When citing an abstract from the 2022 annual meeting, please use the format below.

[Authors]. [Abstract Title]. Program No. XXX.XX. 2022 Neuroscience Meeting Planner. San Diego, CA: Society for Neuroscience, 2022. Online.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.01

Topic: A.09. Adolescent Development

Support: DK124727
DK124727DS
S10RR025671
MD006988
GM060507

Title: Neuropathological Correlates Linking Early-Life Stress to Disordered Eating and Obesity

Authors: *P. ONTIVEROS-ANGEL[1], V. E. WILLIAMS[2], T. B. SIMON[1], F. SHARAFFEDIN[1], J. LOU[3], I. DE LA PEÑA[4], C. VIET[5], J. D. FIGUEROA[1];
[1]Basic Sci. Dept., [2]Ctr. for Hlth. Disparities and Mol. Med., [3]Sch. of Behavioral Hlth., [4]Sch. of Pharmaceut. Sci., [5]Sch. of Dent., Loma Linda Univ., Loma Linda, CA

Abstract: Background: Adverse childhood experiences significantly increase the risk for eating disorders and adult obesity. With extended COVID-19-related lockdowns, self-isolation, and adversities, there is evidence that unhealthy eating habits and obesity are increasing among youth. However, the mechanisms interconnecting early stress to adult obesity remain poorly understood. The present study identifies novel sex-dependent neuroadaptations mediating the elevated risk of overeating and obesity in a rat model of early-life stress. **Methods:** Adolescent Lewis rats (n=96, 48 males, 48 females) were exposed to a two-hit model of predator-based psychosocial stress (PSS) followed by intermittent access to a high-saturated fat obesogenic diet (WD, 41% kcal from fat) or an ingredient-matched control diet (CD, 13% kcal from fat). The PSS paradigm combined unpredictable predator stressors and social instability. We evaluated physiological and behavioral markers of stress longitudinally. The rats were subsequently exposed to intermittent WD feeding patterns to evaluate binge eating-like behaviors. A battery of behaviors, including Phenotyper home cage observation, was performed. The estrus cycle was staged to assess the effects of PSS and WD in estrus cyclicity. Histological imaging and immunodetection methods were used to determine the impact of stress and diet on the immediate early gene cFos and the synaptic integrity marker SV2A. **Results:** We found sex-dependent differences in rats exposed to traumatic stress and consumed the WD. Female rats displayed robust binge eating-like feeding behaviors when compared to males. This phenotype was associated with blunted acoustic startle reactivity. Interestingly, estrus cyclicity evaluation revealed dysregulated length and variability of stage frequency in female rats exposed to traumatic stress. In addition, PSS/WD exposure promoted distinct cFos and SV2A expression patterns in brain regions regulating feeding behavior and stress. **Conclusions:** We demonstrate that early-life stress is a significant catalyst for a high risk of disordered eating, particularly in females with intermittent access to an obesogenic diet. Our model recapitulates fundamental sex-

specific differences in how humans respond to childhood adversities and presents new evidence for potential neuropathological targets. Given the cumulative effect of early-life stress and nutrition on brain maturation and function, these findings significantly impact our understanding of neuroadaptations that may contribute to eating disorders and obesity.

Disclosures: P. Ontiveros-Angel: None. **V.E. Williams:** None. **T.B. Simon:** None. **F. Sharaffedin:** None. **J. Lou:** None. **I. De La Peña:** None. **C. Viet:** None. **J.D. Figueroa:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.02

Topic: A.09. Adolescent Development

Support: Funding (or support) for this project was provided by the Oklahoma State University Rural Renewal Initiative, one of the Tier 1 research initiatives supported by the Office of the Vice President for Research.

Title: Scientific Abstract

Authors: *C. PIERCE[1], C. QUIJADA[2], D. PEACH[3], G. KOEHLER[1], D. VAZQUEZ[1]; [1]Oklahoma State Univ. Ctr. for Hlth. Sci., Tulsa, OK; [2]Col. of Osteo. Med., Tulsa, OK; [3]Dept. of Nutritional Sci. Allied Health, Oklahoma State Univ., Stillwater, OK

Abstract: Can protective and compensatory experiences (PACEs) affect the saliva microbiomein health care providers?*Pierce, Chelsea[1], Quijada Carolina[2], Peach Darci[3], Koehler Gerwald[4] , and Vazquez-SanromanDolores[1].*[1] Department of Anatomy and Cell Biology, Oklahoma State University Center for HealthSciences.[2] College of Osteopathic Medicine, Oklahoma State University Center for Health Sciences.[3]Department of Nutritional Science Allied Health, Oklahoma State University.[4] Department of Biochemistry and Microbiology, Oklahoma State University Center for HealthSciences.Protective and compensatory experiences (PACEs) are positive experiences that increaseresilience and protect against the risk for mental and physical illness. Adults who had increasedPACEs in their childhood have better health and wellbeing even if they had a history of adversechildhood experiences (ACEs). The potential role of the salivary microbiome (SM), one of themost diverse microbiomes in the body, has been linked to systemic diseases with an underlyinginflammatory cause, such as chronic stress like ACES. This study examines the composition anddiversity of the SM of healthcare providers in Harmon County, Oklahoma and explores whetherthere are correlations to the number of PACEs they have experienced. Twenty-four healthcareproviders answered a PACEs and ACEs survey. Saliva samples were collected at two timepoints. The composition of SM was analyzed through 16S rRNA gene-based next generationsequencing. Results: We found out that 29% of our population present an ACEs score equal to ormore than 4 traits, in addition 64% neither experienced an enriched environment such as physicalexercise, nor social interactions outside their household.

From the 19 PACE items score, wefound that 26% experienced equal or less than 7 protective experiences while growing up, and47% percent of those experienced that their parents divorced or separated. The SM will besequenced, and we will expect that the study participants with lower ACE scores, but higherPACE scores will have a different microbial composition in comparison to those with higherACE and lower PACE scores.

Disclosures: C. Pierce: None. **C. Quijada:** None. **D. Peach:** None. **G. Koehler:** None. **D. Vazquez:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.03

Topic: A.09. Adolescent Development

Title: Neural dysfunction associated with cannabis use in teenage drivers: What we know and what we don't

Authors: *P. E. DIAMOND[1], M. NAUMAN[1], M. MENDOZA[1], C. M. SNOW[1], T. BLEDSOE[1], T. M. ROCHE[1], M. L. ZÚÑIGA[2], H. S. BAWEJA[1];
[1]Physical Therapy, [2]Sch. of Social Work, San Diego State Univ., San Diego, CA

Abstract: In this review we address, identify, and discuss proposed potential neural mechanisms that underpin effects of cannabis on the adolescent driver's brain and their fitness-to-drive. Following recent legalization of cannabis via CASB94 and similar bills in other states, there has been an increase in cannabis use amongst driving-age adolescents (16-19 years old). While, both adolescent and adult drivers under the influence of cannabis (DUI-C) are unsafe relative to their unintoxicated peers, sober adolescent drivers are involved in more MVAs when compared with novice adult drivers. Many of the cortical and sub-cortical regions implicated with driving (pre-frontal cortex, thalamus, basal ganglia, cerebellum) do not completely mature until the age of 25. Thus, driving associated executive function, response inhibition, error recognition, visuospatial processing, and potentially motor control are limited in adolescent drivers. These further influence the necessary ability to multitask, process information to make decisions, and execute driving tasks. Importantly, development of these cortical and sub-cortical regions is negatively affected in cannabis users who begin using at an early age. Therefore, we posit that adolescents using cannabis would be at an even greater risk of MVA when compared with their age matched sober counterparts. Currently, little is known about the neurological impairments associated with cannabis use and MVA rates in adolescents. Direct evidence addressing the neurological components that drive the adolescent DUI-C and MVA rates remain obscure due to challenging ethical and legal barriers to objectively study this population. Current evidence uses ineffective outcome measures (e.g., reaction time), as well as indirect self-reported measures of driving and observational data on DUI-C and MVAs. Our work suggests cannabis use impairs adolescent brain development and function, leading to riskier driving behaviors, which in turn leads to

higher rates of MVA in this population. However, a lack of empirical evidence potentially obscures the severity of the problem. It will be important for future studies to quantify fitness-to-drive in adolescents with DUI-C to develop targeted driver safety interventions and inform policy to address this dangerous problem.

Disclosures: P.E. Diamond: None. **M. Nauman:** None. **M. Mendoza:** None. **C.M. Snow:** None. **T. Bledsoe:** None. **T.M. Roche:** None. **M.L. Zúñiga:** None. **H.S. Baweja:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.04

Topic: A.09. Adolescent Development

Title: Effects of methamphetamine exposure during adolescence on development of inhibition in the rat mPFC

Authors: *A. S. BRINKS[1], L. K. CARRICA[2], D. J. TAGLER[2], J. M. GULLEY[2], J. M. JURASKA[2];
[1]Neurosci. Program, Univ. of Ilinois At Urbana-Champaign, Champaign, IL; [2]Psychology, Univ. of Illinois, Champaign, IL

Abstract: Adolescence is a time of heightened vulnerability to the environment, in part due to ongoing development of prefrontal cortical regions. Inhibitory neurotransmission through fast-spiking inhibitory cells (PV) increases across adolescence, contributing to medial prefrontal cortex (mPFC) development (Caballero et al., 2016). Additionally, the number of perineuronal nets (PNNs) increases across adolescence in the rat mPFC, where they preferentially surround PV cells (Baker et al., 2017; Drzewiecki et al., 2020) and support PV cell function. PNNs have been shown to be influenced by drugs of abuse (Slaker et al., 2018; Dannenhoffer et al., 2022). In the mPFC, PNNs also show a temporary decrease at puberty in females but not males (Drzewiecki et al., 2020) which could be a window of enhanced vulnerability for females.To assess the effects of exposure to methamphetamine (METH) during this vulnerable period, male and female Sprague Dawley rats were given IP injections of either 3mg/kg METH or saline at one of three timepoints: early adolescence P30-38, late adolescence P40-48, or adult P60-68. Brains were collected 24 hours after final METH exposure and sections of mPFC were immunofluorescently labeled for PNN's and PV cells. Density and stain intensity of PV cells and PNN's were quantified using ImageJ. Results suggest sex specific effects of METH on stain intensity. Females exposed to METH had more intense PNN's and PV cells at all ages ($p < 0.05$). Ongoing data analysis will assess the effects of METH on colocalization of PV cells and PNNs as well the effects of METH on the number of PNNs and PV neurons by multiplying density by the volume of the mPFC. At present, METH exposure appears to uniquely impact female PNN and PV expression at all timepoints.

Disclosures: **A.S. Brinks:** None. **L.K. Carrica:** None. **D.J. Tagler:** None. **J.M. Gulley:** None. **J.M. Juraska:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.05

Topic: A.09. Adolescent Development

Support: NSERC Grant 206686
Janeway Foundation Trainee Grant

Title: Legacies of stress: Investigating the impacts of repeated trauma on stress transmission across generations

Authors: *L. DAWSON, R. BENNETT, A. JONES, K. RANDELL, K. GRACE, K. IVANY, P. E. MAC CALLUM, A. FLEMING, J. J. BLUNDELL;
Psychology, Mem. Univ. of Newfoundland, St. John's, NL, Canada

Abstract: The underlying mechanisms by which some individuals become more vulnerable to the development and persistence of stress-related and anxiety disorders remains poorly understood. Generational transmission of changes in behaviour and genetic regulation in response to traumatic experience may play a role. This study aimed to assess the effects of chronic predator stress in the parental generational on brain and behavioral changes in the F1 and F2 generations. F0 adult mice were exposed to a predator (rat) or control condition daily for seven days then assessed for anxiety-like behaviour (ALB) using a battery of tests consisting of the elevated plus maze (EPM), open field test (OFT), light-dark box (LDB), and social interaction test (SIT). F0 mice were also monitored for circadian locomotor activity for the 12 days before and after stress exposures. While both control and predator stressed F0 mice showed habituation to the exposures (reduced freezing across days), predator stressed F0 mice froze more than F0 control mice across all seven days. As expected, predator stressed F0 mice showed increased anxiety-like behavior in the EPM compared to F0 controls. Following the behavioral tests, F0 mice were bred (control males to control females, predator stressed males to predator stressed females). We measured anxiety-like behavior (EPM, OFT, LDB, SIT) in adolescent F1 mice. These behaviors, along with novelty suppressed feeding (NSF) and circadian rhythm monitoring, were also assessed following a mild stressor (foot shock) in adult F1 mice to determine if parental experience altered offspring stress sensitivity. Adolescent offspring from predator stressed parents showed decreased anxiety-like behavior as assessed in the EPM, but increased social behavior in the SIT. In adulthood, following a mild foot shock, F1 mice from predator stressed parents show less anxiety-like behavior in the EPM compared to offspring from control parents. Interestingly, in the OFT, female offspring from predator stressed parents showed increased ALB compared to female offspring from control parents. Current studies in the lab are examining changes in stress-related mRNA transcription in the brain in F1 offspring and

assessing anxiety-related behavioral changes in the F2 generation. Our results demonstrate that chronic pre-conception parental exposure to predation risk engenders lasting effects on subsequent generations, a result that may improve our understanding of the etiology of stress-related psychopathologies such as posttraumatic stress disorder as well as anxiety disorders.

Disclosures: **L. Dawson:** None. **R. Bennett:** None. **A. Jones:** None. **K. Randell:** None. **K. Grace:** None. **K. Ivany:** None. **P.E. Mac Callum:** None. **A. Fleming:** None. **J.J. Blundell:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.06

Topic: A.09. Adolescent Development

Support: 2R01MH091068

Title: A Machine Learning Approach to Predict Hyperactive/Impulsive Symptoms Using Irritability Symptoms

Authors: S. KOMIJANI[1], P. MUKHERJEE[2], D. GHOSAL[1], *J. B. SCHWEITZER[3]; [1]Dept. of Computer Sci., Univ. of California, Davis, Davis, CA; [2]Psychiatry and Behavioral Sci., Univ. of California, Davis, Sacramento, CA; [3]Psychiatry and Behavioral Sci., Univ. of California Davis Dept. of Psychiatry and Behavioral Sci., Sacramento, CA

Abstract: There is limited existing research to show the relation between irritability and hyperactive/impulsive symptoms in adolescents with attention-deficit/hyperactivity disorder (ADHD). Impulsivity and ADHD are associated with lower academic and occupational outcomes, higher accident rates, substance use disorders, and suicidality. Identifying the correlations between the symptoms may help us intervene earlier and prevent costly negative outcomes. In this project, we apply machine learning (ML) models to investigate if irritability can predict hyperactive/impulsive symptoms. The model used 108 items scored between 0 (never) to 3 (very often), from the Conners' Parent Rating Scale, including the DSM-oriented ADHD subscales, measuring hyperactive/impulsive and irritability symptoms. We analyzed data for 80 participants (48 males) aged 12-16 years for two consecutive years, T1 and T2. Analysis of raw data showed that higher irritability scores are correlated with a higher hyperactive/impulsive score. We developed a random forest regression model using age, gender, and irritability along with hyperactive/impulsive raw scores at T1, as predictor variables, to predict hyperactive/impulsive scores at T2. Our model shows a reasonable prediction in terms of precision (80%), recall (78%), and balanced accuracy (81%) metrics. Ongoing work includes exploring gender differences in prediction of hyperactivity/impulsivity using Gaussian Graphical Model (network approach) and feature importance (ML approach). Additionally, to address the data imbalance and replicability, we are investigating consolidation of self-report rating scale

along with data resampling techniques to refine the performance and interpretability of the model for prediction of hyperactive/impulsive symptoms in later years.

Disclosures: S. Komijani: None. **P. Mukherjee:** None. **D. Ghosal:** None. **J.B. Schweitzer:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.07

Topic: A.09. Adolescent Development

Support: National Research Foundation of Korea(NRF) grant 2021R1C1C1006503
NRF grant 2021K2A9A1A01102014
NRF grant 2021K1A3A1A21037512
NRF grant 2021M3E5D2A01022515
Seoul National University (SNU) Research Grant No.200-20210083
Creative-Pioneering Researchers Program through SNU No. 200-20210107

Title: Interplay between multigenerational family history of depression and polygenic background for psychopathology in children

Authors: *E. LEE[1], M. T. VAN DIJK[2], E. MURPHY[2], A. TALATI[2], M. M. WEISSMAN[2], Y. Y. JOO[3], J. CHA[1];
[1]Psychology, Seoul Natl. Univ., Seoul, Korea, Republic of; [2]Psychiatry, Columbia Univ., New York, NY; [3]Data Sci., Korea Univ., Seoul, Korea, Republic of

Abstract: All types of mental illness are known to tend to run in families. Family history of depression is reported to increase the overall risks in offspring for the onset of common mental disorders and suicidal behaviors in children. Complementary to familial information, polygenic risk scores have been suggested to assess overall psychiatric disorders risk. Familial risk and inherited genetic factors are significantly involved in developing mental illness, but their relationship remains unclear. In this study, we aimed to investigate the association of family history of depression with polygenic scores (PGSs) of multiple common traits and their effects on childhood psychopathology. We analyzed the phenotype and genotype data of 8,111 multiethnic preadolescents (including 6,151 of European ancestry) from the Adolescent Brain Cognitive Development (ABCD) study. The variable describing risk levels of depression history over two generations was created, assuming that children with a depression history of both parents and grandparents have the highest risk and children with no family history have the lowest. We computed the PGSs of 30 different human complex traits. Based on logistic regression analysis, children with the highest familial risk tended to have higher polygenic chances than those with one ever-depressed generation or no familial risk. Among the tested 30 PGSs, PGSs for depression and bipolar disorder were significantly associated with a family

history of depression. Additionally, our mediation analysis revealed that PGS for depression was a significant mediator between a family history of depression and psychiatric disorders. These results showed the mediating role of PGS in the association of familial depression with psychiatric disorders and suicidality.

Disclosures: E. Lee: None. **M.T. van Dijk:** None. **E. Murphy:** None. **A. Talati:** None. **M.M. Weissman:** None. **Y.Y. Joo:** None. **J. Cha:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.08

Topic: A.09. Adolescent Development

Support: SC1GM139696-01
U54AA019765
U01AA019925-11

Title: Sex-specific effects of adolescent intermittent ethanol exposure on the hippocampal neurogenic niche

Authors: *K. NWACHUKWU[1], K. HEALEY[2], S. SWARTZWELDER[2], A. MARSHALL[1]; [1]North Carolina Central Univ., Durham, NC; [2]Psychiatry & Behavioral Sci., Duke Univ., Durham, NC

Abstract: According to the Centers for Disease Control and Prevention, alcohol is the most commonly abused drug among adolescents, and underage alcohol consumption accounts for more than 3,900 preventable deaths in the United States. Studies have shown that binge-like ethanol exposure during adolescence promotes dysregulation of inflammatory cytokine responses and a reduction of newly regenerated neurons in the dentate gyrus subregion of the hippocampus. These effects include changes in proliferation, regulation, differentiation, and maturation of neurons, and there is indication that such effects may be disproportionate between sexes. This study determined whether sex impacts the neurogenic makers Ki-67 and SOX2 as well as the proinflammatory cytokines TNF-α and IL-1β in adulthood after adolescent intermittent ethanol (AIE) exposure. To determine this, 16 adolescent male and 16 adolescent female rats underwent AIE with 10 doses of ethanol (5 g/kg) over a course of 16 days, on a 2-days on, 1-day off, 2-days on, 2-days off pattern, to mimic the sporadic drinking of adolescents. Half of each sex group received either AIE or adolescent intermittent water (AIW) as the control. In adulthood, animals were sacrificed and immunohistochemical techniques and ELISAs were used to distinguish AIE effects on sex-specific neurogenic markers and proinflammatory markers, respectively. A random number generator was used to assign experimental cohorts with cage numbers to blind experimenters to the treatment groups. Our results indicated that AIE exposure led to a significant decrease in neurogenesis in the dentate gyrus of the hippocampal

formation indicated by reductions in the numbers of Ki-67+ and SOX2+ cells in male and female AIE-exposed rats. Additionally, AIE increased the protein expression of pro-inflammatory cytokines, TNF-α and IL-1β, in the hippocampus of male AIE-exposed rats only. Altogether, our findings indicate that AIE does reduce neurogenesis in the dentate gyrus subregion and pro-inflammatory cytokine expression in the hippocampus. The neurogenic impairment was not sex-specific as both male and female AIE groups had approximately a 10-20% decrease in the number of Ki-67+ and SOX2+ cells compared to their AIW control, though the pro-inflammatory cytokine increase was observed solely in male AIE-exposed rats. A persistent impairment in neurogenesis may alter hippocampally driven behaviors including memory consolidation and retrieval.

Disclosures: K. Nwachukwu: None. **K. Healey:** None. **S. Swartzwelder:** None. **A. Marshall:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.09

Topic: A.09. Adolescent Development

Support: NIH Grant R56AG073965

Title: Multi-omic profiling of the Down syndrome brain reveals novel aspects of aging in DS.

Authors: *C. PALMER[1], C. S. LIU[2], N. WILLIAMS[3], J. CHUN[3];
[1]Univ. of California San Diego, La Jolla, CA; [2]UCSD, La Jolla, CA; [3]Sanford Burnham Prebys Med. Discovery Inst., La Jolla, CA

Abstract: Down syndrome (DS), caused by the triplication of human chromosome 21, is characterized by lifelong cognitive changes and the development of the neuropathological hallmarks of Alzheimer's disease (AD). Previous single-nucleus RNA-sequencing studies identified numerous changes in the DS prefrontal cortex, including changes to neuronal ratios, microglial activation states, and the existence of previously unannotated transcript isoforms containing intra-exonic junctions. The genomic and epigenomic changes that lead to the observed transcriptomic changes, as well as the transcriptome's effects on the cellular landscape are not understood. To elucidate the mechanisms responsible for these changes, we studied the DS postmortem brain via numerous approaches at a single cell resolution, providing insight into the cellular, epigenomic, and genomic factors responsible for the observed transcriptomic changes associated with DS. These results highlight global cellular and epigenomic differences as novel features of the aging DS brain. Supported by NIH R56AG073965 (J.C.)

Disclosures: C. Palmer: None. **C.S. Liu:** None. **N. Williams:** None. **J. Chun:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.10

Topic: A.09. Adolescent Development

Support: NIH Grant G12MD007592
NIH Grant R24DA029989
NIH Grant R25DA033613
NSF Grant HRD-1202008
Mary S. Kostalos and John Kostalos, Jr. Fund

Title: Effects of early life stress on the hippocampal expression of molecules in the BDNF signaling pathway in adolescent female rats

Authors: H. DUNSTER[1], C. AUGUSTINE[1], S. SWEGER[1], A. DIAZ AYALA[1], A. MOORE[1], J. N. HAMDAN[2], R. RODRIGUEZ[2], A. PILLAI[2], A. RASTEGARI[2], K. L. GOSSELINK[3], *J. SIERRA FONSECA[1];
[1]Sci., Chatham Univ., Pittsburgh, PA; [2]Univ. of Texas At El Paso, El Paso, TX; [3]Physiol. and Pathology, Burrell Col. of Osteo. Med., Las Cruces, NM

Abstract: Early-life stress (ELS) is known to induce long-term neurochemical effects that affect neuronal circuit formation. These persistent alterations can result in the development of neurological disorders such as depression and neurodegenerative diseases later in life. The hippocampus (HIPP) can be especially vulnerable to stress, potentially resulting in learning and memory deficits. The brain-derived neurotrophic factor (BDNF) signaling pathway is a primary mediator of neuronal signaling in the HIPP. BDNF binds to and activates tyrosine receptor kinase B (TrkB), initiating an intracellular signaling cascade that leads to the downstream activation of multiple signaling molecules, including the extracellular signal-regulated kinases 1 and 2 (ERK1/2). Phosphorylation of ERK1/2 further signals to transcription factors, leading to changes in gene expression. Evidence shows that proper function of this pathway can be altered by chronic stress. Most studies have been conducted using male subjects; however, female brains are known to have differing levels of BDNF as well as differential stress responses. To address this gap of knowledge, we hypothesized that ELS negatively impacts the expression of protein markers of the BDNF pathway in the female HIPP. Female Wistar rats underwent ELS in the form of neonatal maternal separation for 3h/d on postnatal days 2-14. Hippocampal tissue was harvested from adolescent (45 days old) rats and evaluated for expression of protein markers associated with BDNF signaling, including BDNF intermediate forms (pre-proBDNF and proBDNF), TrkB, and phosphorylated ERK1/2, using immunoblotting analysis. Our results indicate that ELS caused a significant increase in proBDNF expression without affecting levels of pre-proBDNF, thus suggesting that ELS can alter intracellular processing of BDNF. No significant changes were found in the expression of TrkB, pre-proBDNF, and phosphorylated ERK1/2. Collectively, our results indicate that ELS can have persistent molecular effects in the female HIPP, thus highlighting the need to specifically study female responses to chronic stress.

Disclosures: **H. Dunster:** None. **C. Augustine:** None. **S. Sweger:** None. **A. Diaz Ayala:** None. **A. Moore:** None. **J.N. Hamdan:** None. **R. Rodriguez:** None. **A. Pillai:** None. **A. Rastegari:** None. **K.L. Gosselink:** None. **J. Sierra Fonseca:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.11

Topic: A.09. Adolescent Development

Support: UWRF Undergraduate Stipends and Expenses Grant

Title: Adolescent social isolation stress enhances nicotine conditioned place preference and disrupts stress coping behavior.

Authors: D. R. JENSEN[1], A. C. KIRCKOF[2], M. T. DAVIS[1], K. OSTROVIAK[1], E. HOFFMAN[1], *D. G. EHLINGER[1];
[1]Psychological Sci., Univ. of Wisconsin-River Falls, River Falls, WI; [2]Univ. of Kansas, The Univ. of Kansas, Lawrence, KS

Abstract: Adolescence is a sensitive period in brain development that is marked by increased susceptibility to the effects of chronic stress, which may enhance vulnerability to neuropsychiatric conditions such as depression and substance use disorders. In the present study, we used an animal model to examine the effect of adolescent social isolation stress on coping behavior and nicotine reward. During the adolescent period from postnatal day (P)35-P49, male and female C57BL/6J mice were exposed to either social isolation (SI) stress or standard rearing (SR) conditions, as well as nicotine exposure (0.35mg/kg) four times between P35-P49 during a nicotine conditioned place preference (CPP) procedure. On approximately P50, stress-coping behavior was examined following a 6-minute forced-swim test (FST). Our behavioral results show that both male and female SI mice more rapidly develop nicotine CPP compared to SR mice, that SI mice exhibit increased levels of immobility in the FST, and that prior nicotine exposure during social isolation decreases immobility in the FST. These results suggest that adolescent social isolation stress enhances the rewarding effects of nicotine and negatively impacts stress-coping behavior. To determine whether adolescence is a sensitive period for these effects, ongoing research efforts are aimed at comparing these results to adult social isolation and nicotine exposure. Furthermore, we are examining stress-induced functional (c-fos expression) differences in the brains of SI versus SR mice in response to the FST via immunohistochemistry of the dorsal raphe ascending serotonergic system. Collectively, these analyses will help determine neurological correlates of adolescent susceptibility to the negative effects of chronic social isolation stress and inform our understanding of adolescent brain development and vulnerability.
Acknowledgements: Funding for this project was generously provided by University of Wisconsin-River Falls Undergraduate Stipends and Expenses grants (awarded to A.C.K-2020,

D.R.J. & M.T.D.-2022) and Faculty Research Grant (awarded to D.G.E.). We would like to acknowledge undergraduate students Emily Hoffman and Katie Ostroviak for assistance with data collection related to this project.

Disclosures: D.R. Jensen: None. **A.C. Kirckof:** None. **M.T. Davis:** None. **K. Ostroviak:** None. **E. Hoffman:** None. **D.G. Ehlinger:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.12

Topic: A.09. Adolescent Development

Support: 20210517

Title: Combined effect of prenatal maternal immune activation and adolescent exposure to THC on stress vulnerable mice

Authors: *D. BEGMATOVA[1], A. PINHASOV[2], K. MURLANOVA[3];
[1]Ariel Univ. Ctr., Ariel, Israel; [2]Ariel Univ., Ariel, Israel; [3]Physiol. and Biophysics, State Univ. of New York, Buffalo, Buffalo, NY

Abstract: COMBINED EFFECT OF PRENATAL MATERNAL IMMUNE ACTIVATION AND ADOLESCENT EXPOSURE TO THC ON STRESS VULNERABLE MICE
Begmatova D[1], Murlanova K,[1,2] Pinhasov A[1].*[1]Department of Molecular Biology, Ariel University, Ariel, Israel[2]Department of Physiology and Biophysics, Buffalo, NY, USA*Exposure to maternal immune activation (MIA) and cannabis use during adolescence have been associated with increased risk for the development of neuropsychiatric disorders. Inborn stress vulnerability is also known as a strong factor affecting fetal programming. Cannabis exposure in adolescence increases risk for psychosis, yet only a minority of cannabis users develop psychosis, suggesting that cannabis use may interact with pre-existing vulnerabilities such as prenatal MIA-exposure and increase the risk for neuropsychiatric illness.Our resent findings demonstrated that MIA activation in stress vulnerable (Sub) mice increased MK-801 provoked locomotor activity in offspring. In this work we explored the impact of MIA and sub chronic exposure to the main psychoactive component of cannabis, delta-9-tetrahydrocannabinol (THC), in adolescence on mouse models of social dominance (Dom) and submissiveness (Sub), which possess innate features of stress resilience and vulnerability respectively.MIA was elicited through prenatal exposure to polyinosinicpolycytidylic acid (poly(I:C)), supplemented with THC provided throughout adolescence (8 mg/kg/day for 21 days). We found that adolescent THC increased exploratory activity and social behavior and decreased MK-801 provoked locomotor activity Sub mice compared to vehicle treated Sub counterparts (136% of control, $p<0.001$). No effect of adolescent THC on Dom mice was observed. Combination of MIA with adolescent THC significantly increased MK-801 provoked locomotor activity with no effect on exploratory

activity and social behavior in Sub mice compared with respective control. No effect of MIA-THC was observed on respective Dom groups. Activation of CB1 receptors is thought to create an imbalance in excitatory-inhibitory signaling in the brain by influencing the GABAergic, glutamatergic, and dopaminergic systems. These findings suggest that induced MIA in combination with adolescent exposure to THC in individuals with inborn stress vulnerability may have cumulative effect on gross neuroanatomical development and affect GABAergic, glutamatergic, and dopaminergic signaling. that the endocannabinoid system may be sensitive to both prenatal MIA, adolescent THC, or a combination of factors.

Disclosures: D. Begmatova: None. **A. Pinhasov:** None. **K. Murlanova:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.13

Topic: A.09. Adolescent Development

Support: NIH Grant K23 DK106528
NIH Grant R03 DK121025
NIH Grant ULTR001881/DK041301
NIH Grant P50 DK064539
NIH Grant R01 DK048351
NIH Grant P30 DK041301
NIH Grant VA CDA2 IK2CX001717

Title: Interactions of Early Life Adversity and Brain-Gut Alterations Predict Obesity-Related Complications

Authors: *J. LOU[1], A. WILLIAMS[1], T. DONG[2], A. GUPTA[2], J. FIGUEROA[1];
[1]Loma Linda Univ., Loma Linda, CA; [2]UCLA, Los Angeles, CA

Abstract: Interactions of Early Life Adversity and Brain-Gut Alterations Predict Obesity-Related Complications Abstract: Due to continuing high prevalence and co-morbidities, obesity remains a complicated public health problem. Evidence shows that early-life adversity (ELA) impacts the brain-gut system and predisposes them to develop various adult-related disorders and symptoms related to stress sensitivity. Alterations to components of the brain-gut axis mediate between environmental pressures and host neurobiology, leading to obesity-related complications such as cognitive dysfunction and emotional dysregulation. However, it is unclear whether ELA predisposes individuals to obesity-related changes and how neuroendocrine and neurochemical brain-gut alterations mediate the interaction between ELA, BMI changes, and increased reward-based eating and cravings. This study performed a correlation and regression analysis to determine ELA as a predictor of increased BMI and a mediation analysis using structural equation modeling to examine the predictability of ELA on the association between

stress-induced alterations of the gut and brain, specifically in the hippocampus, amygdala, hypothalamus, microbiome diversity, and BMI changes. Data is from a sample of 128 healthy adult participants, mostly of Hispanic origin, with a history of ELA and assessed using validated questionnaires. The adjusted models show interactions between ELA, BMI, hippocampal volume, and inflammatory signatures. We found positive associations between BMI and IL-6 [$r(82)=.23$, $p<.05$] and positive associations between TNFα and left and right hippocampal regions [$r(82) =.25$ & $.22$, respectively, $p<.05$]. Selected gut-regulated factors, inflammatory biomarkers, and neurobiological structures may contribute as mediating factors to increased BMI in individuals exposed to ELA. The findings demonstrate that early adversity during a vulnerable period of neural growth predicts inflammation-driven brain-gut changes that have been implicated in obesity-related vulnerabilities and may inform targeted therapeutic approaches for further investigation.

Disclosures: **J. Lou:** None. **A. Williams:** None. **T. Dong:** None. **A. Gupta:** None. **J. Figueroa:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.14

Topic: G.08. Other Psychiatric Disorders

Support: CCB

Title: Gestational Ethanol Exposure Induces Sex-specific Striatal Acetylcholine and Dopamine Deficits

Authors: *S. BARISELLI, Y. MATEO, N. REUVENI, D. M. LOVINGER;
NIH, Natl. Inst. on Alcohol Abuse & Alcohol NIAAA, Bethesda, MD

Abstract: Fetal alcohol exposure has deleterious consequences on the cognitive abilities and motor skills of patients affected by Fetal Alcohol Spectrum Disorder (FASD) and in pre-clinical models of gestational ethanol exposure (GEE). Deficits in striatal cholinergic interneuron and dopamine function impair action learning and execution, yet the effects of GEE on acetylcholine and dopamine striatal release remain unexplored. Here, we report that alcohol exposure during the first ten postnatal days ($GEE^{P0\text{-}P10}$), which mimics EtOH consumption during the last gestational trimester in humans, induces sex-specific anatomical and motor learning deficits in female mice during adulthood. Consistent with these behavioral impairments, we observed increased evoked-dopamine levels in the dorsolateral striatum (DLS) of $GEE^{P0\text{-}P10}$ female, but not male, mice. Further experiments revealed an impaired β2-containing nicotinic acetylcholine receptor (nAChRs)-modulation of electrically evoked dopamine release, pointing to striatal acetylcholine deficits. Using a genetically encoded acetylcholine sensor ($GACh_{3.0}$), we found a reduced decay of acetylcholine transients in DLS of $GEE^{P0\text{-}P10}$ females in the presence of an

acetylcholinesterase inhibitor. Finally, we showed that this effect is associated with decreased excitability of striatal cholinergic interneurons (CINs), pointing to activity-dependent defects in acetylcholine release. Altogether, these data shed a new light on striatal deficits that might underlie cognitive and motor learning symptoms of patients affected by FASD.

Disclosures: S. Bariselli: None. **Y. Mateo:** None. **N. Reuveni:** None. **D.M. Lovinger:** None.

Poster

519. Vulnerability Risk Factors During Development

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 519.15

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: Grant Nos. 31522028
Grant Nos. 31530031
Grant Nos. 81571056
Grant Nos. CNLZD1503
Grant Nos. CNLZD1703

Title: Positive coping promotes hippocampal-neocortical functional maturation in negative emotion processing

Authors: *T. TIAN[1], B. CHEN[1], J. QIU[2], X. CHEN[2], S. QIN[1];
[1]Beijing Normal Univ., Beijing, China; [2]Southwest Univ., Chongqing, China

Abstract: Human susceptibility to stress varies substantially across individuals. Why do some people fall apart in face of stressful events while others bounce back quickly? Such differential susceptibility to stress is one of the major scientific issues to be answered in recent years. Psychological resilience is a key buffering factor of the vulnerability to stress, which allows for rapid recovery and helps maintain long-term mental health. Adaptive responses to diverse challenges are involved in the slow responses of the neuroendocrinal system, of which stress hormone cortisol is greatly sensitive to stress and dependent on the regulation of the hippocampus. Besides, the hippocampus has been thought to play a vital role not only in the process of positive coping style, but also in vulnerability to stress. However, the role of the hippocampus in processing negative events is still elusive. The hippocampus goes through a rapid development stage in childhood, yet little is known about the neurobiological mechanisms behind positive coping style from a developmental perspective in young children. We investigated the effect of psychological resilience and cortisol awakening response (CAR) on the development of the hippocampus in healthy children aged 6-12. In Study 1, we recruited 89 children and collected their positive coping style (PCS) questionnaire as the index of psychological resilience. The CAR and fMRI data during an emotion processing task (Time1) were obtained. In Study 2, 34 children were invited back in the next year (Time2) to examine the interplay of PCS and CAR on brain development. We found that higher PCS was associated with

greater CAR. Such PCS-related greater CAR paralleled with an increase in hippocampal functional connectivity with the ventrolateral prefrontal cortex (vlPFC) and fusiform gyrus one year later (Time2). Critically, CAR mediated the positive asssociation between PCS and longitudinal changes in hippocampal-fusiform functional connectivity (Time2-Time1), but not in hippocampal-vlPFC connectivity. Moreover, the interplay of PCS and CAR modulated the maturity development of vlPFC through the development of hippocampus-fusiform connectivity. CAR mediates the relationship between PCS and the longitudinal development of hippocampal function. Our findings have important implications in the neurobiological mechanisms underlying how positive coping style actively shapes hippocampal-neocortical functional systems invoveld in negative emotion processing. This points toward that psychological resilience may be dependent on the neuroendocrine process of CAR to shape hippocampal development in childhood.

Disclosures: **T. Tian:** None. **B. Chen:** None. **J. Qiu:** None. **X. Chen:** None. **S. Qin:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.01

Topic: A.09. Adolescent Development

Support: Sackler Award

Title: Role of serotonin brain circuit in the developmental emergence of innate fear

Authors: *G. ZANNI[1], A. MACKAY[2], S. YADOLLAHI KHALES[1], S. MODI[1], G. STEVENS[1], M. CAFFREY CAGLIOSTRO[3], N. PINI[4], C. FERRIS[5], M. S. ANSORGE[1], J. A. GINGRICH[1];
[1]Columbia Univ., New york, NY; [2]RFMH/Columbia Univ., New york, NY; [3]Univ. of Connecticut-Stamford, Stamford, CT; [4]Columbia Uiversity, New york, NY; [5]Northeastern Univ. Program In Behavioral Neurosci., Boston, MA

Abstract: Fear is an emotional state that enables the organism to avoid or reduce harm, ensuring its adaptation and survival. Exacerbated or unfounded fear is a common hallmark of anxiety disorders. Fear responses are orchestrated by the activation of stimulus-specific neural circuits that converge in the periaqueductal gray (PAG). Common treatments for anxiety are serotonin reuptake inhibitors (SSRIs) that act by increasing 5-HT levels. Paradoxically, increased 5-HT during early postnatal development, for example due to early life trauma or SSRIs use in pregnancy, exert profound effects on brain structure and function, increasing adult anxiety and decreasing 5-HT innervation of certain brain structures (mPFC and hippocampus) in rodent models. In humans, imaging and genetic studies have linked serotonin-related genetic polymorphisms to anxiety disorders accompanied by structural and functional changes in PAG among other regions. However, it remains unknown if changes in developmental serotonin

signaling alters adult serotonin circuit PAG function to increase anxiety. Here we investigated this question in mice using chemogenetic or pharmacologic methods to increase 5-HT signaling during postnatal (P) day 2 to 11 and to test reactivity to predator-like odor (2-methyl-2-thiazoline, 2MT) in adulthood. To study unlearned fear-like responses in mice we developed a behavioral assay, in which a predator-like odor is presented in a chamber that allows for fast on- and off-set of the stimulus. I use deep learning algorithms to automate behavioral scoring. We found that chemogenetic or pharmacologic increased 5-HT signaling during this sensitive period increased fear-like responses in the adult. Furthermore, functional MRI imaging in this mouse model revealed robust PAG hyperactivity in response to the predator-like odor. Finally, using projection-specific optogenetics, we found that excitation of 5-HT neurons projecting to the dorsolateral PAG was sufficient to reduce unlearned fear-like responses in normal adult mice. Together our data demonstrate that: 1) developmentally elevated 5-HT signaling produces long-lasting changes in adult 5-HT input into the dlPAG resulting in increased fear, 2) boosting 5-HT release in PAG may be sufficient to rescue these circuit-specific deficits. Overall, these findings suggest that serotonin in the PAG plays a pivotal role in fear and anxiety-like behaviors and understanding how this major modulatory neurotransmitter can modify unlearned fear responses in adulthood, and after developmental interference, can help improve diagnosis, prevention, and treatment strategies for anxiety-related disorders.

Disclosures: G. Zanni: None. **A. Mackay:** None. **S. Yadollahi Khales:** None. **S. Modi:** None. **G. Stevens:** None. **M. Caffrey Cagliostro:** None. **N. Pini:** None. **C. Ferris:** None. **M.S. Ansorge:** None. **J.A. Gingrich:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.02

Title: WITHDRAWN

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.03

Topic: A.09. Adolescent Development

Support: Appalachian State BNR&T Fund

Title: Investigating a space with or without objects evokes activity in superficial entorhinal cortex of adolescent rats

Authors: *H. WOLFE[1], K. M. GEISINGER[2], E. A. GIBSON[2], J. L. REGAN[2], M. C. ZRULL[2]; [1]Biol., [2]Psychol., Appalachian State Univ., Boone, NC

Abstract: The entorhinal cortex (EC) leads, and ends, two neural processing loops through the hippocampal formation (HF). In part, activity across two loops, one beginning with medial EC (MEC) and the other with lateral EC (LEC), contributes to neural representations of an animal's environment as a whole (e.g., MEC led loop) and the location of items within that environment (e.g., LEC led loop). In this study, we used c-FOS to examine neural activation in Layers 2 and 3 of MEC and LEC, which receive input derived from various brain regions and relay input to the HF, evoked by exposure to a novel, multi-level environments with or without a variety of moveable objects. Subjects were 15, adolescent Long-Evans hooded rats. After weaning, the animals were housed in groups in standard shoebox cages and handled daily. At 49 days old and prior to sacrifice, 5 rats spent 1.5 h in an enclosure with ramps and platforms (NoObj group), 5 rats spent 1.5 h in the same enclosure but with a number of objects (YesObj group), and 5 rats spent 1.5.h in a standard shoebox cage in a quiet and dark room (control). Brains were processed to visualize c-FOS+ neurons, and cell counts were made using digital microscopy and stereological technique. All data were standardized to the control group cell counts. Enhaced neural activity was observed across Layers 2 and 3 of MEC and LEC of both NoObj and YesObj groups in comparison to controls (all $p<.01$). For LEC Layer 2, NoObj and YesObj groups did not differ in c-FOS+ neuron counts; however, 147% more activated neurons were observed in LEC Layer 3 of NoObj than YesObj brains ($p<.037$). Both Layer 2 and 3 of MEC in NoObj brains exhibited enhanced evoked activity relative to YesObj brains (+133%, $p<.017$ and +827%, $p<.001$, respectively). These results were somewhat unexpected. MEC layers were expected to be activated similarly across NoObj and YesObj groups due to responsivity to spatial cues of the overall environment (i.e., the global scene). However, more neural activity was elicited in three of four EC layers by exposure to an environment devoid of moveable objects than a setting with various objects (i.e., NoObj vs. YesObj groups). In contrast, and somewhat more expectable, more c-FOS+ neurons were found in MEC than LEC (+46%) of the NoObj group, and within the YesObj group more actvated neurons were in LEC than in MEC (+71%). These latter results align with suggestions that while MEC responds to the global nature of an environment, the LEC responds to both the space of an environment and the location of various objects within that space. That said, our data seem to suggest the EC may be more responsive to the global nature of an environment than local cues within that space.

Disclosures: H. Wolfe: None. **K.M. Geisinger:** None. **E.A. Gibson:** None. **J.L. Regan:** None. **M.C. Zrull:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.04

Topic: A.09. Adolescent Development

Support: Haverford College Startup grant to PRD

Title: Differential changes in prefrontal and hippocampal activity following novel object identity-location recognition in adolescent rats

Authors: N. KASSAHUN[1,2], *P. ROBINSON-DRUMMER[2];
[1]St. Joseph's Univ., Philadelphia, PA; [2]Haverford Col., Haverford, PA

Abstract: Novel object recognition tasks rely on rat's natural tendency to explore novelty. The standard object recognition task (OR) requires rats to learn the identity of an object while the object location (OL) task requires memory of an object's spatial location. The object-in-place (OiP) task requires both object identity and location to be learned. Previous reports in adult animals show that the OR, OL and OiP, tasks depend on the perirhinal cortex, hippocampus and medial prefrontal cortex (mPFC), respectively. Furthermore, the OiP task is suggested to require a working circuit between the hippocampus and the prefrontal and perirhinal cortices. However, little is known about the whether these regions function similarly in the recognition tasks during development. The current study examined learning during the OR, OL and OiP tasks and how hippocampal and prefrontal activity varies across these tasks. Animals were trained in either the OR, OL or OiP task during adolescence [postnatal day (PD) 28] and their brains were subsequently examined for changes in cFos expression relative to control animals that only received habituation. In the hippocampus, preliminary results reveal a trending ($p = .09$) decrease in cFos activity in the ventral DG following learning in the OR and OL tasks (but not the OiP task) but no significant changes observed in the dorsal DG or CA1 subregions. In the mPFC, animals that performed the OiP task showed a significant increase ($p < .05$) in cFos activity relative to controls but no difference was observed in the OR and OL groups. Additionally, although activity was positively correlated between the dorsal blade of the dentate and the ventral blade ($p's < .05$) in all three tasks, a trending ($p = .06$) positive correlation between activity in the mPFC and the DG was only observed in animals that performed the OiP task. These data demonstrate similar regional activity as that observed in adult animals across the three tasks. However, these data are the first to demonstrate a potential functional connection between the hippocampus and prefrontal cortex during OiP recognition learning in adolescence. Additional studies will examine the ontogenetic emergence of this relationship across development including infancy (when only identity recognition is observed) and in juveniles when identity recognition and location memory are not integrated.

Disclosures: N. Kassahun: None. **P. Robinson-Drummer:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.05

Topic: A.09. Adolescent Development

Support: NIH Grant 8 K00 MH124183-02

Title: A neural circuit for age dependent control of reward seeking states

Authors: *G. MANZANO NIEVES, C. LISTON;
Cornell University: Weill Cornell Med. Col., New York, NY

Abstract: As we get older, we learn to modulate our behaviors to evaluate reward outcomes. These adaptive choices are orchestrated by current sensory conditions, internal cognitive states and future expectations. **Understanding how neuronal functions develop and bias our behavior is fundamental to understanding developmental vulnerabilities to psychological disorders.** To understand adolescent reward seeking behaviors, we examine how development changes brain function and connectivity. The medial prefrontal cortex (mPFC) is a key structure for emotional regulation, decision making, and reward seeking behaviors. The reward-modulating properties of mPFC are thought to be derived from inputs from the ventral tegmental area (VTA). However, during adolescence, VTA inputs into mPFC are still developing, with adolescence showing decreased dopaminergic innervation of mPFC. Furthermore, mPFC itself is still developing, as indexed by myelination and inhibitory neuron maturation. Here we expand on our and others anatomical work to specifically probe the **impact of development on functional connectivity and information flow within and between VTA and mPFC**. Using simultaneous multisite neuronal recordings, we found that adolescent mPFC and VTA have increased activity during incorrect trials, compared to adults. This suggests that the compulsive reward seeking behavior we observe in adolescence may result from changes to adolescent interpretation of unrewarded attempts. Furthermore, through recording from VTA inputs into mPFC, we found that the VTA dependent reward information received in mPFC differs in adolescence, with chemical ablation of projections differentially affecting behavior at these ages. Together our work thus far suggests that the adolescent brain is not coding incorrect trials to the same extent as it does in adulthood, and therefore altering adolescent behavior. It may be that the compulsive reward seeking behavior we observe in adolescence may be a direct result of the lack of feedback on incorrect trials. Adolescence do not perceive the trials as wrong, so they do not learn to decrease their reward seeking behavior. Ongoing work is attempting to directly test this hypothesis. Overall, this work extends our understanding of motivation and reward-seeking behaviors across development and generate testable hypotheses that tackle why certain psychiatric disorders, such as impulse control and anxiety disorders, tend to emerge during adolescence.

Disclosures: G. Manzano Nieves: None. **C. Liston:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.06

Topic: A.09. Adolescent Development

Support: Appalachian State BNR&T Fund

Title: Investigating a space with or without objects evokes activity in the amygdala of adolescent rats

Authors: E. A. GIBSON, V. S. TACKER, ***M. C. ZRULL**;
Psychology, Appalachian State Univ., Boone, NC

Abstract: Adolescence can be a stressful time that is often marked by stimulation seeking and emotion-driven decision making, both of which can lead to risk-taking behavior during novel experiences. Exposure to an environment for the first time can promote exploration and emotional response through sensory and motor stimulation, provide opportunity to interact with same-sex conspecifics, and allow investigation of objects and space. Given the relative importance of the amygdala in learned and unlearned emotional response as well as in the evaluation of environmental and social cues, we examined how exposure to a novel, multi-level environment with or without a variety of moveable objects might evoke neural activity across lateral (LA) and basolateral (BLA) amygdala. We used c-FOS to study the differential effects of novelty exposure on evoked neuronal response within the LA and BLA of 15 adolescent rats. After weaning, the animals were housed in groups in standard shoebox cages and handled daily. At 49 days old and prior to sacrifice, 5 rats spent 1.5 h in an enclosure with ramps and platforms (NoObj group), 5 rats spent 1.5 h in the same enclosure but with a number of objects (YesObj group), and 5 rats spent 1.5.h in a standard shoebox cage in a quiet and dark room (control). Brains were processed to visualize c-FOS+ neurons, and cell counts were made using digital microscopy and stereological technique. All data were standardized to the control group cell counts. For LA and BLA, any exposure to novelty (YesObj, NoObj) increased neural activity over no exposure (control), +73% ($p<.001$) and +126% ($p<.001$), respectively. While there was no difference in LA neural activity between YesObj and NoObj groups (+15%, $p=.258$), the BLA of YesObj rats showed +70% more activated neurons (i.e. c-FOS+) than in NoObj rats ($p<.001$). Exposure to an unfamiliar space with or without novel objects promoted activity in LA and BLA. This activity likely underlies behavior related to emotion and assessment evoked in response to being in the environment. Activity in LA may underlie changes in emotional state as well as promote emotional memory formation during exploration and investigation of the space. Neural activity in the BLA may also correlate with behaviors accompanying emotional state but also contribute to adding context. While activity in LA was evoked similarly for environments with or without objects, activity evoked activity in BLA differed between YesObj and NoObj conditions suggesting the presence of objects in the unfamiliar space may have required integration of additional contextual cues to promote emotion-driven exploratory behavior.

Disclosures: E.A. Gibson: None. **V.S. Tacker:** None. **M.C. Zrull:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.07

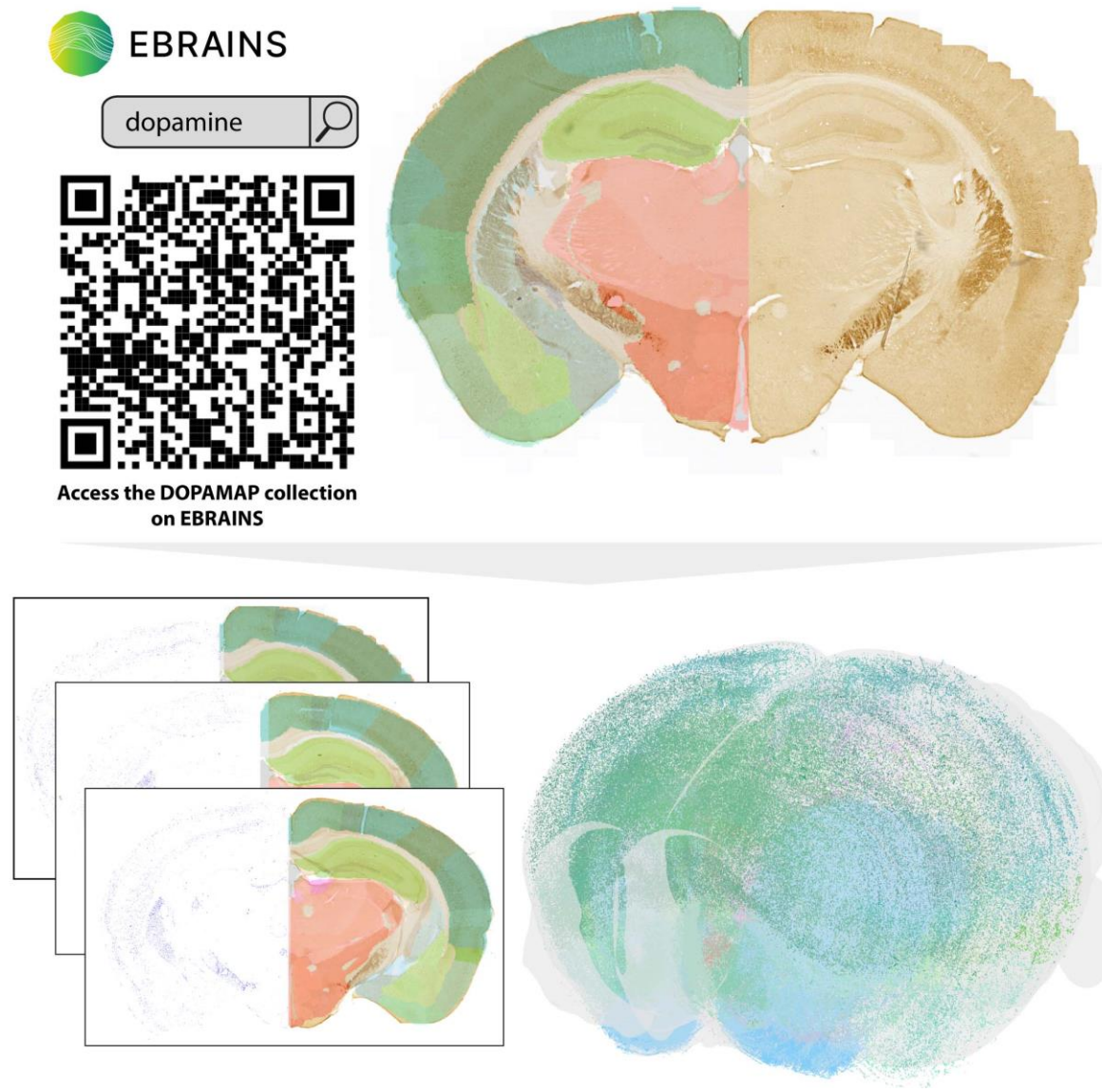
Topic: A.09. Adolescent Development

Support: National Health and Medical Research Council R.D. Wright Career Development Fellowship APP1083309
EU Horizon 2020, Specific Grant Agreement No. 945539 (Human Brain Project SGA3)
HBP voucher program (DOPAMAP voucher)

Title: Quantitative map of dopamine 1- and 2-receptor positive cells in the developing mouse forebrain

Authors: *I. E. BJERKE[1], T. B. LEERGAARD[1], J. G. BJAALIE[1], J. H. KIM[2,3];
[1]Inst. of Basic Med. Sci., Univ. of Oslo, Oslo, Norway; [2]The Florey Inst. of Neurosci. and Mental Hlth., Melbourne, Australia; [3]IMPACT – the Inst. for Mental and Physical Hlth. and Clin. Translation, Deakin Univ., Geelong, Australia

Abstract: The dopaminergic system undergoes major developments during adolescence, a period especially vulnerable to mental disorders. Knowledge about the typical development of this system is required to understand its impact on the ontogeny of different mental disorders, but such information is sparse and scattered across publications investigating one or at most a few brain regions. We have utilized a comprehensive collection of microscopic images of immunostained sections from 152 male and female mice at five stages of development (P17, P25, P35, P49, and adult; DOPAMAP collection available through the EBRAINS Knowledge Graph), showing D1R and D2R expressing neurons across the forebrain. All images are registered to the Allen Mouse brain Common Coordinate Framework tools, with the P17-P35 age groups registered to spatially modified atlas delineations matching the morphology of young brains. We have analyzed all images using the semi-automated QUINT workflow, combining atlas defined regions-of-interest and image segmentation to extract and quantify D1R and D2R positive cells across sex and age groups. The data provide a novel quantitative overview of the regional maturation and spatial distributions of dopaminergic neurons in the mouse forebrain, suitable as a benchmark resource for future experimental studies in mouse models for mental disorders.



Disclosures: **I.E. Bjerke:** None. **T.B. Leergaard:** None. **J.G. Bjaalie:** None. **J.H. Kim:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.08

Topic: A.09. Adolescent Development

Title: Connectivity Differences in the Hippocampus and Amygdala Brain Regions of mice colonized with Infant Gut microbiota

Authors: *A. L. WHITE[1], H. DUBEY, Sr.[2], L. SHENG[3], C. BEST[4], D. PACHECO[5], R. C. KNICKMEYER[6];
[1]Michgan State Univ., East Lansing, MI; [2]Michigan State Univ., The Inst. for Quantitative Hlth. Sci. & Engineering, Michigan State Univ., East Lansing, MI; [3]Michigan State Univ., East Lansgin, MI; [4]3075 Riverwoods Dr., Michigan State Univ., Rockford, MI; [6]Michigan State Univ., [5]Michigan State Univ., East Lansing, MI

Abstract: Background: Rodent and human studies reveal that the gut microbiota can impact brain development and connectivity. For example, our group previously reported that aspects of the human infant gut microbiome are associated with functional connectivity between brain regions involved in processing and responding to threat, assessed via resting state fMRI. The goal of the current study was to test if introduction of human infant microbiomes into germ-free mice produced similar alterations in connectivity. **Method**: We transplanted human infant gut microbiota dominated by Bacteroides (BAC), human infant gut microbiota dominated by Bifidobacterium (BIF), and murine specific pathogen-free gut microbiota (SPF) into pregnant germ-free Swiss Webster mice. At 9 weeks of age, offspring (BAC n=8, BIF n=11, SPF n=8, GF n=8) were anesthetized using isoflurane, then switched to 0.5-1% isoflurane with a subcutaneous injection of dexmeditomidine at 0.05 mg/kg followed by an infusion of dexmedetomidine at 0.1 mg/kg/h. Mice were scanned on a Bruker Biospec 70/30. Anatomical images were acquired using a T2_TurboRARE sequence. Diffusion weighted images were acquired using a 4-segment EPI sequence with 30 directions and B-value 1,000 s/mm2. We acquired resting state fMRI using a T2star_FID_EPI sequence for 2 500 repetition cycles for each mouse. Groups contained similar numbers of males and females but we did not test for sex differences due to limited power. **Results**: ANOVA analysis revealed group differences in functional connectivity between the hippocampus (HIP) and central nucleus of the amygdala (CeA) and between the nucleus accumbens (NAc) and bed nucleus of the stria terminalis (BST). Post-hoc tests (Tukey HSD) showed that BIF mice had significantly greater HIP-CeA connectivity than BAC mice. SPF mice were similar to BIF mice and GF mice were intermediate between BAC and BIF mice for this outcome. For BST-NAc connectivity, SPF mice had significantly greater connectivity than BAC mice. BIF and GF mice were more similar to BAC mice than SPF mice. **Future Directions**: We have created an atlas from our anatomical MRI images and are currently refining a structural segmentation so we can investigate whether regional brain volumes and DTI measures differ between groups.

Disclosures: A.L. White: None. **H. Dubey:** None. **L. Sheng:** None. **C. Best:** None. **D. Pacheco:** None. **R.C. Knickmeyer:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.09

Topic: A.09. Adolescent Development

Support: NIH AA13023

Title: Developmental changes in neuronal excitability in ferret prefrontal cortex

Authors: *D. KEUM, A. E. MEDINA;
Dept. of Pediatrics, Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: Ferrets have a gyerencephalic brain with an organization that is closer to non-human primates than rodents. Because of that, these animals have been used in studies of visual cortex development and plasticity, sensory processing, multisensory integration, TBI and and decision making. Ferrets are altricial animals with eye opening occurring only at postnatal day (P) 32. Developmental events that occur during the second half of gestation in humans, occur after birth in ferrets. Therefore, ferrets have been used as models for neurodevelopmental disorders such as fetal alcohol syndrome and hypoxia-ischemia. Surprisingly, there is no convention of what defines adulthood in the ferret. Studies have considered ferret adulthood as early as P90 and as late as 2 years. Adolescence is a period of development characterized by physical maturation, risk taking behavior and impulsivity. While physical maturation, including puberty, co-occurs with adolescence, they do not indicate when adulthood is reached. For instance, while puberty in humans is completed by 16-17 years of age, the human brain continues to mature until approximately 23-25yrs of age with the prefrontal cortex (PFC) maturing latest. Therefore, the functional maturation of the PFC is proposed to act as a marker of the conversion between adolescence and adulthood. We conducted whole-cell patch clamp of pyramidal neurons in Layers 5 from acute slices from the prelimbic cortex portion of ferret medial PFC at three different ages after puberty (P120, P180 and P220). Injected current-firing curves indicate that the amount of current needed to elicit the same number of action potentials (APs) increase between P120 to P220. APs amplitude and threshold decreased and increased respectively between these ages. Changes in passive membrane properties did not reach statistical significance. Our findings showed that excitability of pyramidal neurons in this region changes between the ages investigated, which strongly suggest that ferret prefrontal cortex does not mature before P220.

Disclosures: D. Keum: None. **A.E. Medina:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.10

Topic: A.09. Adolescent Development

Support: Wellcome Trust Investigator award 108089/Z/15/Z (ACR)

Title: Assessing the common marmoset (Callithrix jacchus) as an animal model of adolescent risk-taking: a cross-sectional study of novel object exploration

Authors: *T. M. LYNN-JONES[1], J. L. ZEREDO[2], A. R. HODGSON[1], M. A. HUGKULSTONE[1], R. A. FROWDE[1], H. F. CLARKE[1], A. C. ROBERTS[1];
[1]Univ. of Cambridge, Cambridge, United Kingdom; [2]Univ. of Brasilia, Brasilia, Brazil

Abstract: Adolescence is a vulnerable period for the onset of mental health problems. It is characterized by widespread changes in both neural and behavioral development, including a peak in exploration, as individuals learn to navigate the world on their own. Many of these exploratory behaviors can be classified as risk-taking, which in humans has been associated with negative mental health outcomes (Smout et al., 2020). The interactions between changes in neural connectivity and patterns of behavior are key to understanding why mental health problems often arise during adolescence, and what preventative measures might be possible. Specifically, changes to threat- and reward-related circuits during adolescence may alter both the prevalence of exploratory behaviors and the likelihood of developing an anxiety disorder (Baker and Galvan, 2020). Neurodevelopmental trajectories in humans are mirrored by those seen in the common marmoset, an increasingly popular primate research model (Sawiak et al., 2018). However, behavioral similarities between the species during adolescence are less well understood. The primary aims of this study are twofold. First, to investigate whether adolescent marmosets, like their human counterparts, are more inclined to take risks than older or younger animals using a Novel Object test (NOt), and in the future to elucidate whether or not juvenile risk-taking correlates with markers of trait anxiety in the same animals assessed at adulthood. The NOt was conducted on thirteen family groups (n = 89 animals), comprised of breeding adult pairs and sets of offspring ranging from infancy to late adolescence. This method captures a wider range of naturalistic behaviors performed by animals as they explore the novel object within their family groups than would be possible if the NOt was performed on individuals temporarily isolated from their families. Novel object interactions were scored as low-risk (approaching and inspecting the object from a distance), medium-risk (approaching within 20cm of the object), or high-risk (touching the object). Adult levels of trait anxiety were assessed using the well-established human intruder test of uncertain threat. Overall, marmosets in late infancy and early adolescence were more likely to touch the novel object, exhibiting greater exploratory risk-taking than either older or younger members of their family groups. These findings support the use of the common marmoset as an animal model for future studies exploring the neurological development underlying changes in risk-taking behavior during adolescence and the potential link between this behavior and trait anxiety in adulthood.

Disclosures: T.M. Lynn-Jones: None. **J.L. Zeredo:** None. **A.R. Hodgson:** None. **M.A. Hugkulstone:** None. **R.A. Frowde:** None. **H.F. Clarke:** None. **A.C. Roberts:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.11

Topic: A.09. Adolescent Development

Title: Pubertal changes on dopaminergic innervation of the striatum and nucleus accumbens

Authors: *K. THOMPSON[1], V. ROUSH[2], V. R. RIESGO[2], J. WILLING[3], H. C. CROMWELL[3];
[1]465 S. Summit, APT 42, [3]Psychology, [2]Bowling Green State Univ., Bowling Green, OH

Abstract: The general goal of this project was to determine if dopaminergic optical density within the dorsal striatum and nucleus accumbens (NAc) changes at the onset of puberty in Long Evans rats. Previous researchers have indicated that there are significant differences in the neuroanatomy between pre-pubertal and adult subjects within the striatum and the NAc, and it is hypothesized that hormones secreted by the "awakening" (onset of puberty) of the Hypothalamic-pituitary-gonadal axis (HPG) may contribute to these developmental changes. Despite this hypothesized connection, few researchers have analyzed the striatum and NAc specifically at the onset of puberty. The current project aims to analyze this gap of pubertal development in the striatum and NAc by using immunohistochemistry and ImageJ software to look at the dopaminergic optical density (OD) of the dorsal striatum and NAc for both male and female Long Evans rats at postnatal (P) days P30 and P40 (pre-pubertal for females and males respectively), 1 day post pubertal onset, 5 days post pubertal onset and P60 (adulthood). The results of our statistical analyses indicated that the NAc has a significantly higher optical density measurement than the dorsal striatum and that the NAc and dorsal striatum were positively correlated. However, the statistical tests indicated that there were no significant differences for sex or age on OD; nor was there a significant difference between interactions of region and sex, region and age, or age and sex. Our data indicates no significant differences between the dorsal striatum and the NAc over the pubertal stage.

Disclosures: K. Thompson: None. **V. Roush:** None. **V.R. Riesgo:** None. **J. Willing:** None. **H.C. Cromwell:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.12

Topic: A.09. Adolescent Development

Title: The effects of age and pubertal onset on development of the ventral tegmental area during adolescence

Authors: *V. RIESGO, T. A. BEEDY, H. M. RUBY, K. THOMPSON, K. A. FLANIGAN, J. WILLING;
Psychology, Bowling Green State Univ., Bowling Green, OH

Abstract: The ventral tegmental area (VTA) contains dopamine-producing cells that are essential for behaviors such as reward processing, memory and cognition, sexual behavior, learning, and mood regulation. Evidence suggests that dopamine cells in the VTA are affected by

estrogen and testosterone, yet potential critical windows of elevated sensitivity to these hormones, such as adolescence, are relatively understudied. Previous work has also shown that dopamine cell number in the VTA changes between the juvenile and young adult period. Despite the known developmental changes and the sensitivity of the VTA to pubertal hormones, the developmental trajectory of this region has yet to be explored during the period of adolescence in relation to the timing of puberty. In the present study, we assess the role of age and puberty in changes in the optical density (OD) of tyrosine hydroxylase (TH) within the VTA in male and female Long Evans rats In the first cohort, VTA tissue was collected on postnatal day (P)30, P40, and P60 brain sections to compare definitively pre- and post-pubertal subjects. To further examine the potential changes in OD immediately following the event of puberty, in a second cohort, we used physical markers to determine the day of pubertal onset in male and female rats. From these subjects, brain tissue was collected from pre-pubertal, 1 day post pubertal onset, 5 days post pubertal onset, and P60 males and females. Lastly, a third cohort was behaviorally tested in a battery of cognitive and affective tasks during adolescence and in young adulthood. We report sex-specific developmental changes in the VTA and in behavior that are mediated by both age and the onset of puberty. The results from these experiments have important implications for our understanding of the connection between dopamine, pubertal hormones, and psychiatric illnesses that manifest during the adolescent period.

Disclosures: V. Riesgo: None. **T.A. Beedy:** None. **H.M. Ruby:** None. **K. Thompson:** None. **K.A. Flanigan:** None. **J. Willing:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.13

Topic: A.09. Adolescent Development

Support: NIMH Grant R21MH127486
Schmitt Program on Integrative Neuroscience (SPIN) GR504304

Title: Immature neuronal change in the primate amygdala: early development and disrupted early environment

Authors: *A. C. MCHALE[1], D. M. DECAMPO[2], J. L. CAMERON[3], J. L. FUDGE[1];
[1]Dept. of Neurosci., Univ. of Rochester Med. Ctr., Rochester, NY; [2]Johns Hopkins Univ. Sch. of Med., Baltimore, MD; [3]Psychiatry, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: The paralaminar nucleus (PL) of the amygdala contains immature neurons that persist throughout life, and may have altered growth depending on early life experiences. Little is known about how PL immature neurons change with age or in response to early-life stress. Here we used stereological techniques to compare cellular features in the PL of 1) infant and adolescent macaques (n=4/group), and 2) infant macaques that experienced early life maternal

deprivation (n=4/group, 2 deprived groups and 1 maternally reared control group). For the normal development study, we found pronounced differences in cellular measurements between maternally-reared infant and adolescent macaques. Adolescent PL had fewer immature neurons, more mature neurons, and larger immature soma volumes when compared to infant PL, providing evidence for PL neuronal maturation between infancy and adolescence. Furthermore, adolescent monkeys had fewer total neurons (immature and mature) in the PL compared to infant, suggesting that neurons may migrate out of the PL by adolescence. Early-life maternal deprivation did not change immature or mature neuron counts by 3 months of age. However, across all infants, immature neuron soma volume was strongly correlated with mature neuron counts. We also previously found that tbr-1 mRNA, which is highly expressed in the PL and associated with glutamatergic neuron maturation, was significantly reduced in the PL in maternally deprived infants in this cohort. Here, we compared tbr-1 mRNA levels with mature neuron counts in the PL and found they correlated positively across all infants. Together, these findings suggest that immature neurons gradually mature by adolescence, and that early maternal deprivation is correlated with subtle slowing in cellular growth, as indicated by decreased tbr-1 mRNA levels and associated correlation with mature neuron counts in infancy.

Disclosures: **A.C. McHale:** None. **D.M. DeCampo:** None. **J.L. Cameron:** None. **J.L. Fudge:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.14

Topic: A.09. Adolescent Development

Support: NIH grant R21MH127486
NIH grant T32NS115705-02

Title: Characterization of microglia during normal development in the primate amygdala subregions

Authors: *D. P. KING[1], A. K. MAJEWSKA[2], J. L. CAMERON[3], J. L. FUDGE[2];
[1]Neurosci., Univ. of Rochester Sch. of Med. and Dent., Rochester, NY; [2]Neurosci., Univ. of Rochester Med. Ctr., Rochester, NY; [3]Psychiatry, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: In primates, the amygdala is comprised of at least thirteen distinct subnuclei. The amygdala is necessary for emotional salience and its dysfunction has been implicated in the onset of many neuropsychiatric disorders. The basal subnucleus (Bpc) has an essential role in processing emotions and is surrounded by a unique group of cells recognized as the paralaminar nucleus (PL). The Fudge lab has previously demonstrated that the PL is different from the other subnuclei because it contains immature post-mitotic neurons which persist into adulthood. However, an increase in the proportion of mature neurons in the PL by adolescence suggests

significant developmental neural growth and differentiation by this age. While we are beginning to understand neural maturation in the PL, the role of microglia, the brain's immune cells, in PL development is unknown. During normal development, microglia can guide the differentiation of precursor cells to neurons, clear excess neuroblasts and prune synaptic contacts. Each of these functions is associated with different morphological and molecular signatures. As a first step in characterizing the role of microglia in the developing PL, we assessed microglia in 3-month-old (infant) and 4-year-old (adolescent) macaques (n=4/group), and used the adjacent Bpc as a relatively more mature brain region for comparison. Four evenly spaced sections through the PL and Bpc were immunoreacted for Iba1, sampled at 40x, and analyzed in FIJI/Image J. The average density of microglia in the PL was 37% greater in adolescents (390 microglia/mm^2) compared to infants (267 microglia/mm^2; p=0.0143), with no differences in density across medial, central, and lateral portions of the PL in either group. Similarly, in the Bpc microglia density was also greater in adolescents (341 microglia/mm^2) compared to infants (290 microglia/mm^2) by 16% (p=0.0143). The spacing index, which measures microglial distribution while accounting for density (calculated as: (average nearest neighbor distance)^2 *microglia density) was similar between the infant and adolescent groups in the PL (p=0.395). In contrast, in adolescent Bpc the spacing index (0.473) was 10% less relative to infants (0.530), indicating closer spacing (p=0.0277). These preliminary data indicate that there is an increase in microglia numbers from infancy to adolescence in the PL and Bpc, with increased clustering of Bpc microglia in adolescence, suggesting potential increased microglia interactions with neurons. Future analyses will determine whether differences microglia densities in each region are associated with increased phagocytosis or pruning.

Disclosures: D.P. King: None. **A.K. Majewska:** None. **J.L. Cameron:** None. **J.L. Fudge:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.15

Topic: A.09. Adolescent Development

Support: R01 MH116675, R01 MH117796

Title: Structural brain changes of the macaque brain during adolescence

Authors: *C. M. GARIN[1], J. ZHU[1], T. R. STANFORD[3], C. T. WHITLOW[4], F. CALABRO[5], B. LUNA[5], C. CONSTANTINIDIS[1,2,6];
[1]Dept. of Biomed. Engin., [2]Neurosci. Program, Vanderbilt Univ., Nashville, TN; [3]Dept. of Neurobio. & Anat., Wake Forest Sch. of Med., Winston–Salem, NC; [4]Dept. of Radiology, Section of Neuroradiology, Wake Forest Sch. of Med., Winston Salem, NC; [5]Dept. of Psychiatry, Univ. of Pittsburgh, Pittsburgh, PA; [6]Dept. of Ophthalmology and Visual Sci., Vanderbilt Univ. Med. Ctr., Nashville, TN

Abstract: Improvement of cognitive abilities, including working memory and response inhibition, that characterize adolescent development is presumably caused by anatomical and functional brain changes over that period. The nonhuman primate model has been particularly fruitful for understanding cerebral function, but developmental data have been lagging. We therefore performed longitudinal morphological MRI to identify cerebral maturation trajectories. A total of 8 macaques (6 males, two females) were scanned over a period of three years over 11 time points spaced approximately 3 months apart (with some variation due to COVID-19 research disruptions). Monkeys' ages ranged from 3.4 to 6.2 years old over that period, covering pre- and post-puberty stages. We also performed morphometric assays to determine objective markers of puberty and adulthood and relied on closure of the epiphysial growth plate of the tibia bones as a common time point to align MRI morphological results rather than relying on absolute age. T1 structural MRI (3T) were obtained under anesthesia. We used AFNI to co-register the images to a study specific template and co-register the macaque CHARM atlases to each individual space. This procedure allowed us to extract biomarkers of cerebral maturation such as volume, thickness and surface. Maturation trajectory (peak, velocity) was estimated using general additive model (*gratia* library). Peak cortical volume (4.66 ± 0.82 years) was observed at an analogous absolute age when compared to humans (5.9 years), which is surprising when considering the 3x faster growth rate of monkeys. In addition, no significant change in cortical volume, suggestive of cortical pruning, was found after the time of the volumetric peak. We also identified the maturation peak times of cortical, subcortical and white matter areas, which occurred in a similar order in macaques and humans. Lobar maturation peak times were also ranked in the following order: parietal, occipital, frontal, temporal. Finer analysis suggested that the temporal pole and the inferior temporal cortex were the main drivers of the protracted temporal maturation. Our results reveal the trajectories of adolescent cortical maturation in a non-human primate model.

Disclosures: C.M. Garin: None. **J. Zhu:** None. **T.R. Stanford:** None. **C.T. Whitlow:** None. **F. Calabro:** None. **B. Luna:** None. **C. Constantinidis:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.16

Topic: A.09. Adolescent Development

Support: R01 MH116675
R01 MH117796

Title: Neuronal activity changes in the macaque prefrontal cortex during adolescence

Authors: *J. ZHU[1], C. M. GARIN[2], Z. WANG[1], A. W. LODISH[3], T. R. STANFORD[3], E. SALINAS[3], C. CONSTANTINIDIS[2];

[1]Neurosci. Program, [2]Biomed. Engin., Vanderbilt Univ., Nashville, TN; [3]Neurobio. & Anat., Wake Forest Sch. of Med., Winston Salem, NC

Abstract: The prefrontal cortex undergoes a protracted period of development in humans and nonhuman primates that is thought to be responsible for the improvement of cognitive abilities, including working memory and response inhibition, in adolescence. Little is known about the association between changes in prefrontal neuronal activity and consequent executive function over this time period. To address this question, we quantified behavioral performance, neurophysiological activity and cortical structure in areas 8a and 46 in adolescent animals. Eight (2F, 6M) macaques (Macaca mulatta) were trained to perform variations of the Oculomotor Delayed Response (ODR) task for assessing working memory, with delay periods of 1.5 s or 3.0 s, including versions that included distracting stimuli. Behavioral performance and neural activity were collected from the animals at time points spaced approximately 3 months apart from 3.4 to 6.2 years old. The monkeys performed the ODR task robustly, with a mean accuracy of 86% correct (excluding aborted trials). Modest improvement in performance in in both ODR variants, with and without distracters was observed with age, with greatest improvement for the most difficult task conditions involving the presentation of distractor stimuli. A total of 994 neurons were recorded in area 8a and 1440 neurons in area 46 across all monkeys and time points. An increase in the percentage of neurons that were responsive to task events (from 35% in the earliest stage to 47% in the latest stage) was observed. Changes in prefrontal activity were also observed over time. These were characterized by increased baseline activity in the fixation period as well as increased evoked activity relative to this baseline during the delay period of the task. Thus, adolescent development in PFC can be characterized by an increase in firing rate during working memory tasks. The results relate the development of cognitive capacity to specific changes in prefrontal neural activity.

Disclosures: J. Zhu: None. **C.M. Garin:** None. **Z. Wang:** None. **A.W. Lodish:** None. **T.R. Stanford:** None. **E. Salinas:** None. **C. Constantinidis:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.17

Topic: A.09. Adolescent Development

Support: HHMI
NIHP50MH112491

Title: Age-related changes in the structure and function of the prefrontal cortex across adolescence in mouse and marmoset

Authors: *K. MASTRO[1], W.-C. LEE[2], E. SCHOENBECK[3], W. WANG[2], B. L. SABATINI[4], B. A. STEVENS[5];

[1]Harvard Med. School/Boston Children's Hosp., Boston, MA; [2]Harvard Med. Sch., Boston, MA; [3]The Stanley Ctr. for Psychiatric Res., The Broad Inst., Cambridge, MA; [4]Neurobio., Harvard Med. Sch. Dept. of Neurobio., Boston, MA; [5]Harvard Med. Sch. Neurobio., Boston Children's Hosp., Boston, MA

Abstract: Over the course of life, there are periods of structural changes that are tied to the development or deterioration of functions. Adolescence, the transition from juvenile to adulthood, is a developmental period marked by a prolonged structural change in prefrontal cortex (PFC) and associated circuits that coincides with an increase in cognitive capacity, sensory seeking, and risk-seeking behaviors. While inroads have been made to understand the mechanisms that drive PFC maturation and how the PFC contributes to an array of cognitive tasks, *the connection between the PFC's developmental changes and its impact on circuit dynamics and behavior remain unclear. Using a cross species approach, mice and marmoset allow for a comprehensive overview of the structural and functional changes that are shared and divergent across rodents and non-human primate. For mice, we used* synaptic physiology, anatomy, behavior, and computational modeling where we uncovered a prolonged period of structural maturation that mirrors change in behavior. Specifically, we examined L2/3 of the prelimbic region of the medial prefrontal cortex in the mouse across the first four months of life. We found changes in the long-range connections, intrinsic properties and the strength of the local microcircuitry. All evidence points to a significant and prolonged maturation of the inhibitory network that may alter the way the animal behaviors. To test whether changes in inhibition has an impact on behavior, we trained animals to perform a simple reversal learning to a more complex 2-armed bandit task. In all cases, younger mice performed differently than the adult mice. To better describe the changes, we have implemented a set of computational models where they suggest that younger animals are guided by new information to a greater extent than older adult animals. For marmoset, we have taken the behavioral and computational modeling and trained a cohort of young and adult marmosets to perform the identical two armed bandit task. Our preliminary results suggest a similiar strategy across the adult cohorts. By running these species in parallel, we will gain a deeper understanding of the shared properties before dissecting the underlying anatomical changes within the nonhuman primate. Together, these data provide a tractable developmental trajectory for which perturbations to genetic and environmental risk factors can be assessed across age, sex and ultimately species. Our next goal is to uncover the in vivo physiologically impact of these structural changes that may explain the changes in cognitive function.

Disclosures: K. Mastro: None. **W. Lee:** None. **E. Schoenbeck:** None. **W. Wang:** None. **B.L. Sabatini:** None. **B.A. Stevens:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.18

Topic: H.07. Long-Term Memory

Support: James S. McDonnell Foundation https://doi.org/10.37717/2020-1208
SSHRC Postdoctoral Fellowship

Title: Neural retrieval of infant memories during childhood

Authors: *D. CHOI[1], J. E. TRACH[1], T. S. YATES[1], C. T. ELLIS[2], N. B. TURK-BROWNE[1,3]; [1]Psychology, [2]Haskins Lab., [3]Wu Tsai Inst., Yale Univ., New Haven, CT

Abstract: As adults age they can retrieve autobiographical memories that trace back several decades, and yet even as young adults could not recall memories from infancy. Such infantile amnesia may reflect a deficit in episodic encoding of infant experiences that results from hippocampal immaturity. However, recent evidence from fMRI in awake human infants suggests that the hippocampus plays a functional role in infant learning, raising the possibility that it may be able to encode memories during this time. An alternative explanation is that infantile amnesia reflects a deficit in episodic retrieval rather than encoding, resulting from greater interference or accelerated forgetting across childhood. Accordingly, it may be possible to find evidence of infant memories in younger but not older children. This hypothesis is difficult to test because the ground truth of what a child experienced as an infant, which is needed to probe memories for these experiences, is typically unknown. We took a novel approach to address this challenge by conducting a case study of two children, aged 3 and 9 years at the time of participation, whose first-person experiences as an infant (6-20 months) were recorded daily on a head-mounted camera. Rather than relying on behavioral assays of episodic memory, which have found impoverished performance in young children, we assessed memory retrieval in the brain using a novel neural measure that does not require explicit reports. During fMRI, each child viewed 30 s video clips from their own infancy and from the infancy of the other child, equating the stimulus set across children. After playing the first 15 s of each video intact, we probed retrieval by distorting the remaining 15 s in one of two ways: First, we tested recall given a partial cue (pattern completion) by blurring the second half, during which memory could allow reinstatement of missing visual information. Second, we tested for detection of mismatches with encoded memories (relational violation) by replacing the soundtrack or scrambling the order of the second half of the video. Other videos were presented intact for the full 30 s, providing a baseline for these additional component processes. We tested retrieval in subregions of the hippocampus and medial temporal lobe that were segmented manually from high-resolution anatomical scans. We found pattern completion in CA1-3, DG, and PHC but only in the younger child. There was no neural detection of relational violations in the younger or older child. This unique case study provides tentative evidence that some infant memories are encoded and retained with sufficient episodic detail to support later cued recall, at least into early childhood.

Disclosures: D. Choi: None. **J.E. Trach:** None. **T.S. Yates:** None. **C.T. Ellis:** None. **N.B. Turk-Browne:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.19

Topic: A.09. Adolescent Development

Support: Canadian Institutes of Health Research (CIHR) Project Grant 2021/4 - 2026/3

Title: Repeated mild traumatic brain injury affects synaptic plasticity in the hippocampus

Authors: *A. GROSS, A. WILLOUGHBY, J. BRAND, E. BOSDACHIN, J. MORRISON, F. RAMNARAIGN, B. CHRISTIE;
Univ. of Victoria, Victoria, BC, Canada

Abstract: Traumatic Brain Injury (TBI) is one of the leading causes of traumatic death and disability globally. Concussion, or mild TBI (mTBI), constitutes up to 75% of all brain injuries that occur annually in the US. There is growing evidence that repeated mild traumatic brain injury can cause chronic neuroinflammation, changes in hippocampal synaptic plasticity, and associated cognitive deficits. In this study, we used an awake closed head injury (ACHI) model to administer an mTBI repeatedly (8 times, spaced 2 hours apart) over the course of the day in juvenile male rats (P23-29). At 1 or 7 days after the injury, hippocampal slices were prepared for *in vitro* electrophysiological recordings, and the capacity for long-term depression (LTD) in both NMDA-mediated receptors and endocannabinoid-mediated receptors, as well as long-term potentiation (LTP) was examined in the dentate gyrus (DG) synapse. We found that r-mTBI did not produce significant alterations in presynaptic neurotransmitter release or in the size of fEPSPs generated with different stimulus intensities. Preliminary results indicate an impaired capacity for LTD in the endocannabinoid-mediated receptors 1-day post-injury, however, 7 days post-injury LTD increases compared to control.

Disclosures: A. Gross: None. **A. Willoughby:** None. **J. Brand:** None. **E. Bosdachin:** None. **J. Morrison:** None. **F. Ramnaraign:** None. **B. Christie:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.20

Topic: A.09. Adolescent Development

Support: CIHR Grant 468837

Title: Prenatal THC exposure alters interneurons and microglia in the hippocampus of adult rats

Authors: *H. REID[1], K. BALL[2], A. K. HINDE[5], O. TREPANIER[5], T. SNOWDEN[3], C. RODRIGUEZ[6], K. R. BREIT[8], J. D. THOMAS[7], B. R. CHRISTIE[4];
[2]Biol., [3]Neurosci., [4]Div. of Med. Sci., [1]Univ. of Victoria, Victoria, BC, Canada; [5]Med., Univ. of British Columbia, Victoria, BC, Canada; [6]Psych, [7]Dept Psychol, San Diego State Univ., San Diego, CA; [8]Psychology, West Chester Univ., West Chester, PA

Abstract: A growing segment of the population has reported cannabis use, including those who are pregnant. Misconceptions around cannabis use are common, and a subset of people may consider it a remedy for morning sickness, despite its effects as a potential teratogen. This research focuses on the effects of prenatal THC exposure in the rat hippocampus. THC acts primarily on CB1 receptors, one of the most ubiquitous G-protein coupled receptors in the brain. They are primarily $G_{i/o}$ coupled, and found presynaptically on interneurons. The work of others has shown that repeated THC exposure causes receptor downregulation. In the case of prenatal exposure, the GABA switch has not yet been flipped, so this causes excitation. This has ramifications for network establishment. Further, CB1 receptors have been shown to be integral for axon guidance, and possibly for the migration of interneurons from the caudal and medial ganglionic eminences to the hippocampus.
Our data suggests that there is an effect of prenatal THC exposure in parvalbumin and somatostatin interneuron densities. Parvalbumin interneurons make up 20% of all interneurons in the hippocampal formation. They are thought to be integral to lateral inhibition and memory consolidation. In 70-day old animals, in the ventral CA1 and DG subfields, the number of PV interneurons decreased, however this effect was smaller when related to the overall cell density. There were fewer changes in interneuron numbers overall in the DG, however a decrease in the number of PV interneurons in the THC exposed group was observed in the ventral DG. Somatostatin interneurons show a regional specific decrease (in the CA1 but not DG subfield). To ensure scientific rigor, power analysis was performed in all data sets. Sex differences were also considered (and were powered appropriately) since adult rats have sexual dimorphism in body size. Thus, profile counts were divided by the area of each cell layer counted. This also serves to increase robustness against brains that had slices that were slightly dorsal leaning (smaller area relative to ventral), to ventral leaning. All researchers were blinded to experimental condition.
High-throughput macro analysis of ~19,000 Iba1+ cell circularity was investigated in adult animals to assess if there are probable long term inflammatory ramifications of prenatal THC exposure. Iba1+ cells circularity was found to be decreased in the dorsal dentate gyrus and the ventral CA1 in both sexes. Sex differences were observed in the dorsal CA1, where only females showed decreased circularity.

Disclosures: **H. Reid:** None. **K. Ball:** None. **A.K. Hinde:** None. **O. Trepanier:** None. **T. Snowden:** None. **C. Rodriguez:** None. **K.R. Breit:** None. **J.D. Thomas:** None. **B.R. Christie:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.21

Topic: A.09. Adolescent Development

Support: NIH Grant AA012446

Title: The effects of postnatal choline supplementation and exercise intervention in ethanol-exposed rats

Authors: *F. O. RAMNARAIGN[1], P. DONAGHY[1], C. SAHASRABBUDHE[1], A. GROSS[1], J. D. THOMAS[2], B. R. CHRISTIE[3];
[1]The Univ. of Victoria, Victoria, BC, Canada; [2]Dept Psychol, San Diego State Univ., San Diego, CA; [3]Div. of Med. Sci., Univ. of Victoria, Victoria, BC, Canada

Abstract: Fetal Alcohol Spectrum Disorder (FASD) is among the leading causes of neurodevelopmental damage persisting into and throughout adulthood, yet a universal treatment for FASD does not currently exist. There is evidence that the nutrient, choline, and exercise interventions can reduce impaired phenotypic outcomes in FASD subjects - particularly in memory-related tasks. FASD subjects exhibit decreased neurogenesis in the hippocampal dentate gyrus (DG), a region of the brain required for information consolidation, while choline-treated and exercise-treated subjects both individually increase neurogenesis in the DG. This study explores whether choline and exercise produce additive recovery effects to neurogenesis in the DG following postnatal ethanol exposure. A third-trimester binge-like ethanol exposure model of a gavage-fed liquid diet was administered to eight randomly assigned groups of neonate rats from PD 4-9 (blood alcohol concentration ~300-340 mg/dl). Groups were given subcutaneous injections of either choline (100 mg/kg/day) or saline from PD 10-30 and were placed into a daily forced exercise task on either a moving (10 m/min) or non-moving automated running wheel from PD 26-35. All groups received intraperitoneal Bromodeoxyuridine (BrdU) (400 mg/kg) injections on PD 35 and were euthanized within 24 hours of BrdU treatment (on PD36). Immunofluorescent staining against BrdU, ionized calcium-binding adapter molecule (Iba1) and glial fibrillary acidic protein (GFAP) was used to visualize changes in the distribution and abundance of dividing cells and microglial and astrocytic subpopulations across conditions. Preliminary results suggest that ethanol-exposed rats displayed a decrease in the abundance BrdU+ cells compared to controls. This effect was reduced in choline-treated and exercise-treated rats. Choline-exercise-treated rats displayed the highest abundance of BrdU+ cells, in both ethanol-exposed and control conditions. The abundance of BrdU+ cells was significantly higher than the abundance of co-positive BrdU and IBA1 cells, as well as co-positive BrdU and GFAP cells. These results suggest that the administration of choline and exercise to ethanol-exposed and wild-type rats may increase the DG's capacity for neurogenesis relative to either intervention alone, indicating a potential additive interaction between the cognitive changes elicited by both interventions, occurring as early as PD36 (adolescence). Choline and exercise are both promising treatment options for the recovery of impaired neurogenesis in the DG, and the use of both interventions together could increase this capacity for recovery in FASD subjects.

Disclosures: F.O. Ramnaraign: None. **P. Donaghy:** None. **C. Sahasrabbudhe:** None. **A. Gross:** None. **J.D. Thomas:** None. **B.R. Christie:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.22

Topic: A.09. Adolescent Development

Support: NIGMS Grant SC3GM130467

Title: Social defeat stress induces a depression-related phenotype in male prairie voles (Microtus Ochgaster)

Authors: *M. RODRIGUEZ[1], S. A. CASTILLO[2], B. CUSHING[2], S. D. INIGUEZ[1];
[1]Dept. of Psychology, [2]Dept. of Biol. Sci., The Univ. of Texas at El Paso, El Paso, TX

Abstract: Major depressive disorder (MDD) is a mental illness that affects millions of people worldwide, making it one of the leading causes of disability. Chronic stress exposure is a well-recognized risk factor for the development of MDD, in which social stress, in particular, plays a role in the etiology of mood-related disorders. While the traditional social defeat stress (SDS) paradigm has been useful in investigating the development/expression of behavioral responses associated with mood-related illnesses, mouse and rat models, unfortunately, display limited social structures that more directly resemble human behavior; consequently, limiting their translational implications. For this reason, the purpose of this study was to examine if SDS, in male prairie voles specifically, results in a depression-related phenotype. To do this, sexually naïve male voles experienced physical defeat bouts by a pair-bonded male resident aggressor, 5 minutes per day, for 7 consecutive days. Non-stressed control male voles (same-sex siblings) were handled daily and housed in cages fitted with perforated Plexiglass partitions. Twenty-four hours after the last SDS exposure, experimental voles were evaluated on the social interaction, sucrose preference, and forced swim tests - behavioral endpoints that are commonly implemented to evaluate sociability, anhedonia, and despair-like behavior. When compared to controls, SDS-exposed voles displayed decreases in body weight, sociability, and preference for a sucrose solution, along with increased immobility on the forced swim test. Importantly, no differences in general locomotor activity were observed as a function of SDS between the groups. Collectively, these findings indicate that SDS exposure induces depression-related behavior in male prairie voles.

Disclosures: M. Rodriguez: None. **S.A. Castillo:** None. **B. Cushing:** None. **S.D. Iniguez:** None.

Poster

520. Developmental Regulation of Brain and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 520.23

Topic: A.09. Adolescent Development

Support: SIP-Instituto Politécnico Nacional
Instituto Nacional de Psiquiatría Ramón de la Fuente Muñiz

Title: Preeclampsia as a possible origin for development of anxiety, depression disorders and memory alterations in offspring

Authors: B. R. VÉLEZ-GODÍNEZ, P. LÓPEZ-SÁNCHEZ, *N. PAEZ-MARTINEZ;
Inst. Politecnico Nacional, Mexico City, Mexico

Abstract: It has been described that preeclampsia might affect health in offspring at adult life. Low birth weight has been associated to cardiovascular and metabolic alterations in adults. There are, however, few studies relating this illness to development of anxiety, depression and memory alterations in offspring. The objective of this work was to evaluate the possible relationship of preeclampsia with development of anxiety, depression, and memory alterations in offspring from preeclamptic rats. Thirty Swiss Webster females 12-14 weeks old, and 15 males 14 weeks old were mate in a two females per male ratio. Day 1 of pregnancy was recorded when spermatozoa were found in a vaginal smear. Pregnant females were divided into control group receiving vehicle, and preeclampsia group receiving L-NAME in drinking water at a dose of 60 mg/Kg from day 10 of pregnancy until delivery. Offspring was weaned and sexed at 4 weeks after birth. For study purposes, 2 females and 2 males were taken from every control and preeclamptic dam to set 4 study groups with 15 males and 15 females. Each group was evaluated using the elevated plus maze test (anxiety), tail suspension test (depressive-like behavior) and the recognition of novel objects test (memory), in addition to the open field test was performance to corroborate their motor activity and validate our results. We found that the male offspring from preeclampsia showed an enhancement in the time that mice spend in the close arms in the elevated plus maze test, in addition to a longer immobility time in the tail suspension test, compared to the offspring from healthy pregnancies. On the other hand, female offspring from preeclampsia showed a lower percentage of recognition in the memory test compared to offspring from normal pregnancy. These results seem to suggest that preeclampsia predisposes young male offspring to develop anxiogenic and depressive-like behavior as well as memory impair in young female offspring.

Disclosures: B.R. Vélez-Godínez: None. **P. López-Sánchez:** None. **N. Paez-Martinez:** None.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.01

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NIH Grant NS113530 (CRC)
NIH Grant MH127404 (HY)
NIH Grant HD082373 (HY)
NIH Grant NS111619 (SFT)

Title: Glun2c/d selective positive allosteric modulator of nmda receptors reveals particular gabaergic interneuron subtypes that do not rely on glun2d-mediated neurotransmission

Authors: *C. R. CAMP[1], H. XING[1], T. BANKE[1], M. EPPLIN[2], R. FRITZEMEIER[2], S. KIM[1], J. ZHANG[1], H. YUAN[1], D. LIOTTA[2], S. F. TRAYNELIS[1];
[1]Pharmacol. and Chem. Biol., [2]Chem., Emory Univ., Atlanta, GA

Abstract: GABAergic interneurons make up a small proportion of total CNS cells, yet they control circuit excitability and behavior. Through opto- and chemogenetic manipulation, hippocampal GABAergic interneuron function can be directly linked to spatial navigation, memory, and overall network oscillations. Additionally, interneuron dysfunction is a central hypothesis to multiple neuropathological diseases including epilepsy, schizophrenia, and autism. Thus, the therapeutic potential of interneuron modulation is immense. One potential avenue for interneuron-specific control is through activation of GluN2D-containing NMDA receptors. The GluN2D subunit is expressed in GABAergic interneurons, with little to no expression on glutamatergic principal cells. In line with these data, our novel GluN2C/D-specific positive allosteric modulator, (+)EU1180-453, was able to significantly increase the charge transfer of evoked NMDA receptor-mediated currents onto *stratum radiatum* interneurons (0.59 pC for baseline vs 0.92 pC for 10 µM (+)EU1180-453; paired t-test, p=0.003; n=13). Interestingly, only 54% (7/13) of these recorded cells showed robust potentiation above 1.25-fold, while the remaining cells had minimal response to (+)EU1180-453. Given heterogeneity of interneurons found within *stratum radiatum*, we hypothesized that the differential effects of (+)EU1180-453 may be driven by lack of GluN2D expression on a particular interneuron subtype. To answer this, we used genetically-driven fluorescent mice to test (+)EU1180-453's efficacy on cholecystokinin (CCK) only, CCK/vasointestial peptide (VIP), VIP only, and neuropeptide-Y (NPY) positive cells in *stratum radiatum*. We have shown mixed effects of (+)EU1180-453 on CCK only and CCK/VIP cells, while VIP only cells showed no potentiation, and NPY cells showed strong potentiation. Since GluN2C is not expressed on these interneuron subtypes, these data suggest that functional synaptic GluN2D expression may be limited to a unique subset of interneurons. We are currently assessing parvalbumin- and somatostatin-expressing interneurons to obtain a broad picture of GluN2D synaptic expression and the interneuron landscape. These data demonstrate how positive allosteric modulators of GluN2D-containing interneurons could alter network function.

Disclosures: **C.R. Camp:** None. **H. Xing:** None. **T. Banke:** None. **M. Epplin:** None. **R. Fritzemeier:** None. **S. Kim:** None. **J. Zhang:** None. **H. Yuan:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Sage Therapeutics. **D. Liotta:** F. Consulting Fees (e.g., advisory boards); NeurOp. **S.F. Traynelis:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Janssen. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); NeurOp. F. Consulting Fees (e.g., advisory boards); CureGRIN, Sage Therapeutics, Eumentis Therapeutics, GRIN2B Foundation, CombinedBrain.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.02

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NS113530
NS111619
MH127404
HD082373

Title: A positive allosteric modulator of GluN2D-expressing N-methyl-D-aspartate receptor regulates CA1 pyramidal cell excitability via facilitating synaptic input from GABAergic interneuron

Authors: *H. XING[1], C. R. CAMP[1], T. BANKE[2], M. P. EPPLIN[2], R. FRITZEMEIER[2], S. KIM[2], J. ZHANG[2], D. LIOTTA[2], H. YUAN[3], S. F. TRAYNELIS[4];
[1]Emory Univ., [2]Emory Univ., Atlanta, GA; [3]Emory Univ. Sch. Med., Emory Univ. Sch. Med., Atlanta, GA; [4]Emory Univ. Sch. of Med., Sch. of Med., Atlanta, GA

Abstract: NMDA receptors (NMDARs) mediate a slow, Ca^{2+} permeable component of excitatory synaptic transmission in the CNS, and are tetrameric assemblies of two glycine binding GluN1 subunits and two glutamate-binding GluN2 subunits. There are four different GluN2 subunits (A-D), which are differentially expressed throughout the CNS. The GluN2D subunit appears absent in most principal cells but is present in many interneurons. We have developed a series of positive allosteric modulators that show selectivity for the GluN2C/2D subunits. Here we explore the actions of these modulators on hippocampal function using current clamp recording from CA1 pyramidal cells and interneurons in hippocampal slices from C57BL/6J mice (P20-25). Horizontal brain slices that contained the CA1 region were prepared and spontaneous firing of CA1 pyramidal cells and interneurons was recorded for a 5-minute baseline period, followed by a 10-minute of (+)-1180-453 (10 μM), which is a selective GluN2C/2D positive modulator (EC_{50} 3 μM). The recording was concluded by co-applying (+)-1180-453 with DL-AP5 (400 μM) for 5 min. The intrinsic membrane properties were measured during each treatment, including current-voltage (IV) curve, input resistance, the membrane time constant (τ), rheobase, evoked-spike firing frequency, and resting membrane potential. (+)-1180-453 hyperpolarized the membrane potential from -68±3.4 mV to -72±3.3 mV (mean± SEM; n=10), shifted the IV curve more negative, but did not significantly alter input resistance, spike firing or rheobase. These effects were blocked by APV. To investigate the mechanism of the hyperpolarizing effect of (+)-1180-453, we repeated this protocol in pyramidal cells (n=8) with inclusion of the $GABA_A$ receptor inhibitor bicuculline (20 μM). The administration of bicuculline did not significantly change the intrinsic membrane properties of pyramidal cells, but eliminated the effects of (+)-1180-453. Recordings from CA1 interneurons showed two different responses. In 5 of 10 interneurons, (+)-1180-453 increased spike firing frequency, whereas it hyperpolarized the remaining 5 interneurons. These results suggest that potentiation of NMDAR responses in GluN2D-expressing GABAergic interneurons can increase their activity and

suppress the excitability of CA1 pyramidal cells. We interpret the hyperpolarizing actions on interneurons of (+)-1180-453 as indicative of interneurons that do not express GluN2D but receive input from other GluN2D-expressing interneurons. These results show that enhancement of GluN2D can sculpt interneuron function in a complex fashion that alter circuit properties.

Disclosures: **H. Xing:** None. **C.R. Camp:** None. **T. Banke:** None. **M.P. Epplin:** None. **R. Fritzemeier:** None. **S. Kim:** None. **J. Zhang:** None. **D. Liotta:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; member of the Board of Directors for NeurOp Inc., co-inventors on Emory-owned Intellectual Property that includes positive allosteric modulators of NMDA receptor function. **H. Yuan:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; PI on a research grant from Sage Therapeutics to Emory, co-inventors on Emory-owned Intellectual Property that includes positive allosteric modulators of NMDA receptor function. **S.F. Traynelis:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; PI on a research grant from Janssen to Emory, member of the SAB for Sage Therapeutics, Eumentis Therapeutics, the GRIN2B Foundation, the CureGRIN Foundation, and CombinedBrain, member of the Board of Directors for NeurOp Inc., co-inventors on Emory-owned Intellectual Property that includes positive allosteric modulators of NMDA receptor function.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.03

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: GAUK 376221
ProgramLT—INTER-EXCELLENCE, LTAUSA19122

Title: Functional analysis of genetic variations in the cytosolic domain of NMDA receptors

Authors: ***V. KUCHTIAK**[1], E. TOMOVIC[1], L. VYKLICKY[1], S. A. SWANGER[2], A. BALIK[1]; [1]Dept. of Cell. Neurophysiol., Inst. of Physiol. CAS, Prague, Czech Republic; [2]Fralin Biomed. Res. Inst., Roanoke, VA

Abstract: The expression and activity of ionotropic glutamate receptors controls signal transduction at the excitatory synapses in the CNS. The major role is played by the calcium-permeable NMDA receptors (NMDARs) that are represented by three types of subunits: GluN1, GluN2A-D, and GluN3A-B. The NMDAR is composed of two obligatory GluN1 subunits and

two GluN2 or GluN3 subunits in different combinations. Each subunit consists of four domains: the extracellular amino-terminal and agonist-binding domains; the transmembrane domain; and the intracellular C-terminal domain (CTD). The CTD representing up to half of the entire NMDAR subunit (GluN2A/B) has the lowest homology between NMDAR subunits. The CTD of the NMDAR is crucial for trafficking of the receptor to synapses, endocytosis, and subsequent degradation of the receptor. The direct effect of altered CTD on the functional properties of the NMDAR ion channel has also been shown. CTD interacts with several intracellular proteins, such as cytoskeletal proteins, scaffold proteins, or proteins involved in intracellular signaling. In addition to the binding of various proteins, CTD is a target of numerous post-translational modifications regulating the functional properties of the receptor. Amino acid mutations in the cytosolic part of NMDAR subunits have been identified in individuals with various neurological impairments. These mutations could potentially contribute to the emergence and development of the disorder. We employed electrophysiological and microscopy techniques to study the impact of four (P1386L, N1076K, T1064A, V967L) disease-diseases associated mutations (schizophrenia or epilepsy) in the CTD of the GluN2A subunit, which we identified in our NGS data set from patients. We analyzed the NMDAR ion channel properties, their changes in surface expression, and their trafficking to synapses. Our results suggest that the mutations we tested affect the ion channel properties, such as glutamate potentiation, desensitization, and open channel probability. Besides that, studied mutations significantly decreased NMDAR surface expression. Furthermore, the *de novo* mutation P1386L found in schizophrenia patients shows a defect in synaptic localization. We demonstrated that decreased synaptic localization of P1386L mutated NMDAR is presumably linked to lower affinity for the scaffold protein PSD-95. In summary, we showed that genetic changes in the CTD of the NMDA receptor altered its functions, impairing its delivery to the cell surface and synaptic localization, which might contribute to the emergence of neurological disorders.

Disclosures: **V. Kuchtiak:** None. **E. Tomovic:** None. **L. Vyklicky:** None. **S.A. Swanger:** None. **A. Balik:** None.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.04

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: ProgramLT—INTER-EXCELLENCE, LTAUSA19122
Technology Agency of the Czech Republic: TN01000013
BIOCEV project (CZ.1.05/1.1.00/02.0109)

Title: The effects of genetic variations and methylation profiles of GRIN gene promoters in schizophrenia

Authors: *A. BALIK[1], V. KUCHTIAK[1], E. TOMOVIć[2], K. HIRSCHFELDOVA[1], J. HORACEK[3], L. VYKLICKY[1];
[1]Inst. of Physiology, CAS, Prague, Czech Republic; [2]Cell. neurophysiology, Inst. of Physiol. CAS, Prague, Czech Republic; [3]NIMH, Klecany, Czech Republic

Abstract: Schizophrenia (SCH) is a severe mental disorder characterised by a high heritability rate. Genome-wide association studies revealed several individual loci contributing to disease susceptibility included GRIN genes encoding NMDA receptor subunits. A substantial number of NMDAR mutations and rare variants have been reported, but much less is known regarding the common population variability of the genes. In our recent study, we demonstrated that genetic variability in ionotropic glutamate receptors (iGluR) plays a meaningful role in the majority of SCH patients. We found genetic variability associated with SCH for variants in intronic sequences and regulatory regions of GRIN genes, suggesting possible alterations in gene expression and mRNA processing leading to NMDAR signaling disfunction. In addition, previous studies also reported different levels of gDNA methylation in various neurological diseases, also suggesting altered epigenetic regulation. Our goal was to analyse both the genetic variations and the methylation profiles of the regulatory parts of the GRIN genes to assess their effects on gene control. Our custom neuropanel covers, among other loci, all genes for the iGluR subunits including their promoter, 5'UTR and 3'UTR regions. The clinical cohort comprises 62 SCH and corresponding control subjects. We mapped the genetic profiles of promoters and UTRs in individual GRIN genes and patients and used the CADD tool for scoring the deleteriousness of variants. The highest number of SNPs was observed in GRIN3B subunit, 27 in total. We also identified 5 SNPs in GRIN1 promoter region, 10 in GRIN2A, 13 in GRIN2B, and 8 in GRIN3A. Two variants rs181870048 (GRIN2A) and rs12338602 (2B) had a high CADD_phred score. The selected promoters (combination of SNPs) identified with higher frequency in SCH and the control were cloned into a luciferase reporter plasmid and their activity was analysed. We observed differential activity between the selected promoters of each GRIN gene. However, the expected decreased activity of the promoters was not exclusive to the promoters found more frequently in SCH. In parallel, we analysed the methylation sites overlapping with the promoter regions using real-time PCR and bisulfite sequencing. We found an increased level of DNA methylation in GRIN1 and GRIN3A/B genes in SCH. Methylation was decreased in the analysed region of GRIN2A, whereas the GRIN2B gene was not affected. Our results suggest that both genetic changes in the promoter region of Grin genes and changes in methylation can alter the expression of GRIN genes in SCH. The data presented also help us to understand the possible role of several SNPs in NMDAR regulation.

Disclosures: A. Balik: None. **V. Kuchtiak:** None. **E. Tomović:** None. **K. Hirschfeldova:** None. **J. Horacek:** None. **L. Vyklicky:** None.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.05

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: CIHR FDN #154276

Title: NMDA receptor mediated deficits in a mouse model of the disease-associated G620R missense variant in the GluN1 subunit

Authors: *J. LEE[1,2,3], P. TIDBALL[1,2], Y. LEE[1], J. THACKER[1], F. JIN[1], J. GEORGIOU[1], G. COLLINGRIDGE[1,2,3];
[1]Lunenfeld-Tanenbaum Res. Inst., Sinai Hlth., Toronto, ON, Canada; [2]TANZ Ctr. for Res. in Neurodegenerative Dis., [3]Dept. of Physiol., Univ. of Toronto, Toronto, ON, Canada

Abstract: N-methyl-D-aspartate receptors (NMDARs) are synaptic glutamate receptors that mediate a Ca^{2+}-permeable component of excitatory transmission and are critical for synaptic plasticity and brain development. Recently, whole exome sequencing has identified multiple patients with heterozygous *de novo* missense mutation in the M2 transmembrane domain of the NMDAR GluN1 subunit (encoded by the *GRIN1* gene). All patients present with severe intellectual disability, developmental delay, motor and sensory deficits. The genetic variant (c.1858G>C/A) results in a substitution from glycine to arginine at position 620 of GluN1 (p.G620R). However, the impact of this variant on synaptic function is unknown. We therefore generated a mouse model (*Grin1*-G620R) mimicking the heterozygous variant reported in the patients. Both male and female mice were used in this study. *Grin1*-G620R mice were smaller and weighed less than their wild-type (WT) littermates across a range of ages. However, we found no differences in gross brain anatomy or neuron number in Nissl-stained sections from adult mice. In behavioural testing, *Grin1*-G620R mice (8-12 weeks) exhibited locomotor deficits in the open field test, as well as increased startle response during pre-pulse inhibition. We used acute slice electrophysiology to examine the impact of the G620R variant on synaptic function in the CA1 hippocampus. Extracellular field recordings revealed no deficit in basal AMPAR-mediated synaptic transmission of *Grin1*-G620R mice; however, NMDAR-mediated field EPSPs were reduced to ~50% of WT levels, and long-term potentiation (LTP) induced by theta-burst stimulation was similarly reduced by ~50%. In voltage clamp recordings from CA1 pyramidal neurons, NMDAR-EPSCs from *Grin1*-G620R mice were not only reduced compared to WT, but also exhibited significantly faster decay kinetics. In contrast, analysis of NMDAR-EPSC current-voltage plots showed no difference in the sensitivity of NMDARs to blockade by Mg^{2+} ions. Finally, we found no difference in surface GluN1 protein level in *Grin1*-G620R mice using a surface biotinylation approach. Together, our data suggest that variant GluN1-G620R subunits are incorporated into synaptic NMDARs where they result in reduced channel function and impair activity-dependent synaptic plasticity, but do not significantly impact circuit formation during brain development.

Disclosures: J. Lee: None. **P. Tidball:** None. **Y. Lee:** None. **J. Thacker:** None. **F. Jin:** None. **J. Georgiou:** None. **G. Collingridge:** None.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.06

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NINDS R01 NS088479 (LPW)
SBU-BNL SEED Grant (LPW)
Stony Brook University GSO Distinguished Travel Award

Title: Activation dynamics of NMDA receptors

Authors: *M. HE, L. P. WOLLMUTH;
Stony Brook Univ., Stony Brook Univ., Stony Brook, NY

Abstract: Glutamate is the primary excitatory neurotransmitter in the central nervous system. The dynamics of glutamatergic signaling associates with all forms of brain activity including learning and memory. Ionotropic glutamate receptors (iGluRs) are glutamate-gated ion channels that convert presynaptically released glutamate into a biological signal by opening of the ion channel pore, a process referred to as gating. N-methyl-D-aspartate receptors (NMDARs), a particular iGluR subtype, are obligate hetero-tetramers, formed by two glycine-binding GluN1 subunits and typically two glutamate-binding GluN2(A-D) subunits. NMDARs contribute to normal brain physiology, which are highlighted in its contribution to neurological disorders such as autism, epilepsy and Alzheimer's diseases. Understanding the mechanistic basis of NMDAR function has broad implications to brain function as well as clinical interventions. Fast glutamate applications to single channel patches is a powerful approach for studying underlying gating details. Notably, single NMDARs show either successes (activation) or failures (no activation), with successes showing variations in the delay to opening. Failures and delays to opening will impact how strongly the channel contributes to fast synaptic signaling. Here, I addressed potential mechanisms regulating failure rates and delays to opening that are relevant to the function of NMDARs at synapses. In terms of failures, one factor is the time between glutamate applications, as failure rates are significantly reduced with longer wait times. Additionally, the status of pH and CTD impact the activation efficiency of NMDARs, which indicates their important role in channel activity. In terms of subunit dependence, GluN2B-NMDARs with a much lower open probability than GluN2A-NMDARs, have a significantly reduced efficiency and shows greater variation. Interestingly, GluN2C-NMDARs with an even lower open probability shows comparable failure rates to GluN2A-NMDARs. Our data indicate intermediate activation pathways are a mode by which NMDARs are modulated: the physiological environment and subunit composition will have profound effects on NMDARs activation during repetitive activity, which would strongly impact Ca2+ influx mediated. Characterization of subunit-specificity of failures and delays to opening will allow us to understand the functional diversity of NMDAR subtypes, which is critical to the improvement of clinical approaches for neurological diseases.

Disclosures: M. He: None. **L.P. Wollmuth:** None.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.07

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: CIHR MOP119298

Title: Investigating the role of N-methyl D-Aspartate Receptors in learning related myelin remodelling.

Authors: *N. VASWANI, P. S. FINNIE, M. T. SULLIVAN, A. J. RAMSEY;
Pharmacol., Univ. of Toronto, Toronto, ON, Canada

Abstract: Consolidation of fear, spatial and motor learning has been demonstrated to be accompanied by proliferation of oligodendrocyte precursor cells (OPCs) and to require generation of new oligodendrocytes and de novo myelination. While studies have demonstrated immediate differentiation of pre-existing OPCs into immature oligodendrocytes as important for both motor and spatial learning, transgenic animals incapable of OPC differentiation exhibit normal recall of recent fear memories, leading authors to conclude that deficits in memory at remote time points are solely due to a lack of differentiation of newly proliferated precursor cells. Here, using in-situ hybridization and immunohistochemistry colabelling, we investigate the expression and spatial distribution of “newly differentiated” pre-myelinating oligodendrocytes and cFos in the brains of adult male and female c57Bl6/J mice 90 minutes after contextual fear conditioning. In vitro studies in OPC-neuron co-cultures and rodent studies following remyelinating white matter lesions suggest that N-methyl D-Aspartate receptors (NMDARs), a type of ionotropic glutamate receptor, are involved in activity dependent differentiation of OPCs. We show that subchronic treatment with NMDAR antagonist dizocilpine (MK801) post conditioning (i.p, 0.2 mg/kg, 1 time daily, 25 days) does not alter locomotor activity, yet reduces freezing behavior (P=0.038, n=6) during remote fear memory retrieval. In order to evaluate whether this is related to learning related myelin remodeling we investigated the effects of post conditioning MK801 treatment on the immediate OPC differentiation and proliferation using in-situ hybridization and immunohistochemistry. Through these studies we attempt to further elucidate mechanisms of neuronal activity dependent white matter plasticity and explore putative pharmacological intervention for the attenuation of fear memory.

Disclosures: N. Vaswani: None. **P.S. Finnie:** None. **M.T. Sullivan:** None. **A.J. Ramsey:** None.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.08

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NIH R01GM128195

Title: Characterization of NMDAR membrane to channel inhibition by (+)-MK-801

Authors: *A. NIGAM, E. G. NEUREITER, J. W. JOHNSON;
Dept. of Neurosci., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: *N*-methyl-D-aspartate receptor (NMDAR) inhibition is extensively studied due to its therapeutic potential. NMDARs are calcium-permeable glutamate receptors expressed broadly in the brain that are implicated in many neurological disorders including Alzheimer's disease (AD). They are tetrameric ionotropic receptors composed of two obligatory GluN1 subunits and two GluN2 (A-D) and/or GluN3 (A-B) subunits. Channel blocking inhibitors such as memantine (an FDA-approved drug for treatment of AD) and MK-801 exhibit two distinct inhibitory mechanisms: the extensively studied traditional channel block, and membrane to channel inhibition (MCI). Traditional channel block occurs when charged blocker molecules from the extracellular solution enter the open channels of agonist-bound NMDARs, blocking ion flux. Our lab recently showed that memantine MCI occurs when uncharged memantine molecules in the extracellular solution enter the membrane, and then transit from membrane to open channels of agonist-bound NMDARs through a fenestration, blocking ion flux. Here, we use tsA201 cells transfected to express NMDARs to investigate properties of MK-801 MCI using previously-established protocols. We previously showed that the MK-801 MCI IC_{50}s for diheteromeric GluN1/2A and GluN1/2B receptors are similar. In contrast, GluN1/2C and GluN2D receptors showed weak MCI measured by Min I_{MCI}/I_{Con} (minimum NMDAR-mediated current during MCI relative to control current), following application of 10 μM MK-801 to GluN1/2D (Min I_{MCI}/I_{Con} 0.95 ± 0.02) or 100 μM MK-801 to GluN1/2C (Min I_{MCI}/I_{Con} 0.48 ± 0.05) receptors. We investigated the time course of MK-801 exit from the membrane using GluN1/2A and GluN1/2B receptors and found that MK-801 exits the membrane with time constants of 9.27 ± 0.22 s and 8.12 ± 0.23 s respectively. When compared with previous measurements, these data suggest that MK-801 exits the membrane much more slowly than memantine. We studied voltage dependence of MK-801 entry into the membrane using 1 μM MK-801 and GluN1/2A receptors. Min I_{MCI}/I_{Con} was compared using two protocols: with MK-801 applied at -65 mV and MCI measured at -65 mV, or with MK-801 applied at 35 mV and MCI measured at -65 mV (the voltage jump protocol). We found that Min I_{MCI}/I_{Con} was not significantly different when the entire protocol was performed at -65 mV (0.38 ± 0.04) and when the voltage jump protocol was used (0.25 ± 0.05; p =0.11). Thus, MK-801 entry into the membrane is not voltage dependent. We expect that continued characterization of MK-801 MCI will help deepen understanding of NMDAR channel block.

Disclosures: A. Nigam: None. **E.G. Neureiter:** None. **J.W. Johnson:** None.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.09

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Title: Simulative Analysis of NMDA Spikes by Using a Circuits Simulator for VLSI design

Authors: *F. HONDO[1], T. ISHIKAWA[2,3], A. KOSUGE[1], M. HAMADA[1], T. KURODA[1];
[1]Sch. of Engin., The Univ. of Tokyo, Tokyo, Japan; [2]Pharmacol., Keio Univ., Tokyo, Japan;
[3]The Picower Inst., MIT, Cambridge, MA

Abstract: Neuromorphic computing has gained much attention as a method of reduction of power consumption of artificial intelligence (AI) processors. On the other hand, neuron models employed in neuromorphic computing are single-compartment, and therefore they cannot implement active properties such as dendritic spikes. Incorporating dendritic nonlinearities into the computational models is expected to improve the computing capability of neuron models, hence reducing the cost and power consumption of AI Processor. In this study, we simulated *N*-methyl-D-aspartic acid (NMDA) spikes, a type of dendritic spikes, by using Virtuoso, a computer-aided design (CAD) software for integrated circuits (IC). Models of α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptors and NMDA receptors were expressed by equivalent circuits consisting of some electrical components such as, resistor (R), inductor (L), capacitor (C), voltage-controlled current source, voltage-controlled voltage source, and operational amplifier. The effects of varying the NMDA/AMPA ratio and the number of inputs were examined. The output of each receptor was modeled as an electrical current. The waveform was reproduced by the output current proportional to the step responses of the built-in RLC band pass filter. The rise and fall time constants were set independently by adjusting R, L and C. The magnitude of the current depends on the membrane potential. The I-V curve was approximated by a piecewise linear (PWL) function. The simulation proved that NMDA spikes requires an NMDA/AMPA ratio greater than 1. This result corresponds with simulations with a multi-compartment model of a barrel cortex layer 5 pyramidal neuron. The excitatory postsynaptic potentials (EPSPs) were superlinear when the membrane potential was lower than -30 mV, where the I-V curve of NMDA receptors reaches the maximum. Otherwise, the EPSPs were compressed. These characteristics are consistent with experimental data of neocortical layer 5 pyramidal neurons recorded by patch-clamp technique. The implementation of the I-V curve by a PWL function is directly applicable to the experimental results, and thereby useful for reflecting the latest reports of properties of receptors and ion channels. Moreover, because semiconductor circuits simulators are designed to apply to the large networks, it is expected that the presented method is extended to the analytics of more complex neural models.

Disclosures: F. Hondo: None. **T. Ishikawa:** None. **A. Kosuge:** None. **M. Hamada:** None. **T. Kuroda:** None.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.10

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: For K. B. Hansen: NIH NS097536
For K. B. Hansen: NIH NS116055
For K. B. Hansen: NIH GM103546
For F. Zhao: China Scholarship Council

Title: Design and characterization of potent NMDA receptor glycine site agonists with GluN2 subunit-specific activity

Authors: *E. DIAMANT[1], F. ZHAO[1], S. MARIOTTINI[1], U. ATXABAL[1], F. YI[2], J. S. LOTTI[2], N. ROUZBEH[2], N. LIU[1], L. BUNCH[1], K. B. HANSEN[2], R. P. CLAUSEN[1];
[1]Dept. of Drug Design and Pharmacol., Univ. of Copenhagen, Copenhagen, Denmark; [2]Dept. of Biomed. and Pharmaceut. Sci., Univ. of Montana, Missoula, MT

Abstract: N-methyl-D-aspartic acid receptors (NMDARs) are implicated in several psychiatric and neurological diseases. NMDARs are tetrameric subunit complexes composed of two glycine-binding GluN1 and either two glutamate-binding GluN2 subunits (GluN2A-D) or two glycine-binding GluN3 subunits (GluN3A-B). Due to high sequence identity among the GluN2 subunits, the development of subtype-selective NMDAR ligands targeting the glutamate binding site has to this date been unsuccessful. We aimed to explore potency, efficacy, and NMDAR subtype-selectivity for a new class of glycine site agonists that bind GluN1 to activate NMDARs in a GluN2-subunit specific manner. We designed 19 furanylamide derivatives of the GluN1 glycine site agonist AICP, which is superagonist at GluN1/2C, full agonist at GluN1/2A, and partial agonist at GluN1/2B and GluN1/2D. The structure-activity relationship of the compounds was evaluated at recombinant NMDAR subtypes using two-electrode voltage-clamp recordings. No discernible agonist activity was seen at the GluN1/2B subtype for any of the analogs, but all compounds showed activity at the GluN1/2C subtype. The agonists show partial, full, or superagonism ranging from <10% to 157% relative to the maximal response compared to glycine. Six analogs were GluN1/2C-specific agonists with no discernible agonist activity at GluN1/2A, GluN1/2B, and GluN1/2D subtypes. Several of the agonists showed high potencies with EC_{50} values in the low nanomolar range (< 100 nM). Induced-fit computational docking into the GluN1 glycine binding site demonstrated that fundamental ligand-receptor interactions for the agonist -amino part were conserved and consistent with previous literature. In addition, the agonists were predicted to protrude into a side pocket of the orthosteric GluN1 binding site oriented toward the interface of GluN1 and GluN2 subunits. Consistent with this predication, it was found that a non-conserved residue at the position of GluN2A Val783 in the GluN1-GluN2 dimer interface influenced the GluN2-specific agonist efficacy. Furthermore, the results from computational docking were consistent with the binding mode found in crystal structures of the isolated GluN1/2A agonist binding domain heterodimer. Our findings expand the synthetic pharmacology of NMDARs and contribute to a deeper understanding of subtype-specific NMDAR-modulation through the orthosteric GluN1 glycine binding site. With the significant variation in potency, efficacy, and selectivity, this new class of NMDAR agonists may provide innovative therapeutic strategies in the treatment of neurological and psychiatric disorders.

Disclosures: **E. Diamant:** None. **F. Zhao:** None. **S. Mariottini:** None. **U. Atxabal:** None. **F. Yi:** None. **J.S. Lotti:** None. **N. Rouzbeh:** None. **N. Liu:** None. **L. Bunch:** None. **K.B. Hansen:** None. **R.P. Clausen:** None.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.11

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NIH NINDS intramural research program 1ZIANS002994-20

Title: A novel function of CNKSR2 associated with NMDA receptor

Authors: *S. WON, K. ROCHE;
NINDS, NIH, Bethesda, MD

Abstract: N-methyl-D-aspartate receptors (NMDARs) are ionotropic glutamate receptors, which are widely expressed throughout the nervous system. NMDARs play crucial roles in neuronal development, synaptic plasticity, learning and memory, and neuropsychiatric disorders. The functional NMDAR subunits assemble as hetero-tetramers consisting of two GluN1 subunits and two subunits that are either GluN2 (A-D) or GluN3 (A-B). In particular GluN2 subunits define many of the functional properties of the channel, including their trafficking and their synaptic expression. In cerebral cortex and hippocampus, NMDARs are primarily composed of two GluN1 subunits, and two GluN2A and GluN2B. GluN2A and GluN2B have common binding protein regions and posttranslational modification sites such as phosphorylation, whereas they have distinct endocytic motifs in their C-terminal tails. The most well-known region in their cytosolic tail is the postsynaptic density-95 (PSD-95)/Dlg-A/ZO-1 (PDZ) binding motif (ESDV). PSD-95 is a member of the membrane-associated guanylate kinase (MAGUKs) family of proteins that is well-known for binding to the PDZ binding motif in the C-terminus of the GluN2 subunits of NMDARs. This PSD-95 binding stabilizes the surface expression of GluN2 subunit containing-NMDARs at synaptic membranes. Connector Enhancer of Kinase Suppressor of Ras2 (CNKSR2), also known as CNK2 or MAGUIN, is a synaptic scaffolding protein and was identified as a binding protein for PSD-95 and synaptic scaffolding molecule (S-SCAM). CNKSR2 is a homologue of CNK discovered first in association with Ras/Mitogen-activated protein kinase (MAPK) signaling during eye development in *Drosophila*. The CNKSR2 protein is encoded by the *CNKSR2* gene located on the X-chromosome. Many CNKSR2 human mutations are associated with neurodevelopmental disorders including X-linked Intellectual Disability (XLID), Autism Spectrum Disorder (ASD), Attention Deficit Hyperactivity Disorder (ADHD), and epilepsy. However, the molecular mechanisms have not been elucidated. CNKSR2 is a multi-domain protein including Sterile Alpha Motif (SAM), Conserved Region in CNKSR2 (CRIC), PDZ, and Pleckstrin Homology (PH) domains. CNKSR2 is brain specific and enriched at neuronal synapses. We have characterized CNKSR2 expression during development, in

different brain regions, and its subcellular localization. We have also investigated the CNKSR2 binding proteins at neuronal synapses. Interestingly, CNKSR2 is enriched at the PSD in neurons and it binds to NMDARs as well as PSD-95. These results implicate CNKSR2 as an important interactor of PSD-95 and NMDARs.

Disclosures: S. Won: None. **K. Roche:** None.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.12

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NIH grant R01GM128195
NIH grant T32NS007433

Title: NMDA receptor subtype dependence of inhibition by intracellular (+)-MK-801

Authors: *E. G. NEUREITER, A. NIGAM, J. W. JOHNSON;
Neurosci., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: *N*-methyl-D-aspartate receptors (NMDARs) are glutamate gated ion channels present at most excitatory synapses in the brain. They are involved in learning, memory, and synaptic function and are implicated in a range of diseases. Thus, pharmacology of NMDARs has broad therapeutic implications. MK-801 is a high affinity NMDAR channel blocking antagonist that is used as a research tool to study synaptic function, often applied intracellularly to inhibit postsynaptic NMDARs. However, the underlying mechanism by which intracellular MK-801 (iMK-801) inhibits NMDARs is not well understood. Here, we investigate inhibition of NMDARs by iMK-801. We performed whole-cell patch-clamp electrophysiology on tsA201 cells transfected to express diheteromeric NMDARs composed of GluN1 and GluN2A (GluN1/2A) and GluN1/2C receptors. We characterized subtype dependence, amount, and time course of inhibition by 0.1 and 10 mM iMK-801. We quantified iMK-801 inhibition by recording at a depolarized membrane potential (V_m) of 65 mV to alleviate iMK-801 block, followed by a jump to a hyperpolarized V_m of -65 mV to measure current amplitude and rate of reblock. Inhibition by iMK-801 was measured by calculating the ratio between the peak current after the hyperpolarizing step (I_{peak}) and the steady state current after iMK-801 reblock at -65 mV (I_{ss}).We found substantial NMDAR subtype dependence of iMK-801 inhibition: 10 mM MK-801 induced significantly more inhibition of GluN1/2A receptors (I_{ss}/I_{peak} = 0.069 ± 0.01) than of GluN1/2C receptors (I_{ss}/I_{peak} = 0.41 ± 0.01) ($p < 0.0001$). There was also a significant difference in inhibition with 0.1 mM iMK-801 between GluN1/2A (I_{ss}/I_{peak} = 0.23 ± 0.12) and GluN1/2C receptors (I_{ss}/I_{peak} = 0.67 ± 0.06) ($p = 0.02$). I_{ss}/I_{peak} was also significantly different within subtypes between 0.1 and 10 mM iMK-801 ($p = 0.04$ for GluN1/2A and $p = 0.01$ for GluN1/2C). We analyzed the time constant of iMK-801 inhibition in GluN1/2A by fitting a double

exponential curve to the NMDAR current decay that followed the V_m jump from 65 to -65 mV. The fast time constant was significantly faster in 10 mM MK-801 (23.2 ± 5.8 ms) than in 0.01 mM MK-801 (2080 ± 1253 ms) (p = 0.04).The difference in inhibition of GluN1/2A and GluN1/2C receptors indicates that even high concentrations of iMK-801 do not effectively inhibit all NMDAR subtypes. The strong subtype dependence of inhibition by iMK-801 also suggests the specific mechanisms of inhibition by intra- and extracellular MK-801 may differ, since inhibition by extracellular MK-801 does not display strong NMDAR subtype dependence. Further exploration of the mechanisms of iMK-801 action will deepen our understanding of drug action on NMDARs.

Disclosures: E.G. Neureiter: None. **A. Nigam:** None. **J.W. Johnson:** None.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.13

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: CIHR PG 156223
NSERC DG 2017-04730
Healthy Brains Healthy Lives
Fonds de Recherche du Québec

Title: NMDA receptors in synaptic plasticity: more than coincidence

Authors: *S. RANNIO[1,2], A. THOMAZEAU[1], J. A. BROCK[1,2], B. KO[1], G. MOFFAT[1], P. J. SJÖSTRÖM[1];
[1]Ctr. for Res. in Neurosci., [2]Integrated Program in Neurosci., McGill Univ., Montreal, QC, Canada

Abstract: Classically, ionotropic postsynaptic NMDA receptors (postNMDARs) act as coincidence detectors in Hebbian plasticity. However, NMDARs can also signal metabotropically even when Mg^{++}-blocked, and can be found presynaptically (preNMDARs), two scenarios where Hebbian coincidence detection is not possible. We previously showed in primary visual cortex (V1) layer-5 (L5) pyramidal cells (PCs) that preNMDARs signal ionotropically via RIM1αβ to boost high-frequency evoked release, but metabotropically via JNK2 to regulate spontaneous release independent of frequency. As L5 PC timing-dependent long-term depression (tLTD) is independent of frequency, we hypothesized that tLTD also requires metabotropic preNMDAR signalling via JNK2. By genetic deletion, we additionally tested if pre- or postNMDARs govern synaptic plasticity.
Quadruple patch in P11-18 mouse acute V1 slices revealed that the JNK2 inhibitor SP600125 abolished tLTD (96% ± 2%, n = 9 vs. tLTD 62% ± 4%, n = 15, p < 0.001) but homozygous RIM1αβ deletion did not (deletion 65% ± 7%, n = 10 vs. tLTD, p = 0.74). Also, 7-CK did not

affect tLTD (73% ± 5 %, n = 9, $p < 0.001$), confirming that tLTD relies on metabotropic but not ionotropic preNMDAR signalling. In agreement with presynaptic induction, pre- but not postsynaptic dialysis of a JNK2-blocking peptide abolished tLTD (pre 96 ± 4%, n = 10 vs. tLTD 52% ± 6%, n = 7, $p < 0.001$; post 61± 5%, n = 12 vs. tLTD, $p = 0.30$). To globally delete NMDARs in all PCs, we created the triple transgenic mouse model $Emx1^{Cre/+}$;$Ai9^{tdTom/+}$;$NR1^{flox/flox}$. To sparsely delete NMDARs, we injected AAV9-eSYN-mCherry-iCre-WPRE in V1 of P1 $NR1^{flox/flox}$ mice. MNI-NMDA uncaging verified postNMDARs deletion in global (0.02 ± 0.5 pA, n = 31 vs. control -48 ± 6 pA, n=19; $p < 0.001$) and sparse models (0 ± 0 pA, n = 9 vs. control -31 ± 8 pA, n = 14; $p < 0.001$). Consistent with preNMDAR deletion, AP5 did not affect mEPSC frequency in the global model (92% ± 4%, n = 18 vs. control 60% ± 12%, n = 5; $p < 0.05$). Spine density was reduced in the global model (2.5 ± 0.2 spines/10 µm, n =29; vs. 3.8 ± 0.2 spines/10 µm, $p < 0.001$), suggesting that NMDARs promote synapse formation/stability. In the sparse model, AP5 failed to suppress EPSPs in PC-PC pairs with preNMDAR deletion (102% ± 10%, n = 4), yet successfully did with no or postNMDAR deletion (pooled: 63% ± 10%, n = 6; $p < 0.05$), indicating that pre- but not postNMDARs control short-term plasticity. In the sparse model, we will next show how pre/postNMDAR deletion impacts tLTD.
In conclusion, preNMDARs control both short and long-term plasticity, but in different ways: the former ionotropically and the latter metabotropically. At V1 L5 PC synapses, NMDARs thus do more than classic Hebbian coincidence.

Disclosures: **S. Rannio:** None. **A. Thomazeau:** None. **J.A. Brock:** None. **B. Ko:** None. **G. Moffat:** None. **P.J. Sjöström:** None.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.14

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: Academy of Sciences of the Czech Republic (RVO 61388963)
Technology Agency of the Czech Republic, National Centres of Competence (NCK1) Project No. TN01000013, Personalized Medicine - Diagnostics and Therapy

Title: Identification of N-methyl-D-aspartate receptor antagonists using the rat postnatal mixed cortical and hippocampal neurons

Authors: *J. VOLDRICH[1,2], M. MATOUSOVA[3], M. SMIDKOVA[3], B. SLAVIKOVA[3], H. CHODOUNSKA[4], E. KUDOVA[4], H. MERTLIKOVA-KAISEROVA[3];
[1]IOCB Prague, IOCB Prague, Prague, Czech Republic; [2]UCT, Prague, Czech Republic; [3]IOCB, Prague, Czech Republic; [4]Inst. of Organic Chem. and Biochem. CA, Inst. of Organic Chem. and Biochem. CA, Prague, Czech Republic

Abstract: One of the most essential steps during drug discovery is to develop a reasonable screening cascade. Typical primary tool used for screening of modulators of *N*-methyl-*D*-aspartate receptor (NMDAR) are heterologous expression systems with defined NMDAR subunit composition. These models are particularly useful for the first selection of the active compounds from libraries. However, the compound activity can be affected by the absence of neuronal microenvironment. The most complex and physiologically relevant *in vitro* models recently used in neuropharmacology research include brain slice cultures and organoids. The main disadvantage of these cultures is low throughput, and therefore they are more suitable for mechanistic studies. The compromise solution that preserves natural physiological functions of neuronal cells but is also amenable for medium-to-high throughput screening is represented by (1) immortalized cell lines differentiated to neuron-like phenotype, (2) iPSC-derived neurons generated from human somatic cells or (3) primary cultures isolated from brain tissue. The aim of this study was to find out, whether mixed cortical and hippocampal primary rat postnatal neuronal culture is suitable *in vitro* tool for identification of NMDAR antagonists and if it is comparable with other generally used primary rat neuronal models differing in the cell type (pure cortical vs. mixed cortical and hippocampal) and developmental stage (embryonal vs. postnatal). The neuron/astrocyte ratio of each culture was first evaluated by flow cytometry and confocal microscopy using neuronal nuclei (NeuN) protein and DAPI immunocytochemistry staining. The cultures were further characterized by gene expression analysis of NMDAR subunits NR2B/NR2A, glutamate transporter GLT1 and glutamate-aspartate transporter GLAST by qPCR. Selected endogenous and synthetic neurosteroids that have been previously identified as antagonists of recombinant GluN1/GluN2B NMDA receptors by patch clamp technique were then assessed for the ability (1) to decrease the excitotoxic effect induced by glutamate or *N*-methyl-*D*-aspartate (NMDA) over 24 hours by measuring cell viability, and (2) to inhibit an acute NMDA or glutamate-induced Ca^{2+} influx using fluorometric probe Fura-2. Postnatal mixed cortical and hippocampal culture showed to be a reliable, easy-to-cultivate and robust tool for NMDAR antagonist screening. Embryonal (E18) cells, that are usually considered as “gold standard” among primary neuronal cultures, offered higher cell yields, but they evinced increased sensitivity to compounds’ cytotoxicity.

Disclosures: J. Voldrich: None. **M. Matousova:** None. **M. Smidkova:** None. **B. Slavikova:** None. **H. Chodounska:** None. **E. Kudova:** None. **H. Mertlikova-Kaiserova:** None.

Poster

521. NMDA Receptors: Biophysical Properties and Pharmacology

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 521.15

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: U.S. Department of Health & Human Services | NIH | Eunice Kennedy Shriver National Institute of Child Health and Human Development (NICHD) - ZIA-HD000711 [Buonanno]

Title: Pathway-specific contribution of parvalbumin interneuron NMDA receptors to synaptic currents and thalamocortical feedforward inhibition

Authors: *E. M. LEWIS, H. E. SPENCE, N. AKELLA, A. BUONANNO;
Eunice Kennedy Shriver Natl. Inst. of Child Hlth. and Human Develop., Bethesda, MD

Abstract: Prefrontal cortex (PFC) is a site of information convergence important for behaviors relevant to psychiatric disorders. Despite the importance of inhibitory GABAergic parvalbumin-expressing (PV+) interneurons to PFC circuit function and decades of interest in N-methyl-D-aspartate receptors (NMDARs) in these neurons, examples of defined circuit functions that depend on PV+ interneuron NMDARs have been elusive. Indeed, it is still controversial whether adult PFC PV+ interneurons contain functional NMDARs at all, which has major consequences for hypotheses of the pathogenesis of psychiatric disorders. Using a combination of fluorescent *in situ* hybridization, pathway-specific optogenetics, cell-type-specific gene ablation, and electrophysiological recordings from PV+ interneurons, here we resolve this controversy. We found that nearly 100% of PV+ interneurons in adult medial PFC (mPFC) express transcripts encoding GluN1 and GluN2B, and they have functional NMDARs. By optogenetically stimulating corticocortical and thalamocortical inputs to mPFC, we show that synaptic NMDAR contribution to PV+ interneuron EPSCs is pathway-specific, which likely explains earlier reports of PV+ interneurons without synaptic NMDAR currents. Lastly, we found a major contribution of NMDARs in PV+ interneurons to thalamus-mediated feedforward inhibition in adult mPFC circuits, suggesting molecular and circuit-based mechanisms for cognitive impairment under conditions of reduced NMDAR function. These findings represent an important conceptual advance that have major implications for hypotheses of the pathogenesis of psychiatric disorders.

Disclosures: E.M. Lewis: None. **H.E. Spence:** None. **N. Akella:** None. **A. Buonanno:** None.

Poster

522. Muscarinic, Serotonergic, and Metabotropic Glutamate Receptors

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 522.01

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: AG005214

Title: Synthesis and pharmacological evaluation of a series of potential allosteric modulators of acetylcholine efficacy at M_1 muscarinic receptors

Authors: C. WIDMAN[1], G. KAUP[2], H. SMITH[2], S. VENTRESCA[2], E. SLANE[2], G. ELMSLIE[3], J. ELLIS[3], W. S. MESSER, Jr.[2], ***I. SCHIEFER**[1];
[1]Medicinal and Biol. Chem., [2]Pharmacol. and Exptl. Therapeut., Univ. of Toledo, Toledo, OH;
[3]Psychiatry and Pharmacol., Penn State Univ., Lebanon, PA

Abstract: M_1 muscarinic receptors play important roles in mediating the effects of acetylcholine in multiple brain regions including the cerebral cortex, hippocampus and neostriatum. Allosteric modulators of M_1 muscarinic receptors are of particular interest due to their ability to shape acetylcholine signaling in precise spatiotemporal patterns. Moreover, compounds that enhance the efficacy of acetylcholine could help restore function in signaling pathways that are damaged due to disease processes. Several compounds have been identified that enhance the potency of acetylcholine at M_1 muscarinic receptors, yet relatively few compounds increase acetylcholine efficacy. A library of structurally-related, drug-like potential allosteric modulators was synthesized and evaluated for the ability to modulate acetylcholine efficacy and/or potency – as well as levels of intrinsic allosteric agonist activity (allosteric modulators of M_1 muscarinic receptors with high levels of intrinsic activity are associated with adverse cholinergic effects). Each compound was characterized by proton and carbon NMR, HPLC-UV, high resolution mass spectral analysis and/or elemental analysis. Pharmacological activity was evaluated by measuring [^{3}H]-arachidonic acid release in CHO cells expressing human M_1 muscarinic receptors in the absence or presence of 10 µM allosteric modulator and in the presence or absence of acetylcholine (0.1 µM or 100 µM). Several compounds were identified that enhanced responses produced by 100 µM acetylcholine greater than 25%, including: CW-6-65, CW-6-77, CW-7-22, CW-7-34, HS-1-40, and GK-1-15. A few analogs, including CW-6-45, CW-6-56, and CW-7-29, did not enhance the activity of 100 µM acetylcholine, yet reduced 0.1 µM acetylcholine activity more than two-fold. All compounds demonstrated relatively low levels of intrinsic allosteric activity (less than 25% of the maximal acetylcholine response). Together, these data delineate important structure activity relationships for modulating acetylcholine responses at muscarinic receptors and identify compounds that may enhance acetylcholine efficacy.

Disclosures: C. Widman: None. **G. Kaup:** None. **H. Smith:** None. **S. Ventresca:** None. **E. Slane:** None. **G. Elmslie:** None. **J. Ellis:** None. **W.S. Messer:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Psyneurgy Pharmaceuticals LLC. **I. Schiefer:** None.

Poster

522. Muscarinic, Serotonergic, and Metabotropic Glutamate Receptors

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 522.02

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NIH Grant AG005214

Title: Pharmacological characterization of novel allosteric modulatorsof ACh efficacy at M_1 muscarinic receptors.

Authors: *J. ELLIS[1], G. ELMSLIE[1], S. VENTRESCA[2], E. SLANE[2], I. T. SCHIEFER[2], C. WIDMAN[2], G. KAUP[2], H. SMITH[2], W. S. MESSER, Jr[2];
[1]Penn State Univ., Hershey, PA; [2]The Univ. of Toledo, Toledo, OH

Abstract: M_1 muscarinic receptors are G protein-coupled receptors that mediate the effects of acetylcholine in multiple brain regions. The development of compounds that can enhance M_1 muscarinic receptor activity could be useful in the treatment of neurological disorders associated with impaired cholinergic signaling. In contrast to directly acting muscarinic agonists, compounds that interact with allosteric binding sites may preserve the spatiotemporal patterning of muscarinic receptor activity that is stimulated by the physiological release of acetylcholine. Allosteric modulators can exert multiple effects, including modulation of potency (binding cooperativity), efficacy (activation cooperativity) and intrinsic activity (allosteric agonist activity).
Preliminary studies identified several compounds that modulated the effects of 0.1 or 100 µM acetylcholine when tested at 10 µM in a [^{3}H]-arachidonic acid release assay from CHO cells expressing human M_1 muscarinic receptors. Full acetylcholine dose-response curves were generated for active compounds to determine effects on acetylcholine potency and/or efficacy. Several compounds, including CW-6-65, CW-6-77, CW-7-22, CW-7-34, HS-1-40, and GK-1-15, enhanced the maximal response to acetylcholine with minimal effects on acetylcholine potency and limited intrinsic activity. These compounds exhibited properties consistent with positive allosteric modulation of acetylcholine efficacy. In contrast, CW-6-45, CW-6-56, and CW-7-29 shifted acetylcholine dose-response curves to the right without appreciably changing the maximal response produced by acetylcholine or elevating baseline activity. These compounds behaved as negative allosteric modulators of acetylcholine potency.
Three compounds (CW-6-56, CW-6-65 and CW-6-77) were evaluated further over a wide range of concentrations to assess the parameters K_B, tau_B, alpha, and beta by fitting the data to the allosteric operational model. Both CW-6-65 and CW-6-77 displayed a high degree of positive activation cooperativity with beta values of 3.26 and 3.89, respectively and high affinity with $logK_B$ values of -6.26 and -5.53 respectively. In contrast, CW-6-56 displayed a high degree of negative binding cooperativity with an alpha value of 0.180 and a $logK_B$ of -6.20. Compounds such as CW-6-65 and CW-6-77 could be useful as starting points for the development of selective allosteric modulators of acetylcholine efficacy.

Disclosures: J. Ellis: None. **G. Elmslie:** None. **S. Ventresca:** None. **E. Slane:** None. **I.T. Schiefer:** None. **C. Widman:** None. **G. Kaup:** None. **H. Smith:** None. **W.S. Messer:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Psyneurgy Pharmaceuticals LLC.

Poster

522. Muscarinic, Serotonergic, and Metabotropic Glutamate Receptors

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 522.03

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NIH Grant AG005214

Title: A phenotypic screen for identifying compounds that decrease repetitive behaviors in zebrafish

Authors: J. DIETRICH[1], A. WESLEY[1], F. E. WILLIAMS[1], C. WIDMAN[1], G. KAUP[1], I. T. SCHIEFER[1], J. ELLIS[2], ***W. S. MESSER, Jr.**[1];
[1]Univ. of Toledo, Toledo, OH; [2]Psychiatry and Pharmacol., Penn State Univ., Lebanon, PA

Abstract: Repetitive behaviors are associated with several neurological disorders, including autism spectrum disorders, obsessive-compulsive disorder and Tourette syndrome. A high-throughput behavioral screen was developed to monitor repetitive behaviors in wild-type (AB) zebrafish and identify compounds that decrease levels of repetitive behaviors. Experiments were conducted using 5 days post fertilization (dpf), wild-type zebrafish larvae. First, a maximum tolerated concentration (MTC) assay was performed to identify the maximum concentration of each compound not causing general toxicity or morphological abnormalities. Then, 5-dpf zebrafish were exposed to a range of concentrations of compounds in a behavioral assay in alternating light and dark periods for a total of 90 minutes. Video tracking software (Noldus DanioVision) was used to measure locomotor activity and other parameters including angular velocity and variance of turn angle - measures of repetitive behaviors. Preliminary studies examined the effects of BQCA, a positive allosteric modulator (PAM) of M_1 muscarinic receptors. At doses of 3 and 10 µM, BQCA significantly decreased locomotor activity (swimming); angular velocity was significantly increased at 3 and 10 µM, while variance of turn angle was increased at 1, 3 and 10 µM. The MTC was 10 µM for BQCA. Several novel allosteric modulators of acetylcholine efficacy, including CW-6-65, CW-6-77, and CW-7-22 also significantly decreased swimming and increased angular velocity and variance of turn angle. The MTC was 30 µM for CW-6-65 and CW-6-77, and 100 µM for CW-7-22. Two compounds (CW-6-65 and CW-6-77) were evaluated further over a wide range of concentrations to identify an EC_{50} value and establish the minimum effective concentration (MEC) for decreasing repetitive behaviors. The EC_{50} values were 2.6 µM and 1.1 µM for CW-6-65 and CW-6-77, respectively, in good agreement with their affinity for allosteric binding sites at human M_1 muscarinic receptors. The MEC was 0.3 µM for both CW-6-65 and CW-6-77 indicating a therapeutic index of 100 for both compounds. Further studies are needed to evaluate the activity of these compounds at zebrafish muscarinic receptors.

Disclosures: J. Dietrich: None. **A. Wesley:** None. **F.E. Williams:** None. **C. Widman:** None. **G. Kaup:** None. **I.T. Schiefer:** None. **J. Ellis:** None. **W.S. Messer:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Psyneurgy Pharmaceuticals LLC.

Poster

522. Muscarinic, Serotonergic, and Metabotropic Glutamate Receptors

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 522.04

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: DBT-Wellcome Trust India Alliance IA/I/12/1/500529/WTDB
DST India, INSPIRE fellowship, 2016/IF60611

Title: Cholinergic activation tunes synaptic transmission and plasticity at Schaffer-collateral synapses during theta band activity

Authors: *R. SHARMA, S. NADKARNI;
IISER-PUNE, Pune, India

Abstract: Acetylcholine is a crucial neuromodulator for memory formation and recall. It acts on a family of ionotropic and metabotropic receptors with distinct expression loci over multiple timescales and has diverse downstream targets and outcomes. We construct a biophysical model of Schaffer-collateral (SC) synapses to study the effect of metabotropic acetylcholine receptors on synaptic transmission and plasticity. Hippocampal pyramidal neurons primarily express M1 and M4 receptors. M1 receptor expression is concentrated in the post-synaptic regions on the soma and dendrites. These are Gq/11 coupled protein receptors that are known to broadly enhance neuronal excitability by suppressing small hyperpolarizing calcium-activated potassium (SK) and KCNQ2/3 channels. The M4 receptors, on the other hand, are most densely populated in the pre-synaptic regions. They are G(i/o) coupled receptors and have been shown to suppress neurotransmitter release via their action on pre-synaptic voltage-gated calcium channels. These counteracting pre-post effects of acetylcholine seem puzzling and inefficient at first glance. Adding to the complexity of the modulatory activity of acetylcholine, neuronal firing and the consequent neurotransmitter release are noisy. We show that the suppression of vesicle release rate via M4 receptors enables the limited vesicle resource to sustain release for a longer time. The prolonged-release of vesicles creates a stronger correlation between the amount of neurotransmitter released and the pre-synaptic firing rate. Additionally, the activation of M1 receptors amplifies the weaker neurotransmitter signal to generate a stronger correlation of the post-synaptic response with pre-synaptic activity. In the hippocampus, theta-band activity coincides with elevated levels of acetylcholine. We investigate the action of cholinergic receptors on SC synapses during theta-entrained activity in CA3 neurons. Interestingly we see that the encoding mediated by acetylcholine is optimally tuned to respond to theta-rhythmic firing activity in CA3 neurons. The modulatory actions of acetylcholine on synaptic transmission filter out noise to support efficient transmission and encoding of information. Our model elucidates the subtle interplay between the M1 and M4 receptors and their role in orchestrating synaptic function.

Disclosures: R. Sharma: None. **S. Nadkarni:** None.

Poster

522. Muscarinic, Serotonergic, and Metabotropic Glutamate Receptors

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 522.05

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Title: Preclinical Characterization of the Potent M4 Muscarinic Receptor Positive Allosteric Modulator SUVN-L1305022

Authors: *J. THENTU, V. BENADE, K. BOJJA, M. RASHEED, A. SHINDE, N. GANUGA, R. MEDAPATI, M. SRIRANGAVARAM, R. KALLEPALLI, R. SUBRAMANIAN, R. NIROGI;
Suven Life Sci. Ltd., Hyderabad, India

Abstract: Cognitive impairments and psychotic symptoms observed in schizophrenia and Alzheimer's disease (AD) result in debilitating disruptions to the activities of daily living in the patients as well as their caregivers. Unfortunately, currently available treatment options like typical and atypical antipsychotics are constrained by modest efficacy, partial responsiveness, treatment resistance, and dose-limiting motor incoordination and extrapyramidal side effects. Selective activation of muscarinic M4 receptor may provide an alternative approach for the treatment of psychotic symptoms and cognitive impairments in patients with schizophrenia, and also for the behavioral disturbances observed in AD. SUVN-L1305022 is a novel, and potent positive allosteric modulator (PAM) acting at muscarinic receptor M4 subtype. SUVN-L1305022 was evaluated for its pharmacokinetics and brain penetration properties in rats. It was tested in behavioral efficacy models to assess its potential in alleviating psychotic symptoms. SUVN-L1305022 was also evaluated in neurochemistry studies to evaluate its effects on dopaminergic neurotransmission in the prefrontal cortex and striatum. When tested in rodents, SUVN-L1305022 showed excellent oral pharmacokinetics and brain penetration. In neurochemistry studies, SUVN-L1305022 negatively regulated dopaminergic tone in striatum and positively regulated dopaminergic tone in cortex suggesting its possible role in addressing both positive and negative symptoms of schizophrenia. In summary, results from the current preclinical studies support the development of SUVN-L1305022 for the potential treatment of both positive and negative symptoms associated with neurological disorders like schizophrenia, and AD.

Disclosures: **J. Thentu:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **V. Benade:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **K. Bojja:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **M. Rasheed:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **A. Shinde:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **N. Ganuga:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **R. Medapati:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **M. Srirangavaram:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **R. Kallepalli:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **R. Subramanian:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **R. Nirogi:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd.

Poster

522. Muscarinic, Serotonergic, and Metabotropic Glutamate Receptors

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 522.06

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: Authors are full time employees of Janssen Research & Development, LLC. The research was funded by the company.

Title: Characterization of Selective M5 Acetylcholine Muscarinic Receptor Modulators on Dopamine Signaling in the Mouse Striatum

Authors: *V. ZELL, S. NEEDHAM, S. MUKHERJEE, N. ROSCOE, G. WOODRUFF, M. MARELLA, P. BONAVENTURE, W. DREVETS, B. BALANA;
Janssen R&D, San Diego, CA

Abstract: The type-5 muscarinic acetylcholine receptor (M5) is almost exclusively expressed in midbrain dopamine neurons of the ventral tegmental area (VTA) and substantia nigra *pars compacta* (SNc), and thus is of particular interest for precise modulation of dopamine signaling that is central to multiple neuropsychiatric diseases. For instance, decreasing abnormally high dopamine transmission in the striatum has been a core approach in treating psychotic and manic symptoms of schizophrenia and bipolar disorders. In this study we characterize the expression of M5 in different cell types of the VTA and SNc, namely dopamine, glutamate and GABA neurons. We found that M5 was predominantly expressed in dopamine neurons. We used *ex vivo* fast-scan cyclic voltammetry (FSCV) in brain slices from wild-type and M5 knockout mice to evaluate the capacity of M5-selective allosteric modulators to modulate dopamine release in the striatum. ML375 (selective M5 negative allosteric modulator; 10 μM) significantly decreased electrically evoked dopamine release, whereas VU 0365114 (selective M5 positive allosteric modulator; 10 μM) significantly increased dopamine release. In the presence of the acetylcholine nicotinic receptor blocker dihydro-β-erythroidine (DHβE, 1 μM), pre-incubation of the slice with ML375 completely abolished the increase in dopamine release induced by Oxotremorine-M (Oxo-M; 10 μM), a non-selective acetylcholine muscarinic receptor agonist. Pre-incubation of the slice with VU 0365114 augmented and prolonged the increase in dopamine release induced by Oxo-M. Recordings in M5 knockout mice showed that Oxo-M did not significantly increase dopamine release in the dorsal striatum, confirming the selectivity of action of both compounds. In vivo, we observed that pharmacological (ML375, 50 mg/kg po) and genetic blockade of M5 (M5 knockout) increased locomotor response to amphetamine (1 mg/kg ip). In contrast to data reported in the literature (Gunter et al., 2018) ML375 did not decrease cocaine self-administration in rats using either fixed ratio or progressive ratio paradigms, and at various doses of cocaine and / or ML375. In conclusion, our comprehensive mapping study of M5 expression in multiple cell types of the midbrain, supports a functional pharmacological response mediated by M5 activation *ex vivo* where we highlighted the important role of M5 in bidirectionally shaping dopamine signaling but our negative in vivo data in the cocaine self-administration study illustrates the complexity of M5 modulation.

Disclosures: **V. Zell:** A. Employment/Salary (full or part-time):; Scientist. **S. Needham:** A. Employment/Salary (full or part-time):; ASSOCIATE SCIENTIST II. **S. Mukherjee:** A. Employment/Salary (full or part-time):; Full time intern. **N. Roscoe:** A. Employment/Salary (full

or part-time):; Scientist. **G. Woodruff:** A. Employment/Salary (full or part-time):; Senior Scientist. **M. Marella:** A. Employment/Salary (full or part-time):; Principal Scientist. **P. Bonaventure:** A. Employment/Salary (full or part-time):; BIOLOGY DISCOVERY SR DIRECTOR. **W. Drevets:** A. Employment/Salary (full or part-time):; VP Disease Area Leader Neuropsychiatry. **B. Balana:** A. Employment/Salary (full or part-time):; PRINCIPAL SCIENTIST.

Poster

522. Muscarinic, Serotonergic, and Metabotropic Glutamate Receptors

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 522.07

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: VRI-UC

Title: New perspectives on 5-HT1A receptor conformational dynamics

Authors: *A. ROBLES-MUÑOZ[1], G. E. TORRES[2], A. FIERRO[1];
[1]Pontificia Univ. Catolica de Chile, Santiago, Chile; [2]Molecular/Cellular & Biomed. Sci., CUNY Sch. of Med., New York, NY

Abstract: The activation of the 5HT1A receptor, a GPCR coupled to Gi protein, results in a decrease in the firing rate of serotonergic neurons. This effect makes 5HT1A receptors an attractive target for the treatment of neuropsychiatric diseases related to the dysregulation of brain serotonin concentrations. Studies on three-dimensional description of 5-HT1A and its conformational changes have been focused on the binding site and, in some cases, on the transmembrane domains (TMs). However, a detailed molecular description of the conformational changes involved in the mechanism of activation and inactivation of this receptor by serotonin is unknown.Here, using long-term *in-silico* studies, we describe the crucial role of the intracellular loop 3 (IL3) in conformational changes of the 5HT1A receptor when serotonin binds to the receptor. A comprehensive description of the 5HT1A/serotonin complex using molecular dynamic simulation over 1μs using correlation maps as well as, statistical significance, was performed. We identified conformational changes that correlated with the distances of the transmembrane domains. Our descriptive and quantitative results on the different conformational states of 5HT1A lead us to propose a concatenated mechanism dependent on the presence of serotonin. Finally, our results show a "*hinge effect*" of IL3 in the receptor activation process, which allows us to propose a new surface on 5HT1A as a possible target to regulate the activity in the intracellular segment. These results identify new target opportunities for the development of therapeutics involving the 5HT1A receptor.

Disclosures: A. Robles-Muñoz: None. **G.E. Torres:** None. **A. Fierro:** None.

Poster

522. Muscarinic, Serotonergic, and Metabotropic Glutamate Receptors

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 522.08

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: FRAXA Research Foundation
NIH/NINDS R15NS118352
NIH Grant RO1DA029639

Title: Activating 5-HT_{1A}Receptors to Correct Neuronal Hyperexcitability in FMR1 Knockout Mice

Authors: T. SARAF[1], *B. YANG[2], C. FOREST[2], C. E. CANAL[1];
[1]Pharmaceut. Sci., Mercer Univ. Col. of Pharm., Atlanta, GA; [2]Georgia Inst. of Technol., Atlanta, GA

Abstract: Fragile X syndrome (FXS)—caused by an epigenetic mutation of FMR1 that reduces or eliminates expression of the RNA-binding protein FMRP—is a neurodevelopmental disorder characterized by intellectual disabilities that are often comorbid with seizures, sensory hypersensitivities, anxiety, social-communication deficits, and repetitive behaviors. Neuronal hyperexcitability is an overarching feature of FXS that may underlie FXS symptoms. Like individuals with FXS, *Fmr1* knockout (KO) mice have seizures, EEG abnormalities, hyperexcitable pyramidal neurons, and altered expression of ion channels. Inhibitory 5-HT_{1A}Rs have not been reported as direct targets of FMRP, but they directly modulate the activity of neural systems disordered in FXS, e.g., the hippocampus and cortex. We are using a combinatorial approach—from receptor pharmacology to single-cell to EEG to behavioral experiments—to test the hypothesis that selectively activating 5-HT_{1A}Rs is pharmacotherapeutic for FXS. At the behavioral level, we tested the antiepileptic effects of the selective 5-HT_{1A}R agonist, NLX-112 (0.25-2.5 mg/kg). NLX-112 prevented audiogenic seizures in *Fmr1* KO mice, an effect that was blocked by the selective 5-HT_{1A}R antagonist, WAY-100635 (0.1 mg/kg). EEG recordings from above the left somatosensory cortex of awake, freely-moving, *Fmr1* KO mice showed significantly elevated relative gamma (30-100 Hz) power compared to control mice. In vitro whole-cell patch clamping data collected from hippocampal CA1 neurons demonstrated increased input resistance of *Fmr1* KO neurons. NLX-112 (10 μM) decreased the action potential duration of cells from control and *Fmr1* KO mice. At the receptor level, we observed higher whole brain 5-HT_{1A}R expression (B_{MAX}), but no changes in agonist or antagonist affinity at 5-HT_{1A}Rs (K_D) in male *Fmr1* KO mice. In vivo, *Fmr1* KO mice showed significantly elevated 5-HT_{1A}R-mediated behaviors when treated with the potent 5-HT_{1A}R agonist (*R*)-8-OH-DPAT (1 and 2 mg/kg), an effect that was blocked by pretreatment with WAY-100635 (0.1 mg/kg). Collectively, these results suggest that upregulation of inhibitory 5-HT_{1A}Rs in the brains of *Fmr1* KO mice may be a compensatory response to neuronal hyperexcitability, and that activating 5-HT_{1A}Rs may be pharmacotherapeutic for FXS.

Disclosures: T. Saraf: None. **B. Yang:** None. **C. Forest:** None. **C.E. Canal:** None.

Poster

522. Muscarinic, Serotonergic, and Metabotropic Glutamate Receptors

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 522.09

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Title: Cannabidiol Mechanisms on 5-HT1A Receptor Desensitization in Neurons

Authors: C. ALEXANDER[1], *M. VASEFI[2];
[1]Biol., [2]Lamar Univ., Beaumont, TX

Abstract: Cannabidiol (CBD) is a popular phytocannabinoid and a potential candidate to treat psychiatric disorders. Multiple studies have suggested that CBD has antidepressant and anxiolytic properties, however, these mechanisms are still debated. Such effects may arise from CBD's interactions with multiple targets, such as 5-HT1A receptors (5-HT1AR) and the survival-regulating ERK1/2 signaling pathway. Clinicians typically prescribe antidepressants such as SSRIs as first-line treatment for psychiatric disorders, however, these drugs have presented minimal effectiveness. On the other hand, CBD has been considered safe and multiple studies have suggested that CBD invokes rapid effects. Thus, the identification of novel mechanisms of CBD is crucial for the treatment of these disorders. SSRI-mediated desensitization of the serotonergic receptors has been documented as part of the antidepressant effect. However, the involvement of CBD in a similar mechanism is still questioned. Like many SSRIs, CBD is also an agonist of the 5-HT1AR, which is vulnerable to desensitization. Interestingly, a recent study in rodent models of neuropathic pain and anxiety revealed that chronic intra-DRN administration of CBD produced anxiolytic effects via reducing 5-HT neuron firing activity likely due to desensitization of 5-HT1A autoreceptors. Thus, we were inspired to investigate, for the first time, the influence of CBD-5-HT1AR desensitization in neurons. The human neuroblastoma cell line, SH-SY5Y, will be dose-dependently and time-dependently treated with CBD. Western blot will be employed to detect the impact of CBD on ERK1/2 and MAPK signaling while qRT-PCR will confirm whether CBD-induced desensitization of 5-HT1AR is associated with the downregulation of 5-HT1AR and even ERK1/2. Finally, agonist-antagonist studies employing CBD, 8-OH-DPAT, and WAY100135 will confirm whether CBD's effects on the ERK1/2 signaling pathway require 5-HT1AR agonism. Data represent the average and standard error of 3-4 independent experiments. Statistical analysis will be applied using GraphPad software to generate a representative bar graph based on current data. All current data have been normalized compared to control. Based on early evidence, CBD evokes a dose-dependent modulation of ERK1/2 signaling. Importantly, studies have suggested that 5-HT1AR internalization may be mediated by interaction with Ras-Raf signaling pathways. Thus, these early results suggest that CBD invokes a non-canonical 5-HT1AR signaling pathway that could later result in receptor internalization. This study will support the utility of CBD in the treatment of various psychiatric disorders.

Disclosures: C. Alexander: None. **M. Vasefi:** None.

Poster

522. Muscarinic, Serotonergic, and Metabotropic Glutamate Receptors

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 522.10

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NIH Grant R01 MH097803

Title: Activity of 5-HT_{2A}Rs in the prefrontal cortex is necessary, but not sufficient, to regulate the head-twitch response of mice to the agonist DOI

Authors: *A. B. OZOLS, J. WEI, J. M. CAMPBELL, S. QIU, A. L. GALLITANO;
Univ. of Arizona Col. of Medicine-Phoenix, Phoenix, AZ

Abstract: Serotonin 2A receptors (5-HT_{2A}Rs) mediate the effects of psychedelic drugs. 5-HT_{2A}R agonists, such as (-)-2,5-dimethoxy-4-iodoamphetamine hydrochloride (DOI), induce head-twitch responses (HTRs) in rodents. However, it is unknown whether the activity of 5-HT_{2A}R expressing neurons is sufficient to produce the HTR in the absence of an agonist. Nor is it known in which brain region 5-HT_{2A}Rs control the HTR. Here, we use an optogenetic approach to examine whether activation of 5-HT_{2A}Rs in the mouse prefrontal cortex (PFC) is sufficient to induce HTRs and if inhibition of these neurons prevents HTRs in mice following DOI administration. We crossed *Htr2a*-Cre mice to cre-dependent optogenetic lines Ai32 (channelrhodopsin) and Ai39 (halorhodopsin) to selectively activate and inhibit (respectively) 5-HT_{2A}R-expressing neurons. Function was validated by whole cell patch clamp recordings performed on cortical slices from Ai32$^{+/-}$;*Htr2a*-Cre$^{+/-}$ and Ai39$^{+/-}$;*Htr2a*-Cre$^{+/-}$ mice, confirming that blue (470nm) and yellow (590nm) light activated and inhibited 5-$HT_{2A}R^{+}$ cells, respectively. Next, fiber optic cannulae were implanted bilaterally into the PFC of male and female Ai32$^{+/-}$;*Htr2a*-Cre$^{+/-}$ vs. Ai32$^{+/-}$;*Htr2a*-Cre$^{-/-}$ and Ai39$^{+/-}$;*Htr2a*-Cre$^{+/-}$ vs. Ai39$^{+/-}$;*Htr2a*-Cre$^{-/-}$ mice. Immediately after administration of either DOI (1mg/kg, i.p.) or vehicle, mice were placed into an automated HTR apparatus and monitored for 30 minutes. All mice were exposed to optogenetic light. For optogenetic stimulation, Ai32^{+} mice received 5 ms pulses of blue light and Ai39^{+} mice received continuous yellow light, for the full recording session. Automated HTRs from the latter 15 min. period were analyzed. Results showed that, when exposed to blue light, there was no difference between DOI-induced HTRs in Ai32 mice that express *Htr2a*-Cre compared to Ai32 mice that do not. In Ai32 mice that received vehicle, there was no difference in HTRs in mice that expressed *Htr2a*-Cre compared with control mice, indicating that optogenetic activation of 5-HT_{2A}R+ cells in the PFC was not sufficient to produce HTRs in the absence of an agonist. In Ai39 expressing mice that lack *Htr2a*-Cre, DOI administration produced a significant increase in HTRs, as expected for wildtype mice. However, in Ai39 mice that expressed *Htr2a*-Cre, DOI failed to significantly increase HTRs in the presence of yellow light, indicating that inhibition of 5-HT_{2A}R+ cells in the PFC prevented DOI from inducing significant HTRs. Together, these findings suggest that activation of 5-HT_{2A}Rs in the PFC is not

sufficient to induce HTRs in the absence of a 5-HT_{2A}R agonist, but is necessary for induction of HTRs by a 5-HT_{2A}R agonist.

Disclosures: **A.B. Ozols:** None. **J. Wei:** None. **J.M. Campbell:** None. **S. Qiu:** None. **A.L. Gallitano:** None.

Poster

522. Muscarinic, Serotonergic, and Metabotropic Glutamate Receptors

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 522.11

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: Indian Institute of Science Education and Research Mohali

Title: Differential regulation of group I metabotropic glutamate receptors (mGluRs) and group I mGluR-mediated AMPA receptor endocytosis by protein interacting with C-kinase 1 (PICK1)

Authors: N. RAMSAKHA, ***P. OJHA**, S. BHATTACHARYYA;
IISER Mohali, Indian Inst. of Sci. Educ. and Research, Mohali, Mohali, India

Abstract: Precise localization and targeting of group I metabotropic glutamate receptors (mGluRs), mGluR1 and mGluR5, to appropriate subcellular and synaptic compartments is important for the proper functioning and activity of these receptors. Dysregulation of group I mGluR signaling is a major contributor to the pathophysiology of various neuropsychiatric disorders like schizophrenia, Fragile X syndrome and autism. Despite this obvious significance, not much is known about the cellular and molecular mechanisms that underlie the trafficking of these receptors in a neuron. Protein interacting with C-kinase 1 (PICK1) is a well-known adaptor protein that interacts with a number of membrane proteins and lipid molecules through its PDZ and BAR domains. It is known to regulate the trafficking of α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptors (AMPARs) and is implicated in various neuropsychiatric disorders. Moreover, group I mGluRs are known to modulate synaptic plasticity by regulating the internalization of AMPARs. In view of the overlapping roles of PICK1 and group I mGluRs in the central nervous system, we hypothesized that PICK1 might regulate group I mGluR trafficking and in turn control the mGluR-mediated AMPA receptor endocytosis.To test this hypothesis, we used dissociated primary hippocampal neurons from P0 C57BL/6 mice of both sexes and used dual antibody labeling to differentially label the surface and internalized receptors for calculating the endocytosis index in each cell. We observed that knockdown of PICK1 inhibited the agonist-mediated endocytosis of only one member of the group I mGluR family, i.e., mGluR1, but had no effect on the internalization of the other member of this family, i.e., mGluR5. Using a “molecular replacement” approach in primary hippocampal neurons, we showed that various regions of PICK1 viz., the N-terminal acidic motif, PDZ domain and BAR domain played crucial roles in the agonist-mediated internalization of mGluR1. Furthermore, PICK1 is important specifically for the mGluR1-mediated AMPAR endocytosis but not for

mGluR5-mediated AMPAR endocytosis because the mGluR1 receptors that could not internalize in the absence of PICK1 stayed as inactive receptors on the membrane and could not initiate downstream extracellular signal-regulated kinase (ERK) signaling. Thus, our findings establish PICK1 as a differential regulator of group I mGluRs and mGluR-mediated AMPAR endocytosis that might be relevant to various neuropsychiatric disorders that report a dysregulation of mGluRs, PICK1 and mGluR-mediated synaptic plasticity.

Disclosures: **N. Ramsakha:** None. **P. Ojha:** None. **S. Bhattacharyya:** None.

Poster

522. Muscarinic, Serotonergic, and Metabotropic Glutamate Receptors

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 522.12

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: Novartis Pharma Research Grant

Title: Analysis of ER retention motifs in the intracellular C-terminal domain of mGluR6

Authors: *I. OGIWARA, A. SHIMOHATA, T. AKAGI, S. USUI, M. KANEDA;
Physiol., Nippon Med. Sch., Tokyo, Japan

Abstract: Metabotropic glutamate receptor 6, mGluR6, is predominantly localized at postsynaptic sites of the retinal ON-bipolar cells, where this receptor recognizes glutamate released from the photoreceptors and contributes to excitability of the ON-bipolar cells via Go-mediated signaling cascades. We have previously shown that the intracellular C-terminal domain (CTD, residues 840-871) of rat mGluR6 is required for receptor cell-surface localization and G-protein coupling, and that this receptor putatively possesses two ER retention motifs (848-arginine-lysine-arginine-850 and 853-lysine-lysine-854) in its CTD. We herein examined whether the ER retention motifs were involved in intracellular trafficking and cell-surface localization of mGluR6 using heterologous expression systems with immunocytochemistry and flow cytochemistry. Surface levels of mGluR6 were significantly diminished by 15- and 16-amino acid deletions at the C-terminus (Δ857-871 and Δ856-871), while substitution of a lysine residue with an alanine residue at the di-lysine ER retention motif in Δ857-871 resumed surface localization. 17 and 18-amino acid deletions at the C-terminus (Δ855-871 and Δ854-871) did not affect mGluR6 surface localization by disrupting the di-lysine motif. Yet again, mGluR6 surface localization was attenuated by 19-and 20-amino acid deletions (Δ853-871 and Δ852-871), while introducing triple-alanine substitution at the arginine-lysine-arginine ER retention motif in Δ852-871 restored surface localization. In the cells co-expressing full-length and surface-deficient deletion constructs, while full-length constructs were clearly present on cell-surface, surface-deficient mutants were still defective on cell-surface localization, even though full-length and surface-deficient constructs in co-immunoprecipitation assays formed heteromeric complexes. These observations suggest that the intracellular CTD of mGluR6 contains the two ER retention

motifs, and that the exposed ER retention motif in aberrant mGluR6 assembly may prevent the protein complex from being located on cell-surface.

Disclosures: I. Ogiwara: None. **A. Shimohata:** None. **T. Akagi:** None. **S. Usui:** None. **M. Kaneda:** None.

Poster

522. Muscarinic, Serotonergic, and Metabotropic Glutamate Receptors

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 522.13

Topic: B.02. Transmitter Receptors and Ligand-Gated Ion Channels

Support: NRF-2020R1A5A1019023
NRF-2022R1A2C1004913
KHIDI-HU21C0071
SNU CDMC 10-10 project
NRF-2021R1F1A1049169

Title: Neddylation is required for presynaptic clustering of mGlu7 and maturation of presynaptic terminals

Authors: *M. KANG[1,2,3], Y. SUH[1,2,3];
[1]Seoul Natl. Univ. Col. of Med., Dept. of Biomed. Sci., Seoul, Korea, Republic of; [2]Seoul Natl. Univ. Col. of Med., Neurosci. Res. Inst., Seoul, Korea, Republic of; [3]Seoul Natl. Univ. Col. of Med., Transplantation Res. Inst., Seoul, Korea, Republic of

Abstract: Neddylation is a posttranslational modification in which NEDD8 is conjugated to a target substrate by cellular processes similar to those involved in ubiquitination. Recent studies have identified PSD-95 and cofilin as substrates for neddylation in the brain and have shown that neddylation modulates the maturation and stability of dendritic spines in developing neurons. However, the precise substrates and functional consequences of neddylation at presynaptic terminals remain elusive. Here, we provide evidence that the mGlu7 receptor is a target of neddylation in heterologous cells and rat primary cultured neurons. We found that mGlu7 neddylation is reduced by agonist treatment and is required for the clustering of mGlu7 in the presynaptic active zone. In addition, we observed that neddylation is not required for the endocytosis of mGlu7, but it facilitates the ubiquitination of mGlu7 and stabilizes mGlu7 protein expression. Finally, we demonstrate that neddylation is necessary for the maturation of excitatory presynaptic terminals, providing a key role for neddylation in synaptic function.

Disclosures: M. Kang: None. **Y. Suh:** None.

Poster

523. Synaptic Plasticity: Spike-Timing, Homeostatic and Other Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 523.01

Topic: B.05. Synaptic Plasticity

Title: Paired motor point and peripheral nerve stimulation to induce STDP on the soleus H-reflex

Authors: *K. FOK[1,2], N. KANEKO[1,2,3,4], S. TAJALI[2], K. MASANI[1,2];
[1]Univ. of Toronto, Toronto, ON, Canada; [2]KITE, Toronto, ON, Canada; [3]Japan Society for the Promotion of Sci., Tokyo, Japan; [4]Univ. of Tokyo, Tokyo, Japan

Abstract: Soleus (SOL) H-reflex is inhibited during standing to provide better control and postural stability in young healthy individuals. However, in individuals with decreased neurological function it is overly excited, which may result in spasticity of the SOL. Reducing the synaptic gain at the Ia-α motoneuron synapse may help to reduce the spasticity and further improve standing performance in individuals with decreased neurological function. Recently, paired associative stimulation (PAS) has been shown to strengthen residual motor pathways via spike timing dependent plasticity (STDP) in the corticospinal tract, but few studies have looked at modulating the spinal reflex pathways and even fewer aiming to inhibit this pathway. Peripheral nerve stimulation (PNS) can be used to send a signal up the Ia-sensory nerve. Also, motor point stimulation (MPS), which electrically stimulates the motor portion of the muscle that is the most sensitive to electrical stimulation, sends a signal up the α motoneuron cell body antidromically. Thus, by using both of PNS and MPS in a PAS protocol, we may be able to induce STDP at the spinal Ia-α motoneuron synapse. Here we aimed to investigate whether MPS and PNS timed to induce STDP would decrease the H-reflex amplitude. To test this, six healthy participants (5M/1F: 26.8 ± 4.1 yrs) received one of the three following conditions on separate days: 1) Inhibition of H-reflex PAS, 2) MPS- or 3) PNS-only. The antidromic signal from MPS was timed to arrive 5ms prior to the orthodromic signal from PNS at the Ia-α motoneuron synapse to inhibit the H-reflex amplitude. For each condition, 200 stimuli were given at a frequency of 0.1Hz. The H- and M-wave recruitment curves of the SOL were measured prior to, immediately after, 30 and 60 minutes after the intervention. H-reflex amplitudes normalized to the corresponding max M-wave amplitudes were then compared across conditions and times using a two-way ANOVA (3 conditions × 4 times). No main effects of condition ($F = 3.035$, $p = 0.093$) or time ($F = 2.437$, $p = 0.105$) were found. Additionally, no interaction between condition and time was found ($F = 0.218$, $p = 0.986$). To test our choice of the timing between MPS and PNS, two additional PAS conditions where MPS preceded PNS by 15ms at the Ia-α motoneuron synapse were tested. No change was found in the H-reflex amplitude. These results suggest that STDP may be insufficient to inhibit the SOL H-reflex. The influence of orthodromic and antidromic activation, pre- or post-synaptic projections onto the Ia-α motoneuron synapse and negotiated spinal neuron equilibrium on this unexpected result was discussed.

Disclosures: **K. Fok:** None. **N. Kaneko:** None. **S. Tajali:** None. **K. Masani:** None.

Poster

523. Synaptic Plasticity: Spike-Timing, Homeostatic and Other Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 523.02

Topic: B.05. Synaptic Plasticity

Support: NIH Grant NS012542
NIH Grant RR00166
NIH Grant NS118781
NSF Grant EEC-1028725

Title: Paired stimulation produces spike-timing dependent plasticity in single neuron responses within primate motor cortex

Authors: *R. YUN[1], J. MISHLER[1], S. I. PERLMUTTER[2], E. E. FETZ[3];
[2]Dept Physiol. & Biophysics, Washington Natl. Primate Res. Ctr., [3]Physiol. and Biophysics, [1]Univ. of Washington, Seattle, WA

Abstract: Previous studies have demonstrated that electrical stimuli can induce targeted spike-timing dependent plasticity (STDP) both *in vitro* and *in vivo*, but these often required long conditioning periods and the induced changes were not consistently seen between connected site pairs. We hypothesized that inducing reliable STDP was difficult because measures of connectivity typically involved macroscopic evoked potentials, whose mechanisms are complex. To test this hypothesis, we delivered paired stimulation to primary motor cortex (M1) of awake primates. To assess the strength of connectivity before and after conditioning we delivered single-pulse stimulation to the presynaptic site and recorded changes in neural responses from the postsynaptic site. Rather than the commonly used cortico-cortical evoked potentials (CCEPs), we used the probability of stimulus-evoked spikes (SESs) at the postsynaptic site as a measure of synaptic strength. During conditioning, single-pulse stimuli were delivered sequentially to pre- and postsynaptic sites with a specific inter-stimulus interval (ISI), ranging from ±0.1 to ±50 ms with sub-millisecond resolution. The probability of SESs showed consistent changes following conditioning that were strongly dependent on the ISI. Negative ISIs resulted in depression of response probability, consistent with classic STDP, but positive ISIs also often resulted in depression. Normalizing the ISIs to the latencies of SESs revealed that potentiation only occurred if the second stimulus was delivered before the SES. Stimuli delivered near or after the SES latency often resulted in depression as strong as with negative ISIs. The inhibitory responses immediately following SESs typically had highly variable changes after conditioning but increased in duration on average during sessions with positive ISI and decreased SES probability. We additionally tracked the CCEPs, which showed changes in size similar to changes in SES probability across all ISIs but with much higher variability. These results demonstrate that the traditional STDP curve may be more difficult to replicate within intact cortical circuitry *in vivo* due to interactions between excitatory and inhibitory networks, and that CCEPs are a less reliable measure of changes in synaptic strength than single unit responses.

Disclosures: R. Yun: None. **J. Mishler:** None. **S.I. Perlmutter:** None. **E.E. Fetz:** None.

Poster

523. Synaptic Plasticity: Spike-Timing, Homeostatic and Other Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 523.03

Topic: B.05. Synaptic Plasticity

Support: NRF-2019M3E5D2A01058328
NRF-2021M3E5D2A01019544

Title: Spike timing-dependent plasticity at feedforward and feedback inhibitory synapses differentially modulate spike synchronization in a feedforward neural network model

Authors: *K. SHIN, J. KWAG;
Dept. of Brain and Cognitive Engin., Korea Univ., Seoul, Korea, Republic of

Abstract: Neural information is believed to be encoded in synchrony of spikes across multiple neurons. Inhibition and spike timing-dependent plasticity (STDP), in which synaptic weight changes based on the temporal relationship of spike timing in presynaptic and postsynaptic neurons both independently have been shown to gate spike synchronization. However, how STDP in inhibitory synapses gate spike synchronization is yet unclear. To address this question, we built a three-layer feedforward network (FFN) model consisting of Hodgkin-Huxley-type excitatory (EX) and inhibitory neuron (IN) models. INs in each layer provided either feedforward inhibition (FFI) or feedback inhibition (FBI) to EXs, which underwent four different types of STDPs: Hebbian, anti-Hebbian, symmetric, and asymmetric STDP. Using these FFN model with FFI (FFN_{FFI}) and FFN model with FBI (FFN_{FBI}), we investigated how different STDP rules in FFI and FBI could differentially synchronize different frequencies of input spikes (5 to 80 Hz) in the final layer of each FFN model by analyzing the correlation coefficient between the spike trains in the input layer and the final layer. In the absence of STDP, FFN_{FFI} could propagate and synchronize input spikes of low frequencies (20, 30 Hz) to the final layer while FFN_{FBI} could propagate and synchronize input spikes of high frequencies (40-80 Hz). However, the four different STDP rules each differentially gated the spike synchrony depending on the input frequency, indicating that the FFI and FBI in the FFN can dynamically gate the propagation of spike synchronization in specific STDP learning rule regimes.

Disclosures: K. Shin: None. **J. Kwag:** None.

Poster

523. Synaptic Plasticity: Spike-Timing, Homeostatic and Other Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 523.04

Topic: B.05. Synaptic Plasticity

Title: Impacts of synaptic plasticity within the cerebellar golgi cell circuit

Authors: *T. RIVERA[1], J. BUSH[2];
[2]Computer Sci., [1]Moravian Univ., Bethlehem, PA

Abstract: Synaptic plasticity is the ability for neurons to make new or stronger connections with other neurons which is thought to be the main mechanism for learning and storing memory in the brain. The cerebellum is responsible for functions such as balance and movement and has been well studied as a powerful experimental paradigm for synaptic plasticity. However, it is not well known what the impacts are for NMDA-driven spike timing dependent plasticity. Golgi cells are the primary inhibitory interneurons in the cerebellum and are responsible for the regulation of activity in the circuit through feed-forward and feedback inhibition. This computational study investigates this main inhibitory circuit within the cerebellum utilizing Hudgen-Huxley models of mossy fiber cells, granule cells, and Golgi cells. Utilizing Python and NEURON programming languages, these computational cell models were developed to be biophysically realistic using spatial, histological, and electrophysiological recordings of actual rat cells. Here, each individual cell type model is connected together to simulate the cerebellar neuronal network. Modulating the precise timing of spikes affects the magnitude of changes in synaptic transmission. I found that the evolution of synaptic weights are likely dependent on the incoming mossy fiber signals along with the number and organization of the Golgi cells. In turn, these Golgi cells dictate the activity and changes in plasticity when implementing NMDA-driven spike timing.

Disclosures: T. Rivera: None.

Poster

523. Synaptic Plasticity: Spike-Timing, Homeostatic and Other Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 523.05

Topic: B.05. Synaptic Plasticity

Support: The Academy of Finland (decision Nos. 318879, 326494, 326495, 345280)
European Union's Horizon 2020 Framework Programme for Research and Innovation under the Specific Grant Agreement 945539 (Human Brain Project SGA3)

Title: Astrocytic modulation of synaptic plasticity: How to integrate biological knowledge, computational modeling, and model sensitivity analysis

Authors: *T. MANNINEN[1], A. SAUDARGIENE[2], M.-L. LINNE[1];
[1]Fac. of Med. and Hlth. Technol., Tampere Univ., Tampere, Finland; [2]Neurosci. Inst., Lithuanian Univ. of Hlth. Sci., Kaunas, Lithuania

Abstract: Astrocytes have been shown to possess important roles in regulating neural development and circuit maturation, for example, by controlling the number of synapses and synaptic connectivity. Recent evidence during postnatal development in various brain areas, such as somatosensory cortex (Min and Nevian, 2012), prefrontal cortex (Petrelli et al. 2020), hippocampus (Falcón-Moya et al., 2020), and basal ganglia (Cavaccini et al. 2020), provides strong support that astrocytes take part in modulation of synaptic plasticity and the activity-dependent change in the strength of the synaptic connections between neurons. This phenomenon seems to be expressed in many forms, over different time scales, and depend on complex biophysical and biochemical mechanisms in the brain. Previously, we have reviewed and analyzed existing computational models for astrocytes and neuron-astrocyte interactions, including astrocytic mechanisms that may participate in synaptic plasticity (Manninen et al., 2018). Based on tens of experimental data and computational modeling publications (see, e.g., Min and Nevian, 2012; Banerjee et al., 2014; Manninen et al., 2018), we have developed a model for spike-timing-dependent long-term depression (t-LTD) in a layer 4 to layer 2/3 synapse in somatosensory cortex during postnatal development (Manninen et al., 2020). Our synapse model consists of an astrocyte in addition to the pre- and postsynaptic neurons. In our model, the glutamate released from the presynaptic neuron activates the postsynaptic neuron, and also part of it is spilled over to the extrasynaptic space. The postsynaptic activation is followed by a release of endocannabinoids that induces an increase in the astrocytic calcium concentration. The increase in astrocytic calcium concentration induces exocytosis of glutamate that can further activate presynaptic N-methyl-D-aspartate receptors (NMDARs) together with the spillover of glutamate from the synaptic cleft. The NMDAR activation coupled with calcineurin signaling influences synapse dynamics. In the present study, we developed a workflow through EBRAINS to describe the data integration and modeling process. In addition, we performed sensitivity analysis of the model parameters (Seppälä et al., 2022) to be able to extend our model to also address the natural spike pattern-induced LTD. The combination of wet-lab experiments, computational modeling, and sensitivity analysis is needed to decipher the roles of spike frequency and astrocytes in synaptic plasticity in developing somatosensory cortex.

Disclosures: **T. Manninen:** None. **A. Saudargiene:** None. **M. Linne:** None.

Poster

523. Synaptic Plasticity: Spike-Timing, Homeostatic and Other Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 523.06

Topic: B.05. Synaptic Plasticity

Support: NIH R01 DC04154
NIH R01 NS072406
NIH R01 NS104926
Simons Collaboration on the Global Brain 543031
Simons Collaboration on the Global Brain 542989

Title: Stdp^2: a synaptic mechanism for meta-learning

Authors: *B. J. BHASIN[1,3], S. JAYABAL[4], J. L. RAYMOND[4], M. S. GOLDMAN[1,2];
[1]Ctr. for Neurosci., [2]Depts. of Neurobiology, Phsiology and Behavior, and Opthamalogy and Vision Sci., UC Davis, Davis, CA; [3]Dept. of Bioengineering, Stanford Univ., Stanford, CA; [4]Dept. of Neurobio., Stanford Univ. Sch. of Med., Stanford, CA

Abstract: Synapses throughout the brain exhibit spike-timing dependent plasticity (STDP) with a wide variety of timing requirements. Recent work suggests that the timing requirements for plasticity at a given set of synapses can themselves be tuned by experience to precisely match behavioral demands. Here, we propose general mechanisms for this new form of synaptic metaplasticity. Specifically, we propose that the same neural spiking statistics that drive STDP can also drive changes in the timing requirements for STDP, a form of metaplasticity we refer to as STDP^2: spike-timing dependent plasticity of spike-timing dependent plasticity. In STDP^2, the timing relationships of pre- and postsynaptic activity drive behaviorally adaptive changes in the parameters of the STDP learning rule. We show a number of distinct ways in which STDP^2 can be implemented, each of which can align the STDP rule with a reliable interval in the spiking statistics, such as a physiological feedback delay. However, for more complex distributions of spike timing, the different STDP^2 metaplasticity rules align the STDP rule to different features of the distribution, namely the median, mode or shape of the distribution. We further show how each of the different STDP^2 metaplasticity rules can be implemented through a plausible biochemical network. By establishing how spike-timing dependent plasticity rules can be tuned through spike-timing dependent metaplasticity rules, our work demonstrates a powerful new range of computational possibilities that may be available to neural circuits. In particular, this form of metaplasticity enables neural circuits to learn associations between inputs that have differences in timing or in characteristic processing delays, solving the temporal credit assignment problem. More generally, our proposal for the experience-dependent tuning of the timing requirements for plasticity provides a mechanism for meta-learning, or “learning to learn”, that is implemented directly at individual synapses.

Disclosures: B.J. Bhasin: None. **S. Jayabal:** None. **J.L. Raymond:** None. **M.S. Goldman:** None.

Poster

523. Synaptic Plasticity: Spike-Timing, Homeostatic and Other Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 523.07

Topic: B.05. Synaptic Plasticity

Support: NIH R01 NS072406
Simons Collaborations on The Global Brain 543031

Title: Adaptive Tuning of Timing Rules for Associative Synaptic Plasticity: a candidate mechanism for meta-learning

Authors: *S. JAYABAL[1], B. J. BHASIN[2,3], M. KOUNGA[1], J. DISANTO[1,4], A. SUVRATHAN[1,5], M. S. GOLDMAN[3], J. L. RAYMOND[1];
[1]Dept. of Neurobio., [2]Dept. of Bioengineering, Stanford Univ. Sch. of Med., Stanford, CA; [3]Ctr. for Neuroscience, and Depts. of Neurobiology, Physiology, and Behavior,, Univ. of California, Davis, Davis, CA; [4]Dept. of Neurol., Univ. of California, San Francisco, San Francisco, CA; [5]Dept. of Neurol. and Neurosurgery, Pediatrics, McGill Univ., Montreal, CA

Abstract: We discovered a new property of synapses that provides a candidate mechanism of meta-learning– the ability to learn how to learn. Previously, we found that the timing requirements for associative long-term depression (LTD) at the parallel fiber (PF)-Purkinje cell synapses in the oculomotor cerebellum precisely compensate for feedback delays in the circuit during oculomotor learning. When there is an oculomotor error, the resulting image motion on the retina is reported by the climbing fiber (CF) input to the cerebellar flocculus at a feedback delay of ~120 ms. Remarkably, associative LTD at floccular PF-Purkinje synapses is selectively induced by the same, 120 ms delay between PF and CF activation. How do the PF-Purkinje synapses "know" what the relevant CF feedback delay is? Here we show that experience tunes the synaptic plasticity rules to the behaviorally relevant feedback delay. In mice dark-reared from birth to eliminate the delayed visual feedback errors carried by the CFs, LTD was induced, not by the normal, 120 ms PF-CF interval, but rather by coincident (0 ms) PF-CF activation. In mice allowed to develop in a normal visual environment and then moved as adults to dark housing, the timing rules for associative plasticity also reverted to the abnormal, coincidence-driven LTD, and the 120 ms PF-CF interval lost its effectiveness. When dark housed mice were moved back to normal housing and visual experience, the timing rule for associative LTD induction also switched back to requiring the behaviorally relevant 120 ms pairing interval, indicating that experience of the feedback delay *in vivo* updates the timing rules driving synaptic plasticity. Adaptive Tuning of the Timing Rules for Associative synaptic Plasticity (ATTRAP) could provide a solution to the temporal credit assignment problem, ensuring that LTD occurs selectively in PF synapses active at the time an error is generated. Accordingly, the learned eye movements of dark-reared mice were delayed by roughly 120 ms, as would be expected if LTD occurred, not in the PF synapses that caused the error, but instead in the PFs active at the later time when the CFs report the error. In adult mice moved from normal to dark housing, and then periodically tested with an oculomotor learning task, the timing of learned eye movements became increasingly delayed over several weeks, and then recovered to normal over several weeks after return to normal housing and visual experience. Together, these findings indicate that matching of synaptic plasticity rules to the behaviorally relevant feedback delay is needed to optimize learning. By implementing this matching, ATTRAP could provide a synaptic mechanism of meta-learning.

Disclosures: S. Jayabal: None. **B.J. Bhasin:** None. **M. Kounga:** None. **J. DiSanto:** None. **A. Suvrathan:** None. **M.S. Goldman:** None. **J.L. Raymond:** None.

Poster

523. Synaptic Plasticity: Spike-Timing, Homeostatic and Other Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 523.08

Topic: B.05. Synaptic Plasticity

Support: NIH grant NS116589
Swiss National Science Foundation grants P2ZHP3-187943 and P500PB-203133
Universidad Nacional de Quilmes
CONICET
The Pew Charitable Trusts

Title: Paradoxical self-sustained dynamics emerge from orchestrated excitatory and inhibitory homeostatic rules

Authors: *S. SOLDADO-MAGRANER[1], M. J. SEAY[1], R. LAJE[2], D. BUONOMANO[1]; [1]Univ. of California Los Angeles, Los Angeles, CA; [2]Univ. Nacional de Quilmes, Bernal, Argentina

Abstract: Self-sustained dynamics maintained through local recurrent connections is of fundamental importance to cortical function. Converging theoretical and experimental evidence indicate that cortical circuits generating self-sustained dynamics operate in an inhibition-stabilized regime. In this regime, unstable positive feedback is held in check by balanced recurrent inhibition. Inhibition-stabilization is increasingly viewed as the default dynamic regime of the cortex and proven to be crucial for many forms of neural computation—such as input amplification, working memory or motor control—but how neural networks self-assemble into this particular dynamic regime remains an open question.

Previous computational models have established that four sets of weights ($W_{E\leftarrow E}$, $W_{E\leftarrow I}$, $W_{I\leftarrow E}$, $W_{I\leftarrow I}$) must obey specific relationships to produce inhibition-stabilized dynamics, but it is not known how the brain can appropriately set the values of all four weight classes in an unsupervised manner to be in the inhibition-stabilized regime. Using numerical simulations and analytical methods we prove that standard homeostatic learning rules are not robust enough to generate inhibition-stabilized dynamics and that their instability is caused by a counterintuitive, yet well described, signature of inhibition-stabilized networks: the paradoxical effect. In contrast, we show that a biologically plausible family of "cross-homeostatic" rules overcome the paradoxical effect and lead to the emergence of stable dynamics. Analytical results confirm that these rules are stable and robust to any set of parameter values. When implemented into a spiking network, cross-homeostatic plasticity autonomously tunes all excitatory and inhibitory weights and leads to the emergence of self-sustained asynchronous activity in the inhibition-stabilized regime.

This work provides the first model of how—beginning from a silent network—self-sustained inhibition-stabilized dynamics can emerge from homeostatic rules governing all four synaptic weight classes in an orchestrated manner. These rules shed new light on how the brain may reach its default dynamic state, and provide a valuable tool to self-assemble artificial neural networks into ideal computational regimes.

Disclosures: **S. Soldado-Magraner:** None. **M.J. Seay:** None. **R. Laje:** None. **D. Buonomano:** None.

Poster

523. Synaptic Plasticity: Spike-Timing, Homeostatic and Other Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 523.09

Topic: B.05. Synaptic Plasticity

Support: NIH Grant MH122023 to SSN
NSF Grant OAC-1730655 to SSN
NHMRC to PS
ARC to PS

Title: Role of NMDA receptors in signal transmission in a model lateral amygdala

Authors: ***B. LATIMER**[1], G. GLICKERT[2], P. SAH[4], S. S. NAIR[3];
[1]Univ. of Missouri, Univ. of Missouri, Columbia, MO; [2]Univ. of Missouri, COLUMBIA, MO; [3]Univ. of Missouri Columbia, Univ. of Missouri, Columbia, MO; [4]Queensland Brain Inst., Brisbane, Australia

Abstract: The Pavlovian paradigm of tone-shock fear conditioning in rodents involves repeated pairing of a conditioned stimulus (CS) tones with an aversive unconditioned stimulus (US) shock that leads to LTP of synapses carrying CS information to the lateral amygdala (LA). This fear conditioning protocol involves the phases of habituation and conditioning. The learning and associated synaptic plasticity in the protocol has been shown to involve N-methyl-D-aspartate (NMDA) receptor-dependent plasticity. Despite this fact being known for more than two decades, the biophysical details related to the involvement of the receptor in such learning remain unclear. Here we use a 4000-cell computational model of the lateral amygdala (LA) to explore the role of NMDA receptors in such learning during CS tones, including in the generation of network activity and in promoting the recruitment of interneurons. The model is constrained using single unit multi-electrode data (up to 30 cells, at 25 kHz), in runs with and without blockade of NMDA receptors. We used state-of-the-art approach to label neurons that are active during CS and US presentations and have channel-rhodopsin ChR2 delivered to axon terminals of CS afferents in LA for subsequent in vitro studies of changes in synaptic efficacies. We then consider the habituation phase with CS tones to investigate the role of intrinsic circuits in signal transmission and in possibly engendering synaptic depression of CS afferents during habituation.

Disclosures: **B. Latimer:** None. **G. Glickert:** None. **P. Sah:** None. **S.S. Nair:** None.

Poster

523. Synaptic Plasticity: Spike-Timing, Homeostatic and Other Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 523.10

Topic: B.05. Synaptic Plasticity

Support: HU20C0233
NRF-2019M3E5D2A01058328
NRF-2021M3E5D2A01019544

Title: Synaptic plasticity in hippocampal cholecystokinin interneuron is impaired in the 5XFAD mouse model of Alzheimer's disease

Authors: *S. OH, J. KWAG;
Dept. of Brain and Cognitive Engin., Korea Univ., Seoul, Korea, Republic of

Abstract: Alzheimer’s disease (AD) is a neurodegenerative characterized by abnormal accumulations of amyloid beta in the hippocampus, leading to progressive decline in memory. Hippocampal CA1 cholecystokinin-positive (CCK) interneurons receive feedforward excitatory inputs from CA3 pyramidal cells (PCs) axons through Schaffer collateral (SC) to gate CA3 input to CA1 PCs, indicating their critical roles in memory processing. However, how SC inputs to CCK interneurons are impaired by amyloidosis to cause memory deficits in AD is yet unknown. To address these questions, we injected an AAV-Dlx-Flex-GFP virus into hippocampal CA1 area of the CCK-Cre and 5XFADxCCK-Cre mice to perform targeted whole-cell voltage-clamp recordings from CCK interneurons in acute hippocampal slices in vitro. Specifically, we measured the changes in paired-pulse ratio (PPR) and short-term plasticity (STP) in SC-evoked excitatory postsynaptic currents (EPSCs) in CCK before and after the induction of the spike timing-dependent long-term potentiation (tLTP) at CA3-CA1 synapse, where tLTP was induced by pairing the presynaptic SC stimulation at 5Hz with CA1 PC postsynaptic spike bursts (4 pulses at 100 Hz) with 10 ms time-window. In the control mice, PPR and STP of EPSCs at the SC-to-CCK synapse showed paired-pulse depression (PPD) and short-term depression (STD) but they were significantly depressed after tLTP induction. However, in 5XFADxCCK-Cre mice, such tLTP-induced decreases in PPD and STD in the control mouse were abolished and no changes in PPD and STD were observed. These results suggest that synaptic dysfunctions at SC-to-CCK synapse may disrupt CCK interneuron-mediated gating of CA3 input to CA1 PC, leading to memory deficits observed in 5XFAD mouse model of AD.

Disclosures: S. Oh: None. **J. Kwag:** None.

Poster

523. Synaptic Plasticity: Spike-Timing, Homeostatic and Other Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 523.11

Topic: B.05. Synaptic Plasticity

Title: Regulation of immediate early gene NPTX2 trafficking by PV circuit and cholinergic modulation in vivo

Authors: *S.-E. ROH, A. DELGADO, P. WORLEY;
Johns Hopkins Med. Institutions, Baltimore, MD

Abstract: NPTX2 (Neuronal Pentraxin 2) is an immediate early gene that is expressed in excitatory neurons and exocytosed onto the excitatory synapses of parvalbumin-interneurons (PV-IN). NPTX2 clusters AMPAR of PV-IN and mediates homeostatic scaling, supporting excitation-inhibition balance. Later NPTX2 is shed from synapses into the cerebrospinal fluid (CSF), where NPTX2 is detectable by ELISA. The CSF NPTX2 is significantly reduced and correlates with cognitive performance in individuals with Alzheimer's disease and schizophrenia. Previous *in vivo* imaging studies established NPTX2-SEP (Super-Ecliptic pHluorin) as a marker of synaptic NPTX2 and found that the trafficking in V1 is reduced on day 1 of monocular deprivation at the same time as the pyramidal to PV-IN connectivity is reduced. This raises a possibility that loss of PV-IN activity can lead to LTD of the excitatory synapse and subsequent NPTX2 shedding via spike-timing-dependent plasticity mechanism. In this study, we employed multiple approaches to modulate the PV-IN activity *in vivo* to investigate the effect of post-synaptic activity on NPTX2 trafficking using two photon microscopy together with chemogenetics and optogenetics. Direct inhibition of PV-IN by hM4d(Gi) resulted in a remarkable reduction of NPTX2-SEP signals, which is regarded as shedding. Optogenetic activation of cholinergic neurons using C1V1 as a natural way to inhibit PV-IN also induced shedding of NPTX2. Administration of low-dose nicotine, known to suppress PV-IN, also downregulated synaptic NPTX2 levels. Conversely, when PV-INs were chemogenetically activated with hM4d(Gq), NPTX2 accumulated in the synapses. This series of observations reveal the regulation of NPTX2 shedding by the level of post-synaptic PV-IN activity.

Disclosures: S. Roh: None. **A. Delgado:** None. **P. Worley:** None.

Poster

523. Synaptic Plasticity: Spike-Timing, Homeostatic and Other Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 523.12

Topic: B.05. Synaptic Plasticity

Support: NIH Grant R35GM142490
NIH Grant R00NS101065
Whitehall Foundation
BrightFocus Foundation

Title: Regulation of protein hunger by DA-WED neurons

Authors: *E. M. PAUL[1], Y. ZHANG[1], A. N. HUSTON[1], S. D. DANIELS[1], Y. XIE[1], M. TABUCHI[2];
[1]Case Western Reserve Univ. Dept. of Neurosciences, Cleveland, OH; [2]Dept. of Neurosciences, Case Western Reserve Univ. Sch. of Med., Cleveland, OH

Abstract: Given the worldwide obesity epidemic, it has been postulated that the body's homeostatic circuits, which regulate hunger, are being overwhelmed by the satisfaction that comes from fats, sugars, and carbohydrates. We previously identified the first neural circuit encoding protein-specific hunger (named DA-WED) in Drosophila. Following protein deprivation, the DA-WED neurons not only promote protein feeding, but also suppress sugar intake, and do so via distinct terminal branches that signal to different downstream neurons (identified as "FB-LAL" neurons for protein feeding and "PLP" neurons for sugar feeding). Remarkably, only the protein-specific branch undergoes intrinsic plastic changes following protein starvation, which drives persistent protein feeding behavior. Under conditions of moderate protein starvation, we found that the DA-WED neurons showed no change in spiking rate but exhibit sub-threshold events in the protein branch. To test the impact of spiking rates on this circuit, we induced spiking ("analog" coding) and performed dual intracellular recordings of the DA-WED protein branch and the FB-LAL cell. To address spiking and non-spiking interactions ("hybrid" coding) in plasticity and hunger, we performed the experiments above, but simultaneously optogenetically increased DA-WED spiking and additionally measured protein branch morphological plasticity. Furthering the research, we are investigating the mechanisms underlying branch-specific analog signaling by harvesting cell bodies and axons of the DA-WED neurons for both single-cell and single- "branch" RNA sequencing. Gaining new insights into how appetite is regulated in the DA-WED neurons in Drosophila will reveal novel targets for addressing human obesity and expand our knowledge of the modulation of motivated behaviors.

Disclosures: E.M. Paul: None. **Y. Zhang:** None. **A.N. Huston:** None. **S.D. Daniels:** None. **Y. Xie:** None. **M. Tabuchi:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.01

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Support: JSPS KAKENHI grant number 16H06544
JSPS KAKENHI grant number 21H05295
JSPS KAKENHI grant number 21K06259

Title: Heterogeneity of Ca^{2+} responses to airflow stimulus in the local non-spiking interneurons of the insect

Authors: *K. SHIRAHATA[1], H. SHIDARA[2,3], H. OGAWA[2];
[1]Grad. Sch. of Life science, [2]Fac. of science, Hokkaido Univ., Sapporo, Japan; [3]Grad. school of Med., Mie Univ., Tsu, Japan

Abstract: Neurons encode information mediated by changes in membrane potentials, which are classified into two types according to their coding scheme. One is a spiking neuron, which uses action potential, and the other is a non-spiking neuron, which uses graded potentials instead of the action potentials as its coding medium. The non-spiking neurons fail to carry the output signals to distal ends of their neurites because the graded potential declines quickly based on electrotonic distance. Therefore, computational function of the non-spiking neurons is possibly based on subcellular local processing mediated by the graded potential unlike spiking neurons. Arthropoda such as insects have many non-spiking neurons in their central nervous system. However, it remains unknown how the non-spiking neurons respond to sensory stimuli in a subcellular scale. In this study, we addressed this question by using wind-sensitive non-spiking neurons identified in the cercal mechanosensory system of the cricket to detect surrounding airflow. The airflow information such as velocity and direction is processed by local circuit in the terminal abdominal ganglion (TAG), where several local spiking and non-spiking neurons sensitive to the airflow have been identified, and conveyed to the brain by ascending projection neurons including identified giant interneurons (GIs). GIs represent distinct selectivity to the airflow direction with the stimulus-evoked spikes. In contrast, the airflow responses in the graded potential of the local non-spiking interneurons (LNIs) vary in directional tuning depending on the recording site of an intracellular electrode. By combining intracellular recording and Ca^{2+} imaging method, we examined both the membrane potential and subcellular Ca^{2+} responses in LNIs to airflow provided from eight different angles. In our present results, all recorded LNIs (n=12) had directional selectivity in their Ca^{2+} response, and some types of LNIs showed Ca^{2+} decrement as well as Ca^{2+} elevation. We especially focused on LNI-9V-2 (n=4), one of the LNIs and analyzed cross-correlation in the response properties of stimulus-induced local Ca^{2+} changes, such as time variation and directional tuning, among all subcellular regions. For the temporal variation, the Ca^{2+} responses in LNI-9V-2 were more highly correlated between the same subcellular regions such as dendrites, axon terminals, and main-neurite, than between those of different regions. For the directional tuning property, the dendrites and axon terminals were different in their tuning curves from each other. These results suggest that LNIs locally process the directional information in different time course.

Disclosures: **K. Shirahata:** None. **H. Shidara:** None. **H. Ogawa:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.02

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Support: NIH Grant DC004274

Title: Shifts in E/I balance and neuronal resonance enhance spike probability in a maturing sound localization macro-circuit

Authors: *A. L. A. DAGOSTIN, H. P. VON GERSDORFF;
Vollum Inst., Oregon Hlth. & Sci. Univ., Portland, OR

Abstract: The mouse auditory brainstem is not fully mature prior to the first month of life. We have studied the neuronal properties and synaptic transmission in the medial nucleus of the trapezoid body (MNTB) and lateral superior olive (LSO) at postnatal day 30-40 (P30 - juvenile) and in young adults (3-6 months old; 3-6 MO). To avoid early age-related hearing loss at high sound frequencies in C57 mice, we used the CBA strain. Early in development, MNTB principal neurons undergo morphological changes which affect membrane passive properties, reducing their input resistance (R_{in}). Our data show differences in the R_{in} for P30 and 3-6MO in both MNTB (144.1±8.7 and 95.29±18.2 MΩ) and LSO principal cells (58.16±4.9 and 39.24±5.5 MΩ). However, the resting membrane potential did not change significantly. Additionally, subthreshold conductances regulate the membrane potential resonance, which is hypothesized to modulate AP firing rate and precision. Using a chirp command wave, we observed resonance characteristics across different membrane potentials and these change with age and tonotopic localization. However, the particular resonant frequency remained nearly unchanged from P30 and 3-6MO for LSO neurons. MNTB and LSO synapses show short term plasticity (STP) after high frequency stimulation (HFS). Synaptic currents depress to a steady state during HFS. In LSO, the depression reduced the EPSC amplitudes to a steady-state of 58 ± 5.5% of the initial amplitude in P30, and 52 ± 5.1% in 3 MO. In the MNTB, EPSCs were reduced to 46 ± 8.0% at P30 compared to only 68.1 ± 3.6% in 3 MO at 100 Hz HFS. STP data also allows us to estimate the readily releasable pool (RRP) size of synaptic vesicles. LSO neurons did not show significant differences in their EPSC RRP size (3.5 and 3.0 nA), but did in their IPSC RRP (3.8 and 1.3 nA). We observed that for adult mice excitation/inhibition (E/I) balance leans towards a more excitable system (P30: E/I = 0.59; 3-6 MO: E/I = 1.28), due to smaller IPSCs in LSO 3-6 MO compared to P30. Finally, IPSPs had a slower kinetic in P30 mice (Tau [ms]: 4.2 in P30; 1.3 in 3-6MO), while EPSPs were similar (Tau [ms]: 1.5 in P30; 1.6 in 3-6MO). These characteristics lead to a reduced ability of juvenile animals to convey information precisely and securely when the nucleus is challenged with intense afferent fiber input. These results point towards a not yet fully developed superior olivary complex at P30 in CBA mice. Although neurotransmission is functional, its temporal characteristics at higher frequencies fail to deliver information fully and precisely to generate action potential spikes, something that is vital for a system that operates within the microsecond realm.

Disclosures: **A.L.A. Dagostin:** None. **H.P. von Gersdorff:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.03

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Support: NIH, NINDS Grant R35NS097212 to GWD

Title: Firing rate homeostasis is controlled by Notch signaling in Drosophila central neurons

Authors: *D. KARMELIC, Y. KULIK, G. W. DAVIS;
Dept. of Biochem. and Biophysics, Kavli Inst. for Fundamental Neurosci., Univ. of California San Francisco, San Francisco, CA

Abstract: Firing Rate Homeostasis (FRH) is a form of homeostatic control that stabilizes neuronal spiking rate and information coding when neurons are confronted by pharmacological, genetic or environmental perturbation (Davis, 2013; O'Leary *et al*., 2014; Kulik *et al*., 2019). FRH has been widely documented within invertebrate neurons (Turrigiano *et al*., 1994; Muraro *et al*., 2008; Driscoll et al., 2013; Kulik *et al*., 2019) and neural circuits (Haedo and Golowasch, 2006) as well as the vertebrate spinal cord (Gonzalez Islas *et al*., 2010), cortical pyramidal neurons (Andrásfalvy *et al*., 2008) and cardiomyocytes (Guo *et al*., 2005; Marrus and Nerbonne 2008; Michael *et al*., 2009). Yet, the underlying molecular mechanisms remain poorly defined. In *Drosophila,* FRH can be induced by the selective deletion or knockdown of the *Shal* potassium channel (Kv4 ortholog). Loss of *Shal* induces selective increases in both the Slo channel (BK channel ortholog) as well as the delayed rectifier ion channel conductance, precisely restoring action potential waveform and firing rates to wild type levels, evidence of a homeostatic signaling system. Here we demonstrate that the canonical Notch signaling pathway is selectively induced by loss of *Shal* in a single identified motoneuron. Notch signaling was implicated in FRH following transcriptional profiling of motoneurons comparing control and *Shal* mutants, following by confirmation using single-cell patch-seq. Taking advantage of available genetic tools, we demonstrate that loss of *Notch* and the downstream transcriptional regulator *Su(H)* both prevent the induction of homeostatic signaling without altering baseline neuronal firing rates. The *Drosophila* orthologue of Presenilin, which cleaves and releases the Notch intracellular domain, is also required for FRH. An ongoing analysis of ionic conductance changes is consistent with activation of Notch signaling being essential for the homeostatic modulation of the Slo channel conductance. No changes in neuronal cell fate or morphology are detected. We propose a model consistent with an essential, post-embryonic function of canonical Notch signaling during FRH, with potential implications for changes in neuronal excitability that are co-morbid with numerous neurodegenerative and psychiatric disorders in human including Alzheimer's disease.

Disclosures: D. Karmelic: None. **Y. Kulik:** None. **G.W. Davis:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.04

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Support: NIH grant R01NS093866
NIH grant R35NS127219

Title: Multiple spikes in dentate granule cells generated by intrinsic and network mechanisms and modified by experience

Authors: *W.-C. SHU[1], M. B. JACKSON[2];
[1]UW-Madison, Madison, WI; [2]Univ. of Wisconsin-Madison, Univ. of Wisconsin-Madison, Madison, WI

Abstract: The dentate gyrus receives synaptic inputs from the entorhinal cortex and processes this information before transmitting to the CA3 region. The principal cells of the dentate gyrus, granule cells (GCs), exhibit weak excitability, possibly reflecting sparse coding, but fire in bursts during distinct behaviors. These bursts may reflect the activity of their afferents, but GCs can also generate multiple spikes with a single stimulation. We explored multiple spiking in GCs with a hybrid voltage sensor (hVoS 1.5) targeted by Cre-drivers. In mature GCs targeted with a Prox1 Cre-driver, a single stimulation of cortical inputs often elicited two spikes separated by intervals of a few milliseconds (doublets), and rarely (incidence<3%) more than two spikes. The incidence of doublets initially increased with the stimulus current but declined with further increases; the decline may reflect the recruitment of GABAergic inhibition. This was confirmed by showing that GABA receptor blockade increased the doublet incidence nearly 2-fold. Thus, recurrent inhibition limits the generation of multiple spikes. To investigate the mechanisms by which multiple spikes are generated, we imaged voltage in GC soma and dendrites. The first spike generally initiated in the middle molecular layer. The second spike, in some cases, initiated in dendrites located in the inner molecular layer. This suggests that mossy cell (MC) projections play a role in GC repetitive firing. However, in other cases, the second spike initiated close to the soma, suggesting an intrinsic mechanism. As an additional test of the role of GC-MC recurrent excitation, we utilized two pharmacological agents to block the two excitatory synapses of this circuit. This reversed the increase in doublet incidence following GABA receptor blockade. Intrinsic excitability may contribute to multiple spiking in GCs because some GCs still fire doublets after blocking recurrent excitation. However, blockade of low-threshold calcium current, known to modulate GC bursting, only prolonged the doublet interval without altering their incidence. We also investigated multiple spikes in GCs targeted by c-fos expression in mice exposed to a novel environment. The doublet incidence of novelty-activated GCs was significantly higher than in control GCs targeted with Prox1, suggesting that multiple-spiking behavior may serve a role in encoding novel aspects of the environment. These results suggest that multiple spiking in GCs reflects both intrinsic excitability and network properties in the dentate gyrus. One or both of these mechanisms may contribute to the increased firing of GCs resulting from experience.

Disclosures: W. Shu: None. **M.B. Jackson:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.05

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Support: NIH R21EB029740
HFSP RGY0069
ORAU Ralph E. Powe Junior Faculty Enhancement Award

Title: The mechanisms underlying spike timing and rate under multiplexed dendritic input streams

Authors: *S. XIAO, S. YADAV, K. JAYANT;
Purdue Univ., Purdue Univ., West Lafayette, IN

Abstract: Synaptic and dendritic integration across the vast expanse of the basal dendritic arbors of a pyramidal neuron is central for coincidence detection, feature binding, and dendritic branch specific plasticity. Central to enabling these properties is the ability of the cell to maintain spike precision and rate control while integrating widespread synaptic inputs across multiple dendrites. The biophysical mechanisms that give rise to such precision and rate control, however, remains poorly understood. Our limited understanding of both intra- and inter-branch integration stems partly from technical barriers in manipulating synaptic properties with high resolution, and, monitoring the effects of these manipulations across dendritic depth. Here, to address this challenge we performed simultaneous electrophysiology and two-photon glutamate uncaging across distributed and clustered synapses located along multiple basal dendrites of cortical pyramidal neurons in acute brain slices using a spatial light modulator (SLM) with custom-built pulse compression. The SLM allows simultaneous activation of multiple spines in 3D, across basal arborizations with near-diffraction-limited resolution. We found that co-activation of clustered spines on some but not all basal dendritic branches gives rise to a complex dendritic spike, characterized with a Na^+ spikelet followed by a NMDA plateau. Compared to NMDA spikes without a Na^+ spikelet, these complex Na^+ dendritic spikes increase the temporal precision of somatic action potentials. Furthermore, when paired with depolarization caused by a distributed synaptic inputs background, we observe that this temporal precision is accompanied by burst rate modulation, with spike frequencies reaching ~200Hz in the form of doublets and triplets. We show that this precision-rate control is made possible by a combination of axo-somatic persistent Na^+ current amplification (via afterdepolarization and the increased input resistance) and stochastic ion-channel gating. Moreover, this temporal precision is preserved under an *in-vivo* like high conductance state, showing that noisy background activity does not saturate dendrites but in fact improves spike-probability, including from synapses that were previously too weak to elicit a spike. Together, our results suggest that multiplexed synaptic integration across multiple basal dendritic branches enables distinct modes of axo-somatic amplification to enable precise clock generation with concomitant gain control.

Disclosures: S. Xiao: None. **S. Yadav:** None. **K. Jayant:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.06

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Support: NIH 1R56NS094831
NIH 1UF1NS115821-01

Title: Intrinsic rebound excitability of basal ganglia projecting neurons reflect learned features of zebra finch song

Authors: *N. D. MEDINA[1], D. MARGOLIASH[2];
[1]The Univ. of Chicago, Chicago, IL; [2]Organismal Biol. and Anat., Univ. of Chicago Dept. of Organismal Biol. and Anat., Chicago, IL

Abstract: Plasticity of neuronal excitability is a fundamental mechanism underlying learning and memory that refers to the signal processing that occurs within neurons after synaptic transmission. The study of plasticity in the intrinsic properties (IPs) of neurons is unveiling mechanisms beyond synaptic plasticity that relate network activity and learning. Prior work in songbirds established a relationship between the IPs of forebrain neurons and the learned song of zebra finches. Within the premotor nucleus HVC, the IPs of basal-ganglia-projecting neurons (HVCx) are developmentally regulated and differ across individual birds. Importantly, HVC_X from birds who sing similar songs also express similar IPs. Given those results, we examined the role of song learning in regulating HVC_X IPs. We evaluated HVCx in vitro firing properties in relation to the bird's song, and found that the song, not parentage, had the stronger effect on HVCx IPs. While examining the relation between IPs and learned song, we discovered a correlation between timing features of song and the rebound excitation of HVCx: Neurons from birds who sang longer songs with long harmonic stacks had a combination of IPs that reflected increased rebound excitation. Given that harmonic stacks are spectrally unchanging over their duration, this result suggests a mechanism underlying HVCx neurons' documented ability to integrate over long periods of time. We then used an instrumental learning paradigm to generate two groups of birds who sang very similar songs, except one group was tutored with an added long harmonic stack. We found that the group who sang the longer modified song had increased evoked sag and firing frequency. Finally, we used these results, in vivo extracellular recordings, and established work to constrain a Hodgkin-Huxley-based network model of HVC that captures in vivo bursting properties and serves as a hypothesis linking neuronal IPs to network structure and behavior. Our results demonstrate an explicit link between neuronal IPs and features of learned behavior and accentuate the importance of including neuronal IPs in developing realistic network-level descriptions of neural circuits.

Disclosures: N.D. Medina: None. **D. Margoliash:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.07

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Title: Modification of the information processing in granule cells depending on the local network

Authors: *N. NAKAJIMA[1], T. KAMIJO[2], H. HAYAKAWA[1], E. SUGISAKI[1], T. AIHARA[1]; [1]Tamagawa Univ., Tokyo, Japan; [2]Univ. of the Ryukyus, Okinawa, Japan

Abstract: The medial dendrite (MD) of granule cells (GCs) receives spatial information through the medial perforant path (MPP). The distal dendrite (DD) of GCs receives non-spatial information (sensory inputs) through the lateral perforant path (LPP). However, it is not clear how information processing though two pass ways are performed in the GCs. In this study, associative the information processing of inputs for MD and DD were reported. First, in physiological experiment, to investigate the differences in response characteristics between MD and DD using rat hippocampal slices, electrical stimuli of ten pulses at 10-40 Hz were applied MPP or LPP, respectively. In our experiments, NMDA receptor and GABAergic receptor as post synaptic factor were blocked by application of D-AP5 (NMDA receptor antagonist) and picrotoxin ($GABA_A$ receptor antagonist). As the result, excitatory post-synaptic potential (EPSPs) for successive input pulses in MD were transiently decreased as the number of inputs. On the other hand, EPSPs in DD showed sustained responses to 10-30 Hz. However, EPSPs in DD did not show sustained responses to 40 Hz. Next, using NEURON simulator of multi-compartment GC (Ferrante et al. 2009) with the dynamic synapses model (Tsodyks et al. 1988), we performed a computational experiment. The dynamic synapses model was fixed by parameter fitting for our physiological experimental data. Theta burst (8 Hz) and random pulse inputs (10-40 Hz) were applied to MD and DD, respectively. As the result, the temporal-pattern sensitivity for burst inputs to MD was shifted depending on the frequency (20-40 Hz) of random pulse inputs applied to DD. On the other hand, the existence of cue cells (non-spatial information) in addition to better characterized place cells (spatial information) in the dentate gyrus was recently reported (Tuncdemir SN et al. 2022). It showed that the response of place cells was blocked by lateral inhibition from cue cells. Therefore, we performed the second experiment using a model with lateral inhibition to block responses of place cells by cue cells. As the result, the temporal pattern sensitivity for burst input to MD was enhanced depending on the frequency of random pulse inputs to DD. Our result suggests that the information processing of GCs for temporal pattern inputs was modified depending on the non-spatial information in the network of dentate gyrus.

Disclosures: **N. Nakajima:** None. **T. Kamijo:** None. **H. Hayakawa:** None. **E. Sugisaki:** None. **T. Aihara:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.08

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Title: Input multiplexing through cell-intrinsic regulation of spike synchrony in the dorsal raphe nucleus

Authors: M. B. LYNN[1], L. MALER[2], ***J.-C. BEIQUE**[2];
[1]Cell. and Mol. Med., [2]Cell. & Mol. Med., Univ. of Ottawa, Ottawa, ON, Canada

Abstract: The serotonergic dorsal raphe nucleus (DRN) receives diverse long-range synaptic inputs, yet the relative contribution of each input to DRN output spiking patterns is unknown. Here, we use electrophysiological, optogenetic and computational tools to compare functional features of excitatory inputs from lateral habenula (LHb) and prefrontal cortex (PFC) onto DRN 5-HT neurons. Dual-color opsin strategies revealed that single 5-HT neurons receive functionally matched input from both PFC and LHb. Subthreshold features of excitatory post-synaptic potentials, including amplitude and decay, were largely overlapping. However, PFC inputs triggered spikes that displayed significantly higher latency and greater jitter than those from LHb. A support vector machine classifier demonstrated that input identity can be accurately decoded from the spike timing of under ten 5-HT neurons, revealing that these timing differences can be robustly parsed by downstream circuits. By examining the intrinsic cellular mechanisms in 5-HT neurons underlying these transformations of EPSPs to spikes, we uncovered a prominent T-type calcium conductance which selectively boosts certain input types, as well as subthreshold, voltage-dependent membrane noise which calibrates spike jitter and latency. Together, these results outline a mechanism by which intrinsic properties of 5-HT neurons functionally segregate LHb and PFC inputs into distinct spiking patterns which could, we hypothesize, be decoded by downstream areas innervated by 5-HT axons - a multiplexing operation.

Disclosures: M.B. Lynn: None. **L. Maler:** None. **J. Beique:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.09

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Title: Serotonin release dynamics in the raphe support emergent value computations

Authors: ***M. LYNN**[1], S. D. GEDDES[1], M. CHAHROUR[1], S. MAILLÉ[2], E. HARKIN[2], E. HARVEY-GIRARD[3], S. HAJ-DAHMANE[4], R. NAUD[1], J.-C. BEIQUE[2];

[2]Cell. and Mol. Med., [3]Cell. and Mol. Medecine, [1]Univ. of Ottawa, Ottawa, ON, Canada; [4]Dept. of Pharmacol. and Toxicology, Univ. of Buffalo, Buffalo, NY

Abstract: Serotonin (5-HT) neurons in the the dorsal raphe nucleus (DRN) receive a diverse constellation of long-range synaptic inputs, yet it is largely unknown how the statistics of these inputs interact with local connectivity motifs and synaptic dynamics within DRN to shape 5-HT output and ultimately behavioral state. Here, we combined optogenetic, electrophysiological, computational and behavioral strategies to identify how the inputs from the lateral habenula (LHb) are locally processed in the DRN to acutely calibrate learned reward predictions. Stimulating LHb afferents to 5-HT neurons triggered a frequency-dependent mixture of fast, monosynaptic excitation and slow inhibition mediated by 5-HT_{1A} receptors. Optogenetic and pharmacological manipulations in DRN demonstrated that 5-HT neurons are organized in a recurrent inhibitory network, accounting for the protracted effects of LHb inputs and refuting the classical autocrine transmission model. At these recurrent connections, 5-HT release is probabilistic, dominated by robust short-term facilitation, and can be captured with a linear-nonlinear plasticity model. Network simulations revealed that these short-term recurrent dynamics generate excitation-driven inhibition from strong habenular inputs, and submodule-specific winner-take-all dynamics which emerge over behavioral timescales. We employed an auditory classical conditioning paradigm in mice to test quantitative model predictions, finding that activating LHb input to DRN suppressed goal-directed anticipatory licking behavior contextually during high reward value states and for high input frequencies, yet did not affect motor output or the learned association itself. We suggest that recurrent 5-HT release dynamics in DRN support an integration of learned associations, internal state and acute stimuli to flexibly adapt behavioural state to dynamic environments.

Disclosures: M. Lynn: None. **S.D. Geddes:** None. **M. Chahrour:** None. **S. Maillé:** None. **E. Harkin:** None. **E. Harvey-Girard:** None. **S. Haj-Dahmane:** None. **R. Naud:** None. **J. Beique:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.10

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Support: Hungarian Artificial Intelligence National Laboratory (RRF-2.3.1-21-2022-00004)

Title: Reliable estimation of neuronal biophysical parameters from whole-cell recordings using model simulations and probabilistic inference

Authors: D. TERBE[1], M. SZOBOSZLAY[1,2], Z. NUSSER[1], ***S. KALI**[1];
[1]Inst. of Exptl. Med., Budapest, Hungary; [2]Dept. of Neurosci., Columbia Univ., New York, NY

Abstract: Biological systems often have properties that are difficult to measure directly but may be inferred from available data using appropriate mathematical tools. As an important example, the electrophysiological behavior of neurons depends on their morphology and biophysical parameters, and one common approach to estimating these biophysical parameters has relied on varying the parameters in a morphologically detailed neuronal model until the behavior of the model best approximates the experimental recordings. Such methods have yielded some of the currently accepted values for biophysical parameters such as axial resistivity and have also been used to argue for a non-uniform distribution of membrane conductances in several types of neuron. However, the precision and reliability of these estimates have not been analyzed, and the effects of measurement noise on the results have not been investigated.
In this study, we combined methods from probabilistic inference with simulations of detailed neuronal models to examine these issues and applied them to morphological and electrophysiological data from CA1 pyramidal neurons to obtain reliable estimates of key biophysical parameters. We first analyzed the noise in our whole-cell recordings, and found that it was characterized by significant temporal correlations at various time scales. Investigation of the effects of noise correlations on the results of parameter inference using simulated data indicated that assuming an incorrect noise model introduced bias into and increased the variance of the estimated biophysical parameters. This is important also because the common method of fitting parameters by minimizing the mean squared error is equivalent to assuming additive Gaussian white noise during inference and is therefore also expected to yield incorrect results. Our methods also allowed us to compare the expected efficacy of different experimental protocols in determining the target parameters and predicted that dendritic whole-cell recordings using a combination of current steps of different length would be suitable to determine axial resistivity. We therefore carried out such recordings in CA1 pyramidal neurons and performed simultaneous inference for three biophysical parameters. Our method predicted with high confidence that the axial resistivity of CA1 pyramidal neurons was severalfold lower than previous estimates based on least-squares fits. A similar analysis suggested that it was not possible to reliably determine whether the distribution of the leak conductance was spatially uniform in the cell using recordings at a single (somatic or dendritic) site.

Disclosures: **D. Terbe:** None. **M. Szoboszlay:** None. **Z. Nusser:** None. **S. Kali:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.11

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Support: NSF-IOS-2002863
NSF-CRCNS-DMS-1608077

Title: Interaction of segregated mechanisms of resonance in CA1 pyramidal neurons: interplay of ionic currents and the spatial structure of the cell

Authors: *H. ROTSTEIN[1], U. CHIALVA[2];
[1]Federated Dept. of Biol. Sci., NJIT & Rutgers Univ., Newark, NJ; [2]Univ. Nacional del Sur and CONICET, Bahia Blanca, Argentina

Abstract: CA1-pyramidal neurons are known to have two spatially segregated resonance mechanisms due to the non-uniform distribution of two different voltage-gated channels: the perisomatic M-current and the dendritic h-current (Hu et al., J. Neurosci. 2009). These play different roles regarding the control of excitability, spike regulation, processing of rhythmic inputs and neuronal integration of CA1 neurons. The two mechanisms have been studied both theoretically and experimentally in vitro, focusing on the properties of those responses that emerge at different polarization levels at which the two resonant currents activate. However, these studies focus on the two resonant mechanisms independently, and do not take into account the possible interactions between them and the interactions with the additional inputs coming from sorrounding cells. In particular, it is not clear how the resonance interactions are affected by the presence of voltage heterogeneities across the cell such as those expected to be present under realistic conditions due to inhibitory inputs from PV+ (proximal) and OLM (distal) interneurons in addition to background noise. We address these issues by using biophysical (conductance-based) modeling and computational simulations. We show that the h- and M-current-based segregated resonant mechanisms can interact at the subthreshold level due to significant voltage differences along the cell membrane. As result, they can generate filtering regimens and resonant profiles that different from the ones produced by the individual mechanisms, and are not uniform along the neuron. Futhermore, we distinguish between strong and weak interactions of the two segregated mechanisms, which give rise to responses with greater or lesser variability, respectively, throughout the neuron. We then describe how this response variability affects the neuron's synaptic integration properties. Finally, by measuring the somatic summation of synaptic-like inputs applied at the distal end of the neuron for a wide range of input frequencies, we were able to establish the contribution of each current to the overall response. We find that the interaction of resonant mechanisms may play a role in regulating the temporal integration of synaptic inputs, by increasing the range of frequencies for which the summation value is negative. Together these results demonstrate that the filtering properties of neurons are flexible and depend not only on ionic currents or voltage regime, but also on the (nonuniform) distribution and spatial segregation of currents, also therefore provide a mechanism that regulates the frequency-processing of synaptic inputs.

Disclosures: H. Rotstein: None. **U. Chialva:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.12

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Support: NIH R35NS097343
NIH R01MH046742

Title: Biological Structural Coupling Enhances Neuronal Robustness to "Noise" Perturbations

Authors: *Y. ZANG, E. MARDER;
Biol., Brandeis Univ., Waltham, MA

Abstract: Despite cell-to-cell variability, biological neurons still have the striking ability to maintain their key firing properties in the face of unpredictable perturbations and stochastic noise. However, the biological strategies that neurons use to reliably maintain their firing properties remain poorly understood. By using a population of compartment models for the lateral pyloric (LP) neuron in the crab stomatogastric ganglion, we explored how the critical pattern of rebound bursting is preserved when the 14 channel conductances in each model undergo random variations. Simulation results suggest that the rebound bursting pattern can be well maintained in many of the neuron models with different ranges of variations, which argues the existence of manifolds for bursting neurons in the 14-dimension parameter space to make this property insensitive to conductance variations. The rebound bursting pattern is more insensitive to channel conductance variations in the soma than in the axon. The degree of soma-axon coupling is critical to the ability of the axon to spike during bursts and consequently determines the size of the manifold that corresponds to the bursting models in the whole parameter space. When the soma-axon coupling deviates from the biological range, the neuronal tolerance of conductance variation is significantly lessened. This work suggests that neurons can still find general strategies to enhance their functional reliability, even when they exhibit significant individual variability.

Disclosures: Y. Zang: None. **E. Marder:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.13

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Title: Oscillatory stimuli produce nonstationary spiking activity in computer model of neocortical pyramidal neuron: spike-phase advance and retreat

Authors: *C. KELLEY[1], J. L. KUBIE[3], S. D. ANTIC[4], W. W. LYTTON[2];
[2]Physiology/Pharmacology, [1]SUNY Downstate Hlth. Sci. Univ., Brooklyn, NY; [3]Cell Biol., SUNY Downstate Med. Ctr., Brooklyn, NY; [4]Neurosci., UConn Hlth., Farmington, CT

Abstract: Impedance phase characterizes the temporal relationship between subthreshold membrane potential oscillations and the stimuli which elicit them. However, impedance cannot characterize suprathreshold neuronal spiking properties because spiking neurons violate the

assumptions of stationarity and linearity. Instead, we employed a *suprathreshold impedance phase framework* for spiking neurons by registering spikes to the phase of a sinusoidal input. We used a model of neocortical layer 5b pyramidal neuron with subthreshold impedance properties we previously found to be in agreement with experimental data. We identified two (A and B) distinct time-dependent relationships between oscillatory stimuli and spiking, depending on whether the sinusoidal input current was of constant or increasing amplitude. A. Constant amplitude: the stimulus phase of each spike advanced over time, initially preceding the peak of each stimulus cycle and eventually following the peak. This occurred for specific frequency-amplitude combinations at different stimulus locations on the cell. For an 8 Hz perisomatic current stimulus of 1.0 nA amplitude spike-phase advanced ~45^{o} over six stimulus cycles. The spike-phase relationship was nearly constant for lower frequency stimuli of the same amplitude and more complicated for higher frequencies. This phase advance was eliminated by blocking Ih (HCN). B. Increasing amplitude: the stimulus phase of each spike decreased over time (retreated), initially following the peak of each stimulus cycle and eventually preceding the peak. Responses were similarly dependent on stimulus frequency, amplitude, and location in the dendritic tree. For a perisomatic stimulus of 8 Hz whose amplitude increased by 1 nA/s, spike-phase retreated by ~60^{o} over 6 stimulus cycles. There was less spike-phase retreat for lower frequency stimuli of the same amplitude, and the spike-phase profile was more complicated for higher frequencies. The range of stimulus phases swept by the spiking activity was shifted upward by blocking Ih. These cellular responses may play a role in network phenomena studied in hippocampal pyramidal neurons where the phase of spiking changes relative to theta oscillations: theta-phase precession and phase "roll" (phase advance over time).

Disclosures: C. Kelley: None. **J.L. Kubie:** None. **S.D. Antic:** None. **W.W. Lytton:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.14

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Support: National Science Foundation grant DMS-CRCNS 1608077
NSF-IOS 2002863

Title: Intrinsic ionic dynamics, oscillations, and resonance are reflected in and can be extracted from neuronal spike-train cross-correlations

Authors: *R. D. PENA[1], M. V. IBARRA[2], H. G. ROTSTEIN[1];
[1]Federated Dept. of Biol. Sci., New Jersey Inst. of Technol. and Rutgers Univ., Newark, NJ;
[2]Univ. Nacional de la Patagonia San Juan Bosco & CONICET, San Juan Bosco, Argentina

Abstract: There has been an increased interest in extracting information about the structure of neuronal networks in different cognitive states from the newly developed multiarray recordings

of the brain. A sharp peak near zero in spike-train cross-correlation functions (CCFs) is consistent with the presence of a monosynaptic connection between a pre- and post-synaptic neuron (Toyama et al., J Neurophysiol 1981; Platkiewicz et al., J Comput Neurosci 2021). Spike-train relationships have been used to infer synaptic connectivity, by using these peaks as a proxy for synaptic strength (English et al., Neuron 2017). Changes in the peak's height under certain appropriate controlled conditions have been interpreted as synaptic plasticity, involved in cognitive processes such as learning and memory (English et al., Neuron 2017, McKenzie et al., Neuron 2021). However, CCFs are complex and contain significantly more information about the spike train relationships between the participating neurons. Some of this information is apparent from the spiking patterns themselves, but spiking patterns are controlled by the neuronal subthreshold (membrane potential) dynamics whose effects often remain hidden. Identifying this type of information is expected to contribute to the inference of synaptic connectivity strengths and synaptic plasticity from spike-train relationships. Here, we address this issue by combining biophysical modeling, numerical simulations, dynamical systems tools (phase-space analysis), and artificial neural networks (ANNs). We show that in the presence of certain combinations of ionic currents (e.g., resonant and amplifying), secondary peaks emerge in CCFs, away from the peak near zero, in addition to and confounded with these resulting from external factors such as background oscillations, or ripples. We then investigate the circumstances under which the ionic currents' features (conductances and time-constants) can be extracted with ANNs. We identify which of the attributes that describe the CCF's shape are relevant to allow for an efficient classification exploiting the fact that the timing of the first postsynaptic spike relative to a presynaptic spike has a strong dependence on the intrinsic dynamics of the postsynaptic cell in addition to the background. This has been overlooked in earlier modeling studies. Our results have implications for the inference of intrinsic cellular and synaptic properties (e.g. resonances) and the detection of synaptic plasticity from spike-train relationships since "first spikes" (after each presynaptic spike) may be spread out beyond the sharp-peak lags commonly attributed to monosynaptic connectivity.

Disclosures: **R.D. Pena:** None. **M.V. Ibarra:** None. **H.G. Rotstein:** None.

Poster

524. Intrinsic Properties

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 524.15

Topic: B.06. Intrinsic Membrane Properties, Electrical Synapses, and Signal Integration

Support: NIH Grant NS011613
NIH Grant 1R43NS125749

Title: Determining the effects of ion channels on spiking behavior by interfacing a real-time dynamic clamp system with computational models of ion channels simulated with NEURON

Authors: M. W. NOWAK[1], L. KORBEL[1], A. KANE[1], B. PANAMA[1,2], M. L. HINES[3], *N. T. CARNEVALE[3], G. C. L. BETT[2,1], R. L. RASMUSSON[2,1];
[1]Cytocybernetics, Pendleton, NY; [2]SUNY Buffalo, Buffalo, NY; [3]Neurosci., Yale Univ., New Haven, CT

Abstract: For detailed investigation of how ion channels affect neuronal spiking behavior, we interfaced the NEURON simulator (www.neuron.yale.edu) with a real-time, high-speed dynamic clamp system. This allowed us to implement "virtual channel expression," in which experimentally observed membrane potential is used to calculate the current that is generated by computational models of ion channels and injected back into the cell (Synaptic Cell Mode configuration, see diagram). In these experiments, the "cell" subjected to virtual channel expression was a real-time simulation of a model Purkinje neuron with seven voltage-gated ion channels developed by Akemann and Knopfel (2006). The channels that were virtually-expressed in this model were drawn from the neuroscience literature. Addition of IK_A and IK_{DR} (from Sheets et al., 2007) reduced AP firing frequency (Control: 48±0 Hz, IK_A: 16±1 Hz, IK_{DR}: 20±1 Hz; n=4), but IK_A affected after-hyperpolarization potential (AHP) magnitude more than IK_{DR} did (Control: 2.8±0.1 mV, IK_A: 13±0.2 mV, IK_{DR}: 9.9±0.1 mV; n=4). Addition of the sodium channel NaV 1.8 (from Han et al., 2014) increased AP firing frequency (Control: 48±0 Hz, NaV 1.8: 60±1 Hz; n=4), increased AHP magnitude (Control: 2.8±0.1 mV, NaV 1.8: 8.3±0.2 mV; n=4) and increased AP width at half-height (Control: 0.7±0.1 ms, NaV 1.8: 1.6±0.1 ms; n=4). These simulations demonstrate the feasibility and applications of combining NEURON AP simulations with a real-time dynamic clamp system for cell-based electrophysiology research. Through the use of computational models of ion channels and neurons, the effects of subtle changes in ion channel properties on AP behavior can be examined in detail. Instead of “virtual ion channel current expression”, real ion channel currents expressed in living cells can be input into the NEURON models. Drug assays targeting the expressed ion channels can be developed in which the measurable endpoints are neuronal function and AP behavior. These studies were supported, in part, by NIH NS011613 and 1R43NS125749.

Disclosures: **M.W. Nowak:** None. **L. Korbel:** None. **A. Kane:** None. **B. Panama:** None. **M.L. Hines:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Cytocybernetics. **N.T. Carnevale:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Cytocybernetics. **G.C.L. Bett:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cytocybernetics. **R.L. Rasmusson:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cytocybernetics.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.01

Topic: B.08. Epilepsy

Title: Use of Calling Cards to record antecedent super-enhancer binding events that alter susceptibility to seizures in mice

Authors: *M. A. GACHECHILADZE[1], B. D. BOROS[1], A. J. CAMMACK[2], M. SHABSOVICH[1], A. YEN[1], T. M. MILLER[1], R. D. MITRA[1], J. D. DOUGHERTY[1]; [1]Washington Univ. Sch. of Med., Saint Louis, MO; [2]Univ. Col. London, London, United Kingdom

Abstract: Transcription factors (TFs) achieve precise patterns of gene expression that define a wide variety of functional states in living systems by binding to specific regulatory elements in the genome. While many current methods are available to profile this binding, they are limited to endpoint views - they are capable of describing the current state of a system, but cannot describe how that state came to be. Calling Cards technology overcomes this limitation using a TF-transposase fusion and a transposon reporter to leave permanent records of TF binding events, allowing us to integrate TF binding over time and connect the binding events to a later phenotype of interest. Susceptibility to GABA antagonist pentylenetetrazol (PTZ)-induced seizures provides a proof-of-concept of this scenario. Preliminary work has demonstrated that genetically identical juvenile C57BL/6 mice housed in similar conditions experience seizures of highly variable severity in response to the same dose of PTZ. If there are individual pre-existing epigenetic differences in the mice that alter their later susceptibility to PTZ-induced seizures, the Calling Cards system should be able to detect them. Using a Syn1-Cre driven transposase with a natural affinity for Brd4, which targets super-enhancers, pilot data has detected differential antecedent super-enhancer usage that may predict which mice go on to have mild and severe seizure phenotypes.

Disclosures: **M.A. Gachechiladze:** None. **B.D. Boros:** None. **A.J. Cammack:** None. **M. Shabsovich:** None. **A. Yen:** None. **T.M. Miller:** F. Consulting Fees (e.g., advisory boards); Ionis, Biogen, Disarm Therapeutics, Cytokinetics, UCB. Other; C2N, licensing agreement, Bioio, LLC, Regeneron, honorarium, Denali, honorarium. **R.D. Mitra:** None. **J.D. Dougherty:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.02

Topic: B.08. Epilepsy

Support: CONACYT PhD grant #489736

Title: P-glycoprotein is overexpressed in amygdala and hippocampus during electrical amygdala kindling process

Authors: *D. FONSECA-BARRIENDOS[1], J. CASTAÑEDA-CABRAL[2], F. MARTÍNEZ-CUEVAS[1], W. G. BESIO[3], S. OROZCO-SUAREZ[4], A. VALDÉS-CRUZ[5], L. ROCHA[1]; [1]Ctr. de Investigación y de Estudios Avanzados, Ciudad de Mexico, Mexico; [2]Univ. de Guadalajara, Zapopan, Mexico; [3]Col. of Engin., Univ. of Rhode Island, East Greenwich, RI; [4]Speciality Hosp, Mexican Inst. Social Sec, Mexico DF, Mexico; [5]Inst. Nacional de Psiquiatría Ramón de la Fuente Muñiz, Ciudad de Mexico, Mexico

Abstract: Purpose: P-glycoprotein (P-gp) is an ATP-dependent drug efflux pump that acts as a xenobiotic extruder and its overexpression is associated to drug resistant epilepsy. Moreover, P-gp overexpression is detected after an acute seizure and has been associated to neuronal hyperexcitability. Therefore, we aim to determine if P-gp expression is progressively increased through epileptogenesis induced by electrical amygdala kindling. Methods: Male Wistar rats (300-350g) previously implanted in right basolateral amygdala were used (n=5 per group). Briefly, kindled group was stimulated daily until 3 consecutive stage V seizures were induced. Stage III group was stimulated until one stage III seizure was elicited. Stage I group in which only one discharge was evoked. Sham group was implanted but did not receive any electrical stimulation. Naïve animals weren't implanted nor stimulated. Amygdala and hippocampi (ipsi- and contralateral) were collected 24 h after the last electrical stimulation or manipulation. P-gp expression was evaluated by Western blot. Results: Stage I group showed P-gp overexpression in ipsilateral hippocampus (86%, $p<0.001$ vs naïve). Stage III group presented P-gp overexpression in ipsilateral amygdala (77.79%, $p<0.05$ vs naïve), and both hippocampi (58.95%, $p<0.05$ and 57.66%, $p<0.05$ vs naïve, ipsi and contralateral respectively). Kindled group had P-gp overexpression in ipsilateral amygdala (89.95%, $p<0.01$ vs naïve) and both hippocampi (92.20%, $p<0.001$ and 91.73%, $p<0.01$ vs naïve, ipsi and contralateral respectively). Sham group presented P-gp overexpression in the ipsilateral hippocampus (98%, $p<0.001$ vs naïve) Conclusion: Progression of electrical amygdala kindling is associated with increased P-gp expression in amygdala and hippocampus. These changes are structure and stage dependent. P-gp overexpression induced by kindling could be associated with progressive neuronal hyperexcitability and epileptogenesis.

Disclosures: D. Fonseca-Barriendos: None. **J. Castañeda-Cabral:** None. **F. Martínez-Cuevas:** None. **W.G. Besio:** None. **S. Orozco-Suarez:** None. **A. Valdés-Cruz:** None. **L. Rocha:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.03

Topic: B.08. Epilepsy

Support: Grant to RJB from the Camden Health Research Initiative of Rowan University

Title: Somatic mutations in human temporal lobe epilepsy

Authors: *R. J. BUONO[1,2,3], M. R. SPERLING[2], Y. LIU[3], C. N. VACCARO[3], R. PELLIGRINO DA SILVA[3], T. N. FERRARO[1], H. HAKONARSON[3];
[1]Biomed. Sci., Cooper Med. Sch. of Rowan Univ., Camden, NJ; [2]Comprehensive Epilepsy Ctr., Thomas Jefferson Univ. Hosp., Philadelphia, PA; [3]Ctr. for Applied Genomics, The Children's Hosp. of Philadelphia, Philadelphia, PA

Abstract: Rationale: Genetic influences on human epilepsy are complex. Most work done to identify gene mutations in patients analyze gene structure in DNA derived from peripheral blood. We hypothesize that somatic mutations exist in the brain cells of most individuals and that specific allelic variations influence both susceptibility and resistance to epilepsy and other brain diseases. Thus, somatic cell mutations may contribute to the cause of epilepsy in humans.
Methods: We studied 5 patients with temporal lobe epilepsy (TLE) using whole exome sequencing (WES) at an average of more than100X depth of coverage. We extracted DNA from both peripheral blood and from a single region of the resected temporal neocortex (Brodmann area 38). We then performed WES and compared results between blood sequences and brain sequences in each patient. We used an established bioinformatics pipeline to identify mutations in brain that are not found in blood. We distinguished mutations into 2 classes: “Rank A”, which are nonsense somatic mutations (stop-gain, frameshift deletion/insertion etc.) and “Rank B” which are non-synonymous mutations. We defined mutations as having a minimum minor allele frequency (MAF) of greater than one percent based on read number and depth of coverage.
Results: Data analysis revealed that each patient harbored thousands of somatic mutations. In total, there were 4010 unique genes that contain at least one Rank A somatic mutation, with 191 of these genes represented at an MAF greater than five percent. We identified 8388 unique genes with Rank B mutations. The 5 patients had 20 genes with Rank A somatic mutations in common including: CCDC30, HMCN1, OR2T2, TTN, XIRP1, FRAS1, MAML3, TACC2, MKI67, ACIN1, RBM25, SLTM, BRD7, HNF1B, FHOD3, DPP9-AS1, MUC16, NRIP1, LOC150051, and CLTCL. Altogether, the patients had 54 genes with Rank B somatic mutations in common.
Conclusion: Somatic mutations occur in the brain cells of patients with TLE and may contribute to the development of disease symptoms. Among the 20 genes with somatic mutations that we identified in the patient cohort, two are associated with epilepsy (BRD7 and CLTCL). Somatic mutations occur in brain cells in many people, and specific allelic variations may influence both susceptibility and resistance to epilepsy and other brain diseases. We are currently expanding the number of blood-brain pairs and the number of brain regions, including additional areas of temporal neocortex as well as hippocampus and amygdala. Convergence of somatic gene mutations and single cell or bulk tissue gene expression profiles will help to identify new brain cell-localized, genetic causes of epilepsy.

Disclosures: R.J. Buono: None. **M.R. Sperling:** None. **Y. Liu:** None. **C.N. Vaccaro:** None. **R. Pelligrino Da Silva:** None. **T.N. Ferraro:** None. **H. Hakonarson:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.04

Topic: B.08. Epilepsy

Support: Brain & Behavior Research Foundation Young Investigator Grant
U01MH108898
R01MH124890
R21AG070462
R01NS083823

Title: Comprehensive multiomic profiling of somatic mutations in malformations of cortical development

Authors: C. CHUNG, X. YANG, J. G. GLEESON;
Univ. of California San Diego, La Jolla, CA

Abstract: Malformations of cortical development (MCD) are characterized by focal disruption of cortical architecture and cellular organization arising during cortical development, largely from somatic mosaic mutations. Identifying the genetic causes of MCD has been a challenge, as mutations remain at low allelic fractions in brain tissue resected to treat epilepsy. Here, we show a genetic atlas from 317 brain resections, identifying 69 mutated genes through intensive profiling of somatic mutations, combining whole-exome and targeted-amplicon sequencing with functional validation and single-cell sequencing. Genotype-phenotype correlation analysis elucidated specific MCD gene sets correlating distinct pathophysiological and clinical phenotypes. The unique spatiotemporal expression patterns identified by comparing single-nucleus transcriptional sequences of mutated genes in control and patient brains implicate critical roles in excitatory neurogenic pools during brain development, and in promoting neuronal hyperexcitability after birth.

Disclosures: C. Chung: None. **X. Yang:** None. **J.G. Gleeson:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.05

Topic: B.08. Epilepsy

Support: SFI Grant 16/RC/3948
SFI Grant 18/SIRG/5646

Title: Initial profiling of RNA methylation (m6A) in human temporal lobe epilepsy reveals prominent hypermethylation of coding transcripts

Authors: *M.-M. WILSON[1,3], G. P. BRENNAN[5], D. C. HENSHALL[1,3], S. BYRNE[2,6,3], A. SANZ-RODRIGUEZ[1,3], N. DELANTY[7,4,3], E. T. DILLON[5], D. O'BRIEN[7], M. FARREL[8], K. LAU E HOW[1,3];
[1]Physiol. and Med. Physics, [2]Paediatrics, Royal Col. of Surgeons In Ireland, Dublin, Ireland; [3]FutureNeuro, [4]Royal Col. of Surgeons in Ireland, Dublin, Ireland; [5]Sch. of Biomolecular and Biomed. Sci., Univ. Col. Dublin, Dublin, Ireland; [6]Children's Hlth. Ireland at Crumlin, Dublin, Ireland; [7]Dept. of Neurology, Beaumont Hosp., [8]Dept. of Neurosci., Beaumont Hosp., Dublin, Ireland

Abstract: Introduction: The underlying pathomechanisms of epilepsy are likely driven by abundant and diverse changes in transcriptional control that results in aberrant gene readout and altered protein levels. This includes altered epigenetic, transcriptional and post-transcriptional gene readout. However, there may be additional layers of gene regulation in epilepsy which remain unexplored to date. Methylation of the sixth position of adenosine residues in RNA results in N6-methyladenosine (m6A) which is the most common internal covalent modification in mRNA and is associated with altered RNA stability, translational efficiency, and sub-cellular localisation. Preliminary data from our lab shows that in the epileptic mouse brain, global increases in m6A levels are seen, however the mechanisms which govern m6A and the levels of m6A itself remain unknown in human TLE. We hypothesise that RNA methylation represents an important mechanism of post-transcriptional regulation which significantly contributes to the aberrant gene expression and protein production involved in the development and maintenance of hyperexcitable networks in epilepsy. Methods: We combine m6A micro-array, qPCR, and proteomics to probe the status and effects of m6A deposition in human hippocampal tissue at the level of the transcriptome, genome, and proteome. Further, in vitro characterisation of neuronal structure and function in response to loss of m6A is carried out in iPSC derived neuron cultures. Results: Western blot analysis revealed selective dysregulation of m6A-associated enzymes. Transcriptome-wide mapping of m6A in TLE uncovered significant restructuring of the m6A-epitranscriptome in TLE Vs control tissue. Similarly proteomic analysis confirmed significant changes in the epileptic proteome Vs control. Conclusion: By integrating the results of these analyses, we identify m6A-modulated gene networks and biochemical pathways that may be involved in the development and/or maintenance of TLE. This project has explored a potentially novel layer of gene dysregulation in human TLE which provides further insight into disease mechanisms and may illuminate novel pathways for therapeutic intervention.

Disclosures: M. Wilson: None. **G.P. Brennan:** None. **D.C. Henshall:** None. **S. Byrne:** None. **A. Sanz-Rodriguez:** None. **N. Delanty:** None. **E.T. Dillon:** None. **D. O'Brien:** None. **M. Farrel:** None. **K. Lau E How:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.06

Topic: B.08. Epilepsy

Support: SFI Grant 16/RC/3948
European Regional Development Fund
FutureNeuro industry partners

Title: Chromatin landscape mapping reveals bi-directional changes to gene accessibility in human temporal lobe epilepsy

Authors: *K. CONBOY[1,2], G. P. BRENNAN[3,2], N. NGUYEN[1,2], A. SANZ-RODRIGUEZ[1,2], N. DELANTY[4,2], D. O'BRIEN[4], R. GWINN[5], D. C. HENSHALL[1,2];
[1]Dept. of Physiol. and Med. Physics, [2]FutureNeuro, the SFI Res. Ctr. for Chronic and Rare Neurolog. Dis., Royal Col. of Surgeons Ireland, Dublin, Ireland; [3]Sch. of Biomolecular and Biomed. Sci., Univ. Col. Dublin, Dublin, Ireland; [4]Dept. of Neurol., Beaumont Hosp., Dublin, Ireland; [5]Ctr. for Neuromodulation and Functional Restoration, Swedish Neurosci. Inst., Seattle, WA

Abstract: Temporal lobe epilepsy (TLE) is the most common form of epilepsy in adults and is associated with drug resistance. Surgical resection of the epileptic focus is often the only possibility for seizure-relief, with the pathological hallmark of hippocampal sclerosis observed to varying degrees. TLE is characterised by global changes in gene expression and regulatory networks. Gene regulatory patterns are controlled by the activity of transcription factors (TFs) that function by interpreting and altering the chromatin landscape. Chromatin is dynamic and its accessibility can be regulated by a host of mechanisms including chemical modifications on both DNA and histone proteins. While chromatin accessibility has been explored for single loci, there has been no global attempt to map the chromatin landscape in human TLE. This could lead to improved understanding of how dynamic changes in our epigenome establish or maintain seizure susceptibility or drug-resistance in epilepsy patients. The assay of transposase accessible chromatin sequencing (ATAC-seq) was employed in order to profile the accessible chromatin landscape in resected human hippocampus from patients with drug resistant epilepsy with different grades of sclerosis. RNA-seq from the same cohorts was performed and incorporated to make inferences about regulatory elements that are potentially regulating the dysregulated transcriptomic landscape. Epigenetic marks and TFs were further mapped in order to determine their localisation within the genome and to elucidate their function in regulation following ATAC-RNA-seq analysis. ATAC-seq experiments resulted in the identification of 1000s of differentially accessible regions of chromatin between epilepsy cases and controls, in contrast with few differences in accessibility between epilepsy cases with different grades of sclerosis. Differential accessible chromatin regions were predominantly found within regulatory enhancer and promoter regions. Gene ontology of sites with increased and decreased accessibility in hippocampal sclerosis TLE versus control suggests that these regulatory regions are associated with metabolic pathways and synaptic transmission. Differentially accessible chromatin in non-hippocampal sclerosis versus control showed enrichment for terms involved in cellular

development and organisation. Here, we characterised genome-wide chromatin accessibility in the resected hippocampi of individuals with TLE and linked this to the differential accessibility on the transcriptome. This analysis provides insight into the structural basis of molecular mechanisms contributing to human TLE.

Disclosures: K. Conboy: None. **G.P. Brennan:** None. **N. Nguyen:** None. **A. Sanz-Rodriguez:** None. **N. Delanty:** None. **D. O'Brien:** None. **R. Gwinn:** None. **D.C. Henshall:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.07

Topic: B.08. Epilepsy

Support: DFG Grant DO 2542/1-1
"Research commission" of the Medical Faculty, University of Freiburg
DON1207/19

Title: Focal cortical dysplasia type-dependent dysregulation of myelination in extratemporal lobe region of the human neocortex

Authors: *C. DONKELS[1], J. M. NAKAGAWA[2], F. RAUE[1], S. HUBER[1], A. VLACHOS[5], C. SCHEIWE[2], M. J. SHAH[2], A. SCHULZE-BONHAGE[3], M. PRINZ[4], J. BECK[2], C. A. HAAS[1,6]; [1]Exptl. Epilepsy Research, Dept. of Neurosurg., [2]Dept. of Neurosurg., [3]Epilepsy Ctr., [4]Inst. of Neuropathology, Univ. Med. Ctr. of Freiburg, Fac. of Med., Freiburg, Germany; [5]Dept. of Neuroanatomy, Inst. of Anat. and Cell Biol., [6]BrainLinks-BrainTools Ctr., Univ. of Freiburg, Freiburg, Germany

Abstract: Focal cortical dysplasias (FCDs) are local malformations of the human neocortex and a leading cause of pharmacoresistant epilepsy. FCDs are characterized by local architectural disturbances of the neocortex and often feature a blurred gray-white matter boundary indicating abnormal white matter myelination. We have recently shown that myelination is also compromised in the gray matter of FCD type IIa in the temporal lobe. Here, we aimed at analyzing the myelination pattern in FCD type IIa and IIb by focusing on extratemporal regions of the human brain. We characterized the gray matter-associated myelination pathology of FCD IIa, FCD IIb and epileptic control specimens derived from curative epilepsy surgery on several levels. First, we performed a morphological analysis by applying immunohistochemistry to visualize myelinated fibers by confocal microscopy and ultrastructural analyses of the myelinated axons. Next, *in situ* hybridization histochemistry for myelin basic protein (MBP) was performed to analyze the density of myelinating oligodendrocytes (OLs). Finally, we investigated the myelination pattern in FCD on the molecular level by applying real-time RT-qPCR to determine the expression levels of myelin-associated transcripts and chromatin immunoprecipitation to analyze the binding capacity of the transcription factor myelin regulatory

factor (MYRF) to its target promoter MBP. We show that the proportion of myelinated gray matter is similar in FCD IIa, IIb and controls with myelinated fibers extending up to cortical layer II. The myelinated fibers, however, appear fractured and distorted in FCD IIa. Electron microscopy revealed that the myelin sheaths of layer V/VI axons are thinner in FCD IIa and IIb specimens than in controls indicated by an increased g-ratio. In addition, the density of MBP-expressing OLs was reduced in FCD IIa, however, contrarily, we observed an increase of OLs in FCD IIb. Similarly, the expression levels of myelin-associated transcripts were decreased in FCD IIa but increased in FCD IIb. Finally, we detected a reduced binding capacity of MYRF at the MBP promoter. Our results indicate that FCD IIa and IIb have different myelination profiles and reveal opposing myelination deficits.

Disclosures: **C. Donkels:** None. **J.M. Nakagawa:** None. **F. Raue:** None. **S. Huber:** None. **A. Vlachos:** None. **C. Scheiwe:** None. **M.J. Shah:** None. **A. Schulze-Bonhage:** None. **M. Prinz:** None. **J. Beck:** None. **C.A. Haas:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.08

Topic: B.08. Epilepsy

Support: AES Pre-doctoral Fellowship
Medical Scientist Training Program Grant: T32 GM140935
University of Wisconsin Madison MSTP Department of Radiology Fellowship
NIH Grant R01NS123378
NIH Grant R01NS105646
NIH Grant R01NS111022
NIH Grant P50HD105353

Title: Area Deprivation Index and its Association with White Matter Connectome Abnormalities in Temporal Lobe Epilepsy

Authors: *D. Y. CHU[1], N. ADLURU[2], V. A. NAIR[3], A. ADLURU[3], K. DABBS[4], J. MATHIS[7], A. NENCKA[8], B. HERMANN[5], A. L. ALEXANDER[6], A. F. STRUCK[5], J. R. BINDER[7], M. E. MEYERAND[6], V. PRABHAKARAN[4];
[1]Radiology, Univ. of Wisconsin Sch. of Med. and Publ. Hlth., Madison, WI; [2]Waisman Ctr., [4]Radiology, [5]Neurol., [6]Med. Physics, [3]Univ. of Wisconsin Madison, Madison, WI; [7]Neurol., [8]Radiology, Med. Col. of Wisconsin, Milwaukee, WI

Abstract: An increasing body of literature has revealed that social determinants of health including socioeconomic status and neighborhood disadvantage immensely impacts epilepsy prevalence and outcomes. To further assess this, our study aims to characterize the relationship of the US census-based neighborhood disadvantage metric, the Area Deprivation Index (ADI),

and its association with white-matter connectome findings of participants from the Epilepsy Connectome Project (ECP). Multi-shell connectome diffusion weighted MRI (ms-dMRI) data from 124 subjects ages 18 to 60 were employed, including 85 TLE patients (34 male, mean age = 39.28 ± 11.71 years) and 39 healthy controls (HCs) (20 male, mean age = 34.87 ± 10.20 years). The IIT Destrieux gray matter atlas was utilized to create the 162 x 162 structural connectivity matrices using MRTrix3. ComBat data harmonization was applied to harmonize the structural connectivity matrices from pre- and post-scanner upgrade acquisitions and threshold free network-based statistics (TFNBS) was used for statistical analysis of these matrices. These findings were correlated with ADI metrics, which contains measures of income, education, employment, and housing quality. ADI is grouped by quintiles with lower quintiles representing least deprived (least disadvantaged) and upper quintiles representing most deprived (most disadvantaged). Our results reveal a much higher number of significant abnormal connections in the direction where the upper quintiles of ADI scores (more disadvantaged) TLE patients, exhibit lower cross-sectional area of white matter tracts when compared with TLE patients of the lower quintiles of ADI scores (least disadvantaged) and both upper and lower quintile ADI scores of HCs. Moreover, TLE patients in the lower quintile ADI also showed a higher significant number of abnormal connections where their cross-sectional area is lower than that of HCs of both upper and lower quintiles of ADI scores. This demonstrates that neighborhood and other socioeconomic disadvantages may have a strong impact on the white matter integrity of TLE patients. Overall, our study illustrated discrete significant white matter tract abnormalities in the most disadvantaged TLE group when compared to other groups. Understanding the relationship of geospatial metric of neighborhood disadvantages and its impact on epilepsy may inform proper care, treatment delivery, and policy for underserved and disadvantaged patients.

Disclosures: **D.Y. Chu:** None. **N. Adluru:** None. **V.A. Nair:** None. **A. Adluru:** None. **K. Dabbs:** None. **J. Mathis:** None. **A. Nencka:** None. **B. Hermann:** None. **A.L. Alexander:** None. **A.F. Struck:** None. **J.R. Binder:** None. **M.E. Meyerand:** None. **V. Prabhakaran:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.09

Topic: B.08. Epilepsy

Support: NIH Grant U01NS117839
NIH Grant R01MH110831

Title: Characterizing autonomic dysregulation by Inter-Ictal Epileptiform Discharges with intracranial recordings in the Human Brain

Authors: M. GESSESSE[1,3], C. P. MOSHER[3], A. N. MAMELAK[3], C. M. REED[4], *U. RUTISHAUSER[3,2];

[1]Computation and Neural Systems, Caltech, Pasadena, CA; [2]Computation and Neural Systems, Caltech, Los Angeles, CA; [3]Neurosurg., [4]Neurol., Cedars-Sinai Med. Ctr., Los Angeles, CA

Abstract: In addition to seizures, patients with epilepsy often exhibit pathological brief electrical discharges called Interictal Epileptiform Discharges (IED) during the quiescent period between seizures. Although IEDs are common in patients with epilepsy, their role in disrupting normal cognition, as well as in causing or preventing seizures, remains little understood. A key signature of both seizures and IEDs is concomitant autonomic dysregulation, with changes in heart-rate variability around a subset of IEDs commonly seen in both humans and animals. However, the reason for why IEDs can result in autonomic dysregulation remains unknown. We examined the relationship between IEDs and the autonomic nervous system using intracranial single-neuron and local field potential recordings in human patients implanted with hybrid depth electrodes for seizure monitoring. We recorded cardiac data (2 lead electrocardiogram) and neural data (single neuron activity and EEGs scored for IED occurrence) recorded simultaneously from these patients. To assess autonomic function, we developed a toolkit of features including, among other metrics, time-frequency analysis of the inter beat interval (IBI) series. We analyzed these cardiac metrics as well as neural activity aligned to the onset of IEDs to identify potential relationships between neural activity and autonomic responses. Motivated by prior work that shows that modulation of low frequency power of the cardiac waveform during a heartbeat can be used as a metric of a change in sympathetic tone, we used the metric of low frequency-high frequency power ratio of the cardiac waveform to assess sympathetic tone and the IBI to assess heart rate. Using n=13 patients, and averaging over all identified hippocampal IEDs, the low frequency-high frequency power ratio ($p=0.0492$; $t=2.12$), and the IBI ($p=0.02$; $t=-2.54$) were significantly modulated by the occurrence of hippocampal IEDs. This result indicates that the heart rate and the sympathetic tone of the patients in the 30 seconds after the IED is, on average, greater than in the 30 seconds before the IED. Additionally, we have observed that the activity of single neurons recorded in the same brain area as the IEDs is modulated around the time of the occurrence of IEDs, thereby reproducing prior findings. The extent to which a given IED modulates neurons varies widely, however. We are therefore now investigating whether the extent of single-neuron modulation is indicative of the degree of autonomic modulation. Together, these new findings shed light on how an epileptic region of the brain that generates IEDs influences both cardiac as well as single neuron activity.

Disclosures: **M. Gessesse:** None. **C.P. Mosher:** None. **A.N. Mamelak:** None. **C.M. Reed:** None. **U. Rutishauser:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.10

Topic: B.08. Epilepsy
American Epilepsy Society Predoctoral Fellowship

Title: Deciphering axon dysfunction in the pathogenesis of ARHGEF9 epileptic encephalopathy

Authors: *W. WANG[1], C. D. MAKINSON[2], W. N. FRANKEL[1];
[1]Dept. of Genet. and Dev't, [2]Dept. of Neurol., Inst. for Genomic Medicine, Columbia Univ. Irving Med. Ctr., New York, NY

Abstract: *ARHGEF9* is an X-linked gene that is associated with developmental and epileptic encephalopathy (DEE) and intellectual disability (ID). *ARHGEF9* encodes the brain-specific protein collybistin (Cb), a guanine nucleotide exchange factor, and more well known as an essential regulator of inhibitory postsynaptic density by binding to gephyrin and to the α2 subunit of type A GABA receptors ($GABA_AR$-α2). Among patients with mutations in *ARHGEF9*, the reported clinical phenotypes vary, depending in part on the region of the protein affected. Missense mutations in the C terminal PH domain do not cause epilepsy while mutations in DH and SH3 domain lead to early-onset epilepsy along with moderate to severe ID.
We investigated the pathogenesis of *ARHGEF9* DEE by studying a missense variant (G55A), the only missense mutation in the SH3 domain identified in severe DEE. Utilizing a novel *Arhgef9*-G55A mouse model, we examined behavioral, cellular and physiological consequences of *Arhgef9*^{G55A} in mice (*Arhgef9*$^{G55A/Y}$). *Arhgef9*$^{G55A/Y}$ mice exhibited spontaneous generalized tonic-clonic seizures and spike-wave discharges, and increased acoustic startle response, recapitulating key phenotypes observed in *ARHGEF9* patients. Immunostaining studies showed that in *Arhgef9*$^{G55A/Y}$ neurons, Cb formed elongated protein aggregates at the axon initial segment (AIS) of hippocampal pyramidal neurons colocalized with gephyrin, leading to a significant decrease in Cb and gephyrin puncta at the AIS and in dendritic compartments. Furthermore, *Arhgef9*$^{G55A/Y}$ neurons had decreased $GABA_AR$-α2 puncta, especially at the AIS, indicating a disruption in GABAergic synaptic function. Using acute brain slice electrophysiology to examine intrinsic neuronal excitability and synaptic transmission, we observed altered frequency and amplitude in miniature postsynaptic currents (mIPSCs and mEPSCs) in *Arhgef9*$^{G55A/Y}$ neurons. More interestingly, with phase plane analysis of action potentials recorded with somatic current clamp, we observed that *Arhgef9*$^{G55A/Y}$ neurons exhibited altered action potential initiation kinetics, pointing to structural and functional disruptions at the AIS. Together, these results identify the possible pathomechanism of *ARHGEF9* DEE and reveal the underappreciated role of *ARHGEF9* at the AIS.

Disclosures: W. Wang: None. **C.D. Makinson:** None. **W.N. Frankel:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

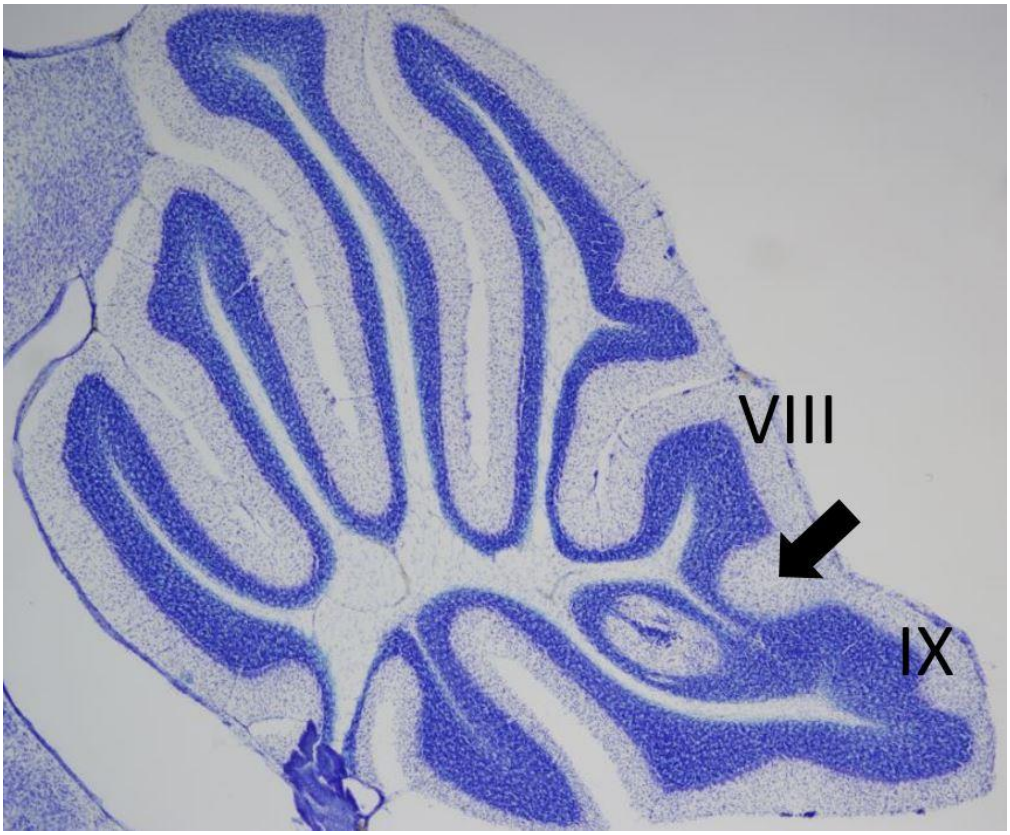
Program #/Poster #: 525.11

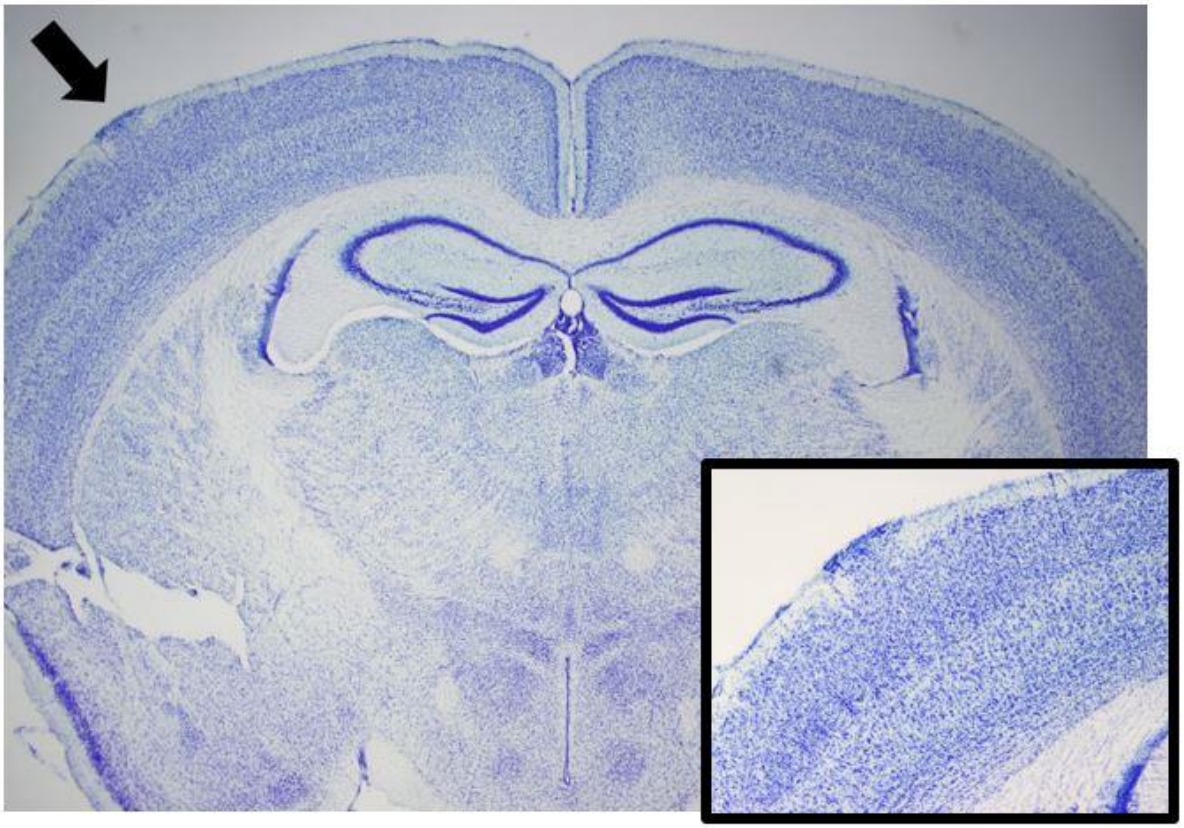
Topic: B.08. Epilepsy

Title: Neocortical and cerebellar malformations affect seizure onset in C57BL/6 mice

Authors: K. M. KEEVER[1], P. D. WOMBLE[2], D. G. SULLENS[2], Y. LI[1], J. N. LUGO[2], *R. L. RAMOS[3];
[1]NYIT Col. of Osteo. Med., Old Westbury, NY; [2]Baylor Univ., Waco, TX; [3]Biomed. Sci., New York Inst. of Technol., Old Westbury, NY

Abstract: Brain malformations cause cognitive disability and seizures in both humans and animal models. Highly laminated structures such as the neocortex and cerebellum, in particular, are vulnerable to malformation affecting lamination and neuronal connectivity as well as causing heterotopia. The objective of the present study was to determine if sporadic neocortical and/or cerebellar malformations in C57BL/6J mice are correlated with reduced seizure threshold. The inhaled chemo-convulsant flurothyl was used to induce seizures in male and female C57BL/6J mice, and the time to seizure onset was recorded as a functional correlate of brain excitability. Following seizures, mice were euthanized and brains extracted for histology. Cryosections of the neocortex and cerebellar vermis were stained and examined for the presence of molecular layer heterotopia as previously describe in C57BL/6J mice (Ramos RL, et al. Dev Neurosci. 2014 ;36(6):477-89; Ramos RL, et al. Neuroscience. 2015; 310:242-51). Over 60% of mice had neocortical and/or cerebellar heterotopia. No sex differences were observed in the prevalence of malformations. Significantly reduced seizure onset time was observed dependent on sex and the type of malformation present. These results raise important questions regarding the presence of malformations in C57BL/6J mice used in the study of brain development, epilepsy, and many other diseases of the nervous system.





Disclosures: **K.M. Keever:** None. **P.D. Womble:** None. **D.G. Sullens:** None. **Y. Li:** None. **J.N. Lugo:** None. **R.L. Ramos:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.12

Topic: B.08. Epilepsy

Support: NINDS NS088776

Title: Early-life seizures and neonatal vocalizations in mice

Authors: ***J. M. REINHART**, J. N. LUGO, Jr., K. J. BLANDIN, D. SANTANA-COELHO; Psychology and Neurosci., Baylor Univ., Waco, TX

Abstract: Early-Life Seizures and Neonatal Vocalizations in MiceJohn M. Reinhart, Katherine J. Blandin, Danielle Santana-Coelho, and Joaquin N. LugoRationale: Early-life status epilepticus (SE) can cause deficits in learning, memory, social behaviors, and communication. To better understand the acute impact of SE, ultrasonic vocalizations (USV) have been used to investigate

communication deficits in mice. Our lab previously found that mice that received SE on postnatal day (PD) 10, have a decrease in the quantity and duration of USVs at PD12. In the current study, we investigated whether SE induced on PD7 had changes in USVs on PD9 to determine if different communication alterations occur at an earlier stage in neonatal development. We also included females in this study to examine sex differences.Methods: In the present paradigm, we induced SE using an intraperitoneal (i.p.) injection of 0.5% kainic acid (2.5 mg/kg), on C57BL/6 male and female mice at PD7. Age matched control male and female pups were given i.p. injections of 0.9% physiological saline. We then investigated the communicative behavior of these mice using USVs recorded on PD9. Two-way ANOVAs for sex [male, female] X treatment[control, SE] were run to assess changes in the total number of calls, total time vocalizing, peak call frequency, maximum call amplitude, and total number of each call individually based on a classification system using ten different categories.Results: We did not find differences in the total number of USVs emitted, total time vocalizing, peak call frequency, and maximum call amplitude between the SE and control group. We did observe a main effect of treatment with an increase in the total number of chevron calls ($p<0.05$) emitted by the SE group compared to the control. There was also a main effect of sex with upward and complex calls($p<0.05$) being emitted at a higher number by females over males.Conclusions: In this study we found a significant increase in chevron calls made by the treated mice. There was also a greater number of upward and complex calls made by females compared to males. We did not find significant alterations in total number of calls following SE on PD7. This result is different from previous findings in SE on PD10. Additionally, these findings support the need for further investigation into the communication differences between males and females reaching SE during this neonatal period and to the broader effects during early development.

Disclosures: **J.M. Reinhart:** None. **J.N. Lugo:** None. **K.J. Blandin:** None. **D. Santana-Coelho:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.13

Topic: B.08. Epilepsy

Support: NIH Grant NS124145

Title: Dissecting the role of mtor compelxes in epilepsy and seizure disorders

Authors: *J. T. OKOH[1], J. L. MAYS[2], A. BACQ[3], K. IMANBAYEV[2], H. ZHOU[2], P. JAFAR-NEJAD[4], J. L. NOEBELS[2], S. BAULAC[2], M. COSTA-MATTIOLI[2];
[1]Baylor Col. of Med., [2]Baylor Col. of Med., Houston, TX; [3]ICM, Paris, France; [4]Neurosci. drug discovery, Ionis Pharmaceuticals, Carlsbad, CA

Abstract: Dysregulation of the mechanistic target of rapamycin (mTOR), which functions via two distinct complexes named mTORC1 and mTORC2, has been causally linked to epilepsy. Currently, it is widely believed that hyperactivation of mTORC1, which is sensitive to the drug rapamycin, drives epilepsy. Most of the evidence supporting the role for hyperactivation of mTORC1 in epilepsy relies on its chronic pharmacological inhibition with rapamycin. However, chronic rapamycin treatment, which reduces seizures in several epilepsy models, also inhibits mTORC2. We recently found that genetic inhibition of mTORC2 improved behavioral deficits and rescued the seizure phenotype in mice lacking the mTOR upstream negative regulator, *Pten*. Thus, it remains unclear whether hyperactivation of mTORC1 or mTORC2 leads to abnormal synchronized neuronal firing during epilepsy. To dissect the role of mTOR complexes in epilepsy, we selectively silenced the activity of either mTORC1 or mTORC2 in forebrain neurons. Using the Cre-lox system, we deleted *Rptor* (encoding the defining mTORC1-defining component, Raptor) and *Rictor* (encoding the mTORC2-defining component, Rictor) to inhibit mTORC1 and mTORC2, respectively. Next, we examined both behavioral and electrographic seizures in the kainic acid (KA) model, which is one of the most widely studied epilepsy models in the field. We found that KA activated mTORC2 more persistently than mTORC1. Surprisingly, we found that genetic inhibition of mTORC1 increased acute KA-induced seizures while genetic inhibition of mTORC2 reduced acute KA-induced seizures. Our results indicate that the mTOR complexes may play distinct roles in seizure generation. More importantly, future studies in other epilepsy and seizure models can help stratify seizures into mTOR-complex-specific subspectra and could inform more effective therapeutic strategies.

Disclosures: J.T. Okoh: None. **J.L. Mays:** None. **A. Bacq:** None. **K. Imanbayev:** None. **H. Zhou:** None. **P. Jafar-Nejad:** None. **J.L. Noebels:** None. **S. Baulac:** None. **M. Costa-Mattioli:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.14

Topic: B.08. Epilepsy

Support: NIH Grant R35GM133440

Title: The astrocyte brain-type fatty acid binding protein, Fabp7, regulates time of day seizure threshold in mice

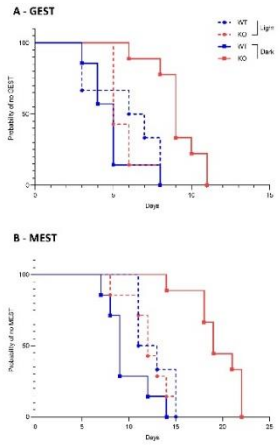
Authors: *M. LEFTON, C. FLORES, C. DAVIS, J. GERSTNER;
Washington State University, Spokane, Spokane, WA

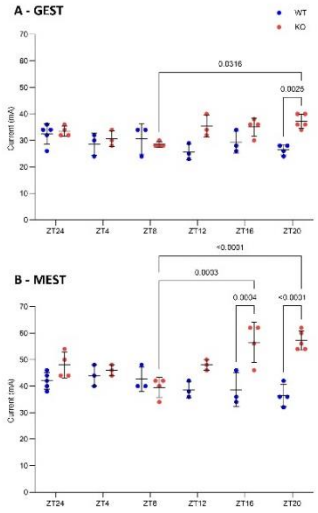
Abstract: Patients with epilepsy often experience increased frequency of seizures at night. Given the role glial cells play in modulating neuronal excitability, we hypothesize that circadian changes in glial cells may affect seizure threshold. Fatty acid binding protein 7 (Fabp7) is

expressed in brain astrocytes and is involved with the fatty acid transport, signal transduction, and gene transcription. Its mRNA expression cycles in a circadian rhythm and is necessary for sleep regulation. We examined if Fabp7 influences seizure threshold upon electrical stimulation. Male C57/BL6N wild type (WT) and Fabp7 knockout (KO) mice were maintained on a 12/12 hour light/dark cycle. Seizure thresholds were measured by administering a once daily, regularly increasing electroshock stimulus. General or maximal seizure was determined by the mouse's response to the stimulus.

Results showed that KO mice required a higher current to elicit both general and maximal seizure compared to WT mice (unpaired t test). Additionally, KO mice had a lower probability of seizure occurrence than both KO mice in the light phase and WT mice in the dark phase (Kaplan-Meier log rank test).

Astrocyte expressed Fabp7 plays an integral role in modulating neuronal excitability. Mice without functioning Fabp7 have an increased seizure threshold to electrical stimulus and therefore have reduced excitability in brain regions where Fabp7 is normally expressed. Further work is now needed to elucidate the pathway by which Fabp7 alters seizure threshold.





Disclosures: **M. Lefton:** None. **C. Flores:** None. **C. Davis:** None. **J. Gerstner:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.15

Topic: B.08. Epilepsy

Support: 5R01NS036692

Title: Changes in extracellular matrix coverage lead to functional alterations in CA1 interneurons in a mouse model of temporal lobe epilepsy

Authors: *A. M. WOO[1], D. C. PATEL[2], B. P. TEWARI[2], H. SONTHEIMER[2];
[1]Neurosci. Dept., Univ. of Virginia Neurosci. Grad. Program, Charlottesville, VA; [2]Neurosci. Dept., Univ. of Virginia, Charlottesville, VA

Abstract: Epilepsy is a neurological condition characterized by recurring, spontaneous seizures, which are often attributed to underlying imbalances in excitation and inhibition. Epileptic brain activity is linked to many cellular and molecular dysfunctions, including alterations in the function of inhibitory interneurons and changes in extracellular matrix, as seen in both human patients and a variety of acquired epilepsy models. Of particular interest are the assemblies of the extracellular matrix, called perineuronal nets (PNNs), which condense into net-like coatings that surround the soma and contribute to the firing of the fast-spiking GABAergic interneurons they envelop. We are interested in the functional differences of PNNs in healthy versus epileptic brains and how PNNs may be altered in multiple acquired epilepsy mouse models, considering their role in interneuron function. In the present study, we have characterized PNNs in a pilocarpine mouse model of temporal lobe epilepsy, focusing on both population-wide, comprehensive changes as well as structural and cell-level changes. While doing so, we observed that a sparse population of cells in the hippocampal CA1 region displays PNNs post-onset of *status epilepticus*; however, mice that received a sham injection or did not acquire epilepsy after treatment do not exhibit PNNs. Additionally, these cells stain negative for parvalbumin, indicating that they are not fast-spiking interneurons. We therefore sought to identify these cells and investigate their properties through immunohistochemical and electrophysiological means. Here, we characterize PNN structures in a pilocarpine mouse model and show that the CA1 "mystery cells" are in fact interneurons via immunohistochemical staining. Furthermore, we show that the electrophysiological activity of the interneurons from pilocarpine-treated mice that developed status epilepticus indeed differs from that of control mice. We propose that our findings signify a functional difference in these cells triggered by pilocarpine-induced epilepsy, as indicated by the development of the surrounding PNN structures, and hypothesize that these interneurons are involved in either a causative or compensatory role in epileptogenesis, requiring further study in this and other models.

Disclosures: **A.M. Woo:** None. **D.C. Patel:** None. **B.P. Tewari:** None. **H. Sontheimer:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.16

Topic: B.08. Epilepsy

Title: Aqp4 immunoexpresion in brain of rats exposed to kainic acid and petilenterazole

Authors: E. CRUZ-ANTONIO[1], C. NAVA-RUÍZ[1], N. LÓPEZ-DÍAZ GUERRERO[2], P. YESCAS-GÓMEZ[1], ***M. MENDEZ-ARMENTA**[3];
[1]Inst. Nacional de Neurologia y Neurocirugia, Ciudad de México, Mexico; [2]Univ. Autónoma metropolitana, Ciudad de México, Mexico; [3]Natl. Inst. Neurol Neurosurg., Mexico City, Mexico

Abstract: Epilepsy is a neurological disorder of the central nervous system characterized by increased and abnormal synchronization of neuronal activity, which is clinically manifested

among others by recurrent unpredictable and uncontrollable spontaneous seizure and the temporal lobe epilepsy is the most common type of acquired and frequent epilepsy in adults. Aquaporine 4 (AQP4) is a transmemabranal protein such as regulates the flow the water in the cell; is expressed in brain by glial cells, especially at specialized membrane domains including astroglial endfeet in contact with blood vessels. In patients with focal cortical dysplasia and epilepsy higher AQP4 expression was found, whereas that in animal models has been poorly studied. In this work we evaluated the immunoexpression of AQP4 in brain of rats exposed to kainic acid (KA) and pentylentetrazole (PTZ). Male Wistar rats were used, were divided into three experimental groups, a control group Saline solution (SS), KA group (10 mg/kg ip) and the third group such as administered with 3 doses of PTZ (25 mg/kg ip) in 15-minute intervals. Cresyl Violet stain and immunohistochemistry antibody for AQP4 were applied. We founded in rats treated with KA strongly AQP4 immunopositive capillaries amidst a weakly labeled neuropil observed mainly in astrocytes of hippocampal brain region in rats; while in neuropil, astrocytes and around blood vessels immunoreactivity remained without obvious changes in its intensity in PTZ-treated rats. These preliminary results support that AQP4 increased expression is a part of protective mechanism to maintain water transport and to modulate ion homeostasis in the rat brain in experimental models of epileptic seizures

Disclosures: E. Cruz-Antonio: None. **C. Nava-Ruíz:** None. **N. López-Díaz Guerrero:** None. **P. Yescas-Gómez:** None. **M. Mendez-Armenta:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.17

Topic: B.08. Epilepsy

Support: NS-45540

Title: Region and cell type specific deletion of neurexin-2 causes spontaneous recurrent seizures and ASD-like behavior impairments

Authors: *M. HAILE[1], G. DE CARVALHO[2], L. Y. CHEN[2];
[1]Anat. and Neurobio., Univ. of California Irvine, Irvine, CA; [2]Anat. and Neurobio., Univ. of California, Irvine, Irvine, CA

Abstract: A well-known central nervous system dysfunction associated with autism spectrum disorder (ASD), and ASD's comorbid disorders such as epilepsy, is hyperexcitability. Hyperexcitability may occur due to dysregulated excitatory glutamatergic and/or inhibitory GABAergic neuronal firing. The underlying mechanisms that cause hyperexcitability and predispose the brain to ASD and epilepsy remains unclear. Neurexin-2 (Nrxn2) is a presynaptic adhesion molecule which plays a critical role in synapse organization and neurotransmission and has been linked to many neuropsychiatric disorders including ASD and epilepsy. A recent study

using Nrxn2 conditional knockout (cKO) mice showed a novel inhibitory role specifically in excitatory synapses. The present study used Emx1-Cre mice to conditionally delete Nrxn2 in excitatory neurons of hippocampus and cortex to identify Nrxn2's role in regulating hyperexcitability. Using intracranial electroencephalogram (iEEG) recording, we demonstrate for the first time that deletion of Nrxn2 in excitatory synapses caused spontaneous recurrent seizures, as confirmed by the Racine scale. Moreover, the Nrxn2cKO has a decreased seizure threshold and increased seizure-related death. Behavioral analysis revealed ASD-like behavior impairments in the Nrxn2cKO. Our electrophysiology recordings showed the disruption of excitatory and inhibitory balance, which likely contributed to the generation of unprovoked spontaneous seizures. This study provides the first evidence that deletion of Nrxn2 induces hyperexcitability that manifests into spontaneous recurrent seizures and ASD-like behavioral impairments.

Disclosures: **M. Haile:** None. **G. de Carvalho:** None. **L.Y. Chen:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.18

Topic: B.08. Epilepsy

Support: NIH R01-NS033310
NIH U54-NS100064
VA Merit BX003485
VA Merit RX000669
DOD AZ14016
Oskar Fisher Foundation, Dr. James Truchard
NIH R01-AG066212

Title: Impaired hippocampal-prefrontal cortex connectivity with development of cognitive disfunction

Authors: *L. LI[1,2], U. KUMAR[2], B. XIE[1], S. A. FRAUTSCHY[2], G. M. COLE[2], H. SHAHPASAND-KRONER[2], J. ENGEL, JR[2], A. BRAGIN[2];
[1]Univ. of North Texas, Denton, TX; [2]Univ. of California Los Angeles, Los Angeles, CA

Abstract: The aim of this study was to explore the neuronal mechanisms underlying seizures in a rat model of Alzheimer's disease (AD) and their relevance to the development of cognitive impairment. Seven AD Tg (FAD+) rats (5-7 months old), which overexpress dominant familial AD mutations. app (APPsw) and Δ exon 9 mutations in human presenilin-1 (PS1EΔ9), and seven age matched controls (FAD-) participated in this study. Rats were implanted with 16-channel depth electrodes to record brain electrical activity in the hippocampus, prefrontal cortex, thalamus, and striatum. All animals performed a Chessboard Maze Task to assess memory scores

and award-seeking motivation. Seizures were observed in 3 of 7 FAD+ rats. Interictal epileptiform discharges (EEG spikes) were observed in all 7 FAD+ animals. No seizure activity was observed in the FAD- control group. The rate of pathological high-frequency oscillations (pHFOs) was significantly higher in the FAD+ group compared to the FAD- group. In animals in the FAD+ group, nearly 17% of the pHFOs are coupled between the prefrontal cortex and the hippocampus. This finding was associated with a significant decrease in memory scores and reduced motivation to seek rewards. Our data suggest that pathological HFOs occur in a rat model of AD which is associated with the ongoing epileptogenesis, and we demonstrate that impaired hippocampal-prefrontal cortex HFO couplings are associated with the development of cognitive dysfunction.

Disclosures: L. Li: None. **U. Kumar:** None. **B. Xie:** None. **S.A. Frautschy:** None. **G.M. Cole:** None. **H. Shahpasand-Kroner:** None. **J. Engel, Jr:** None. **A. Bragin:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.19

Topic: B.08. Epilepsy

Support: Department of Biotechnology, Ministry of Science & Technology, Government of India Grant No: BT/PR20392/MED/122/26/2016

Title: Golgi-cox staining reveals altered apical, basal dendrites and spine density in the hippocampus, atl and neocortex of pilocarpine model of temporal lobe epilepsy (TLE)

Authors: *V. DUBEY[1], A. ROY[2], A. B. DIXIT[5], M. TRIPATHI[3], P. CHANDRA[4], J. BANERJEE[1];
[1]Biophysics, [2]Physilogy, [3]Neurol., [4]Neurosurg., All India Inst. of Med. Sciences, Ansari Nagar, New Delhi, India; [5]ACBR, Delhi Univ., Delhi, India

Abstract: Introduction: Temporal lobe epilepsy (TLE) is a distributed network disorder, which involves the hippocampus and extra-hippocampal structures. Epileptogenesis in temporal lobe epilepsy (TLE) is tightly associated with neurogenesis, plastic changes and neural network reorganization. In the present study, we investigated using Golgi impregnation, tiny neuronal architecture of pyramidal neurons in the in the hippocampus, anterior temporal lobe (ATL) and neocortex at the dendritic and spine levels **Methods:** Li-Pilocarpine was used to induce a status epilepticus (SE) in S. D. rats. Golgi-Cox staining was achieved according to standard protocol to visualize neurons in their cell soma(cell body), axons, dendrites, and spines. In brief, rats were transcardially perfused with 0.9% of saline (PBS) for 3 min followed by 4% PFA in 0.9% saline for a further 5 min. Later the perfusion, brains were dissected out and fixed in Golgi solution for 48 hrs. The brain sectioning was performed using vibratome into 200-μm slices, followed by incubation in 0.1 M Tris buffer (pH 7.3) and then relocated onto gelatin-coated slides. The slides

were dehydrated at room temperature and further followed by alcohols gradient decreasing order series for dehydration. Golgi-Cox stained pyramidal neurons of hippocampal CA1, 5th layer of pyramidal neurons of ATL, and neocortex of pilocarpine and control rats sections were observed under a microscope (Olympus BX50) and analyzed using Neurolucida software (MBF Bioscience, Williston, VT). Dendritic morphology was analyzed by using a 40× objective lens. **Results:** The length of apical and basal dendrites, spine density and soma architecture were altered in pilocarpine rat model of epilepsy. Sholl analysis revealed a significant increase spine density, number of intersections, length of the apical and basal dendrite in pilocarpine treated rats compared to control rats. **Conclusion:** This study demonstrated the alterations in morphology, spine density, and axonal processes of pyramidal neurons in hippocampus, ATL and neocortex suggest restructuring of dendrites and spines, could potentially be involved in production of new circuits, and synaptic reorganization in TLE.

Disclosures: V. Dubey: None. **A. Roy:** None. **A.B. Dixit:** None. **M. Tripathi:** None. **P. Chandra:** None. **J. Banerjee:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.20

Topic: B.08. Epilepsy

Support: University of Iowa Graduate College Post-Comprehensive Exam Research Fellowship (BLK)
NIH/NIGMS T32 GM007337 (University of Iowa MSTP)
NIH/NINDS R01 NS095842 (GFB)
Beth L. Tross Epilepsy Professorship (GFB)

Title: The nocturnal tendency of seizure-induced death is lost in mice genetically lacking central nervous system serotonin neurons

Authors: *B. L. KREITLOW, G. F. BUCHANAN;
Neurol., Univ. of Iowa, Iowa City, IA

Abstract: *Rationale:* People with epilepsy are at risk of sudden death, a phenomenon known as sudden unexpected death in epilepsy (SUDEP). Retrospective studies and captured events from epilepsy monitoring units consistently demonstrate that SUDEP is more common during the night. Historically, this nighttime tendency has been attributed to sleep. Recent evidence from our lab has demonstrated that an independent circadian mechanism contributes to this nocturnal death. In mice housed in constant darkness, seizures induced during wakefulness are more likely to be fatal during the subjective night. Accumulating evidence from mouse models of spontaneous seizure-associated death also demonstrate a nighttime tendency. This conserved nighttime phenotype in diurnal humans and nocturnal rodents suggest that an underlying

circadian rhythm may mediate this time-of-day dependent mortality. The neurotransmitter serotonin (5-HT) is a compelling target of study. In both humans and rodents, levels of 5-HT fluctuate throughout the day, with levels lowest during the night. 5-HT neurons have also been shown to have anticonvulsant effect, regulate breathing, and are necessary to awaken from sleep in response to hypercarbia. *Methods:* Male and female adult (3 - 7 month) *Lmx1b*$^{f/f}$ (homozygous for floxed *Lmx1b* alleles) and *Lmx1b*$^{f/f/p}$ (homozygous floxed *Lmx1b* alleles and hemizygous ePet1 Cre recombinase) mice were used for this study (N = 12 - 16 per group). Conditional knockout of the *Lmx1b* gene in ePet1 Cre-containing neurons eliminates >99% of 5-HT neurons in the central nervous system. Seizure naïve animals were housed in a 12:12 light-dark cycle with *ad libitum* access to food and water. A single maximal electroshock (MES) seizure (30 mA, 60 Hz, 200 ms) was induced at six evenly spaced time points (Zeitgeber Time (ZT) 2, 6, 10, 14, 18, and 22) during wakefulness. *Results:* Like our lab's previous findings from C57BL/6J mice, wild-type *Lmx1b*$^{f/f}$ mice are more likely to die following MES seizures induced during the dark phase of the twenty-four-hour day (58.3% versus 21.4% mortality at ZT 18 and 6, respectively). On the other hand, *Lmx1b*$^{f/f/p}$ mice demonstrate high mortality regardless of time of day (45.5% and 60.0% mortality at ZT 18 and 6, respectively). *Conclusions:* Seizures that occur during the night appear to carry a higher risk of mortality. The time-of-day and circadian mechanisms underlying this nighttime risk are poorly understood. Findings from this work suggest a serotonergic mechanism may be involved. Better understanding how 5-HT neuron-related physiology is influenced by time-of-day and circadian mechanisms may help us develop strategies to mitigate the nighttime risk of SUDEP.

Disclosures: B.L. Kreitlow: None. **G.F. Buchanan:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.21

Topic: B.08. Epilepsy

Support: NIH Grant NS116647
Quillen College of Medicine Summer Scholars Program

Title: Mitochondrial energetics display differential substrate preference in a Scn1a$^{-/+}$ Dravet Syndrome mouse model.

Authors: D. RIDLEN[1], H. COBBLE[1], **J. L. ALDRIDGE**[2], *C. R. FRASIER[2];
[2]Biomed. Sci., [1]East Tennessee State Univ., Johnson City, TN

Abstract: Mutations in *SCN1A,* which encodes the voltage-gated sodium channel Nav1.1, cause Dravet Syndrome (DS). Previous work has suggested that changes in cellular respiration and mitochondrial dysfunction may play, at least in part, a role in seizure development in DS models. We hypothesized that mitochondrial oxygen (O_2) consumption and substrate utilization would be

diminished in *Scn1a*$^{-/+}$ mice. We isolated mitochondria from *Scn1a*$^{-/+}$ (KO) and *Scn1a*$^{+/+}$ (WT) (N=4/genotype) mice at ages P20-P25 and performed high resolution respirometry on an O2k (Oroboros). Mitochondria were exposed to either substrates for Complex I driven respiration (glutamate/malate [G/M]) or Complex II driven respiration (succinate), followed by an addition of ADP. In the succinate group, pyruvate and rotenone were added in sequential order following ADP. In addition, respiration was measured in transverse brain slices in a subset of experiments (N=1/genotype). We found that neuronal mitochondria isolated from KO mice showed a preference for Complex I linked ATP production. Respiration after sequential additions of G/M then ADP were significantly increased in KO mice ($p = 0.027$ and 0.004 respectively). In experiments where succinate was given, there were no differences between KO and WT mice. However, the subsequent addition of ADP led to an increase in O_2 consumption in WT mice but failed to increase O_2 consumption across KO mice. Further addition of pyruvate led to a significant increase in respiration in KO mitochondria but only a modest increase in WT mitochondria. Finally, addition of rotenone in these experiments had a much larger effect on KO mitochondria compared to WT ($p = 0.01$). To determine if differences in respiration could be seen in intact tissue, O_2 consumption was measured in acute brain slices at baseline and following 4 µM kainic acid, a concentration we determined to trigger an increase in hippocampal firing. KO mice exhibited a larger O_2 consumption compared to WT ($p = 0.047$). Taken together, our results suggest that *Scn1a*$^{-/+}$ DS mice may have a heavier reliance upon Complex I mediated electron transport and ATP production. This over-reliance may play a role in changes to energetics during an epileptic event.

Disclosures: D. Ridlen: None. **H. Cobble:** None. **J.L. Aldridge:** None. **C.R. Frasier:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.22

Topic: B.08. Epilepsy

Title: Intraventricular flow barrier in epilepsy blocks fluid transport in the third brain ventricle

Authors: *R. FAUBEL, T. FEINSTEIN, C. LO;
Univ. of Pittsburgh, Pittsburgh, PA

Abstract: A cilia-based flow network resides in the brain ventricular system and confines cerebrospinal fluid (CSF) to flow channels and compartments. Implication of this highly complex transport system for brain functioning and neuropathological conditions has yet to be demonstrated. We investigated a possible role of such guided fluid flow in the context of CDKL5-deficiency disorder (CDD), a neurodevelopmental disorder with early onset intractable epilepsy. We first analyzed multiciliated tissues of patients and *Cdkl5* KO mice, then turned towards mice with heterozygous deletion of the ependyma-specific transcription factor *FoxJ1* and finally addressed the effect of ketogenic diet, a short-term treatment for intractable seizures

in CDD. We applied our methods of tracing cilia-mediated fluid transport along the multiciliated brain ependyma, and developed sophisticated analyses to compare cilia motion parameters and associated intracellular structures from live cell imaging and immunohistochemistry results. Further, we applied an anesthesia-based assay sensitive to CSF homeostatic regulation to determine seizure susceptibility. We applied standards of scientific rigor to verify significance of our results. We found abnormal cilia motion in CDD patients and *Cdkl5* KO mouse airway and brain including loss of synchrony and increased beating range suggest the cellular anchoring might be affected. Consistent with this, we found abnormalities of a newly found cellular structure that regulates the polarized beating of cilia and thus generate directional flow. Loss of uniform cilia beating polarity in an area critical for CSF clearance causes a flow blockage in *Cdkl5* KO mice. This was also observed in *FoxJ1* mice together with increased seizure susceptibility, demonstrating this novel type of ciliopathy is a novel mechanism in epilepsy. When administered a ketogenic diet instead of regular chew, *Cdkl5* KO mice showed normalization of flow patterning, motile cilia polarity, and decreased susceptibility to anesthesia-induced seizure suggesting that postnatal cellular pathways can be triggered to modulate cilia motion and associated fluid transport respectively clearance. This was significant for female but not male mice, consistent with observations in CDD patients. Together, these findings point to a crucial role of intraventricular flow barriers in the etiology of seizure, and point to cilia motion being a promising target for pharmacological intervention.

Disclosures: **R. Faubel:** None. **T. Feinstein:** None. **C. Lo:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.23

Topic: B.08. Epilepsy

Title: Elucidating the role of microglia in acute seizures using chemogenetics

Authors: *A. DHEER, D. B. BOSCO, J. ZHENG, L.-J. WU;
Mayo Clinic, Gu-4-12, Mayo Clin., Rochester, MN

Abstract: Abstract: Recent advances in epilepsy research highlight the significance of microglia, the resident immune cells of the brain in epileptogenesis. Additionally, studies describe the role of microglia Gi-signaling in regulating microglia-neuron interaction and neuronal excitability. To further elucidate the role of microglia in regulating neuronal activity during seizures, the present study was conducted using chemogenetic approaches (Designer Receptors Exclusively Activated by Designer Drugs, DREADDs) to manipulate microglial Gi-signaling. Healthy, adult (~8 weeks old), male transgenic $CX3CR1^{CRE\text{-}ER/WT}$ mice (Control) and $CX3CR1^{CRE\text{-}ER/WT}$: $R26^{LSL\text{-}hM4Di/WT}$ mice (Gi-Dreadd) were used in the study. Our results indicated that Gi activation by CNO reduced seizure scores significantly ($p<0.05$) in Gi-Dreadd mice (n=11) as compared to the Control group (n=11), 30 min after CNO injection. Using

immunostaining and confocal microscopy we determined microglial activation and cell density using Iba-1 and CD68 markers. The results reveal a significant upregulation of Iba-1^+ cell density and mean intensity in the CA3 region. The expression of CD68 was also elevated in the Gi-Dreadd group, suggesting increased activation and phagocytic function of microglia. Thus, Gi-Dreadd activation acutely activates microglia in ICV-KA model of epilepsy. We also determined that there is an increased interaction between microglia (Iba-1^+) and neuronal soma ($NeuN^+$) in the CA3 region. Moreover, we explored that this interaction reduced neuronal hyperactivity as evidenced by reduced cFos positive neuronal cells in the Gi-Dreadd group. Together, our current findings suggest that activation of microglial Gi-signaling has a protective role during *status epilepticus* and may offer a therapeutic target for epilepsy.

Disclosures: **A. Dheer:** None. **D.B. Bosco:** None. **J. Zheng:** None. **L. Wu:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.24

Topic: B.08. Epilepsy

Support: NIH grant R01NS106688

Title: Viral encephalitis-induced seizures are attenuated by Ido1 and Ido2 deficiencies in immune cells

Authors: *Z. A. MACDOWELL KASWAN[1], M. HURTADO[2], E. Y. CHEN[2], A. J. STEELMAN[2], R. H. MCCUSKER[2];
[1]Neurosci. Program, [2]Animal Sci., Univ. of Illinois At Urbana-Champaign, Urbana, IL

Abstract: Viral encephalitis is a serious medical condition, the manifestations of which include neuroinflammation, fever, cognitive impairment, neurodegeneration, and seizure development (ictogenesis). Perhaps the best-characterized preclinical model of viral encephalitis is intracranial infection of juvenile C57BL/6 mice with Theiler's murine encephalomyelitis virus (TMEV). TMEV infection causes changes which allow for the recruitment and entry of peripheral immune cells into the brain parenchyma. Myeloid-derived cells (monocytes/macrophages) are the first to respond, and their infiltration is tied to pathogen-induced ictogenesis. Using single-cell sequencing, we found that these cells contain high levels of *Tmev* RNA. The pro-inflammatory cytokine storm caused by infection also induces central nervous system expression of the indoleamine2,3-dioxygenases (Ido1 and 2). The metabolic pathway initiated by these two enzymes generates ictogenic tryptophan metabolites within immune cells. Thus, we explored the importance of Ido1 and Ido2 expressed by myeloid-derived cells for ictogenesis by breeding Ido1- and Ido2-floxed mice to those expressing Cre recombinase under the myeloid-cell-specific Lyz2 promotor. Seizure incidence was recorded for seven days following intracranial TMEV or sham injection. We found that TMEV-infected Cre-control male and female mice had a 49%

seizure incidence, whereas Ido1 and Ido2 deficiencies in myeloid-derived cells reduced seizure incidence to 16% (n=32-50, p<0.01). Hippocampal gene expression was analyzed by qPCR. Expression of *Tmev*, *Ido1,* cytokines (*IFNγ* and *TNFα*), and infiltrating immune cells markers were induced by TMEV infection and further elevated in mice experiencing at least one seizure compared to mice that did not seize. However, gene expression did not differ across genotype. Next, to study the direct effects of TMEV infection on myeloid-derived cells, peritoneal macrophages from wild-type, Ido1-deficient, and Ido2-deficient mice were treated *in vitro* with TMEV ± IFNγ. Consistent with established literature, IFNγ suppressed viral replication. However, there was no effect of genotype on either viral load or the effect of IFNγ. These results demonstrate that Ido1 and Ido2 expression in myeloid-derived cells drives ictogenesis independent of viral load or the inflammatory response, and suggest peripheral Ido1 and Ido2 as potential targets for anti-seizure drug development.

Disclosures: Z.A. Macdowell Kaswan: None. **M. Hurtado:** None. **E.Y. Chen:** None. **A.J. Steelman:** None. **R.H. McCusker:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.25

Topic: B.08. Epilepsy

Support: KAKENHI 19K07129
KAKENHI 15K07970

Title: Involvement of prostaglandin E_2 in kainic acid-induced acute seizures and febrile seizures

Authors: *Y. IKEDA-MATSUO[1], N. TOMORI[2], S. NARUMIYA[3], T. TAKAHASHI[2], M. TANABE[4];
[1]Hokuriku Univ., Hokuriku Univ., Ishikawa, Japan; [2]Hokuriku Univ., Kanazawa, Japan; [3]Kyoto Univ. Facul.Med., Kyoto Univ. Facul.Med., Kyoto, Japan; [4]Kitasato University, Sch. of Pharm., Kitasato University, Sch. of Pharm., Tokyo, Japan

Abstract: We have recently demonstrated that one of prostaglandin E_2 (PGE_2) receptors, EP3 is involved in stroke-reperfusion injury and glutamate-induced excitotoxicity. In this study, we investigated the involvement of EP3 receptors in seizures of two acute models, kainic acid (KA)-induced seizures and febrile seizures (FS). In a KA-induced seizure model, KA (30 mg/kg, i.p.) was injected to wild-type (WT) or EP3 knockout (EP3KO) mice. In a FS model, seizures were induced by pretreatment of lipopolysaccharide (LPS, 100 μg/kg, i.p.) and increasing body temperature twice (4 h-interval) for 30 min using infrared radiation lamp in 10 day-old WT or mPGES-1 KO (ES1KO) mice. The seizure score for 2 h after KA-injection in EP3KO mice was significantly lower than that in WT mice. The increases in numbers of c-fos-positive cells at 4 h after KA-injection and GFAP- and Iba-1-positive cells at 3 days after injection were significantly

less in EP3KO mice as compared with WT mice. The increases in mRNA expressions of COX-2, mPGES-1 and EP3 receptors, as well as GFAP and Iba-1, and production of prostaglandin E_2 observed in WT mice was also significantly less in EP3KO mice. Furthermore, KA induced severe neuronal loss in hippocampal CA3 region of WT mice, but not in that of EP3KO mice. In behavioral test, 10 mg/kg KA was injected repeatedly every hour until induction of status epilepticus. The total dose of KA in EP3KO mice was higher than that in WT mice, however, the seizure score in EP3KO mice was significantly less than that in WT mice. The spatial memories measured by Y-maze test were impaired by KA, and there were no significant differences between genotypes. Although total migratory distance in open field test was similar in all groups, the time spent in center area in WT mice, but not EP3KO mice, was significantly prolonged by KA. The production of PGE_2 and expression of mPGES-1 mRNA were also upregulated after FS. The upregulation of IL-1β and TNF-α were observed after FS in WT mice, but not ES1KO mice. Furthermore, mRNA of GFAP, but not Iba-1, was increased after FS only in WT mice, but not ES1KO mice. These results suggest that activation of EP3 receptors by PGE_2 through induction of mPGES-1 contributes to seizure susceptibility, glial activation, production of inflammatory cytokines, hippocampal neuronal loss, and reduction in anxiety. Although the role of EP3 receptors in febrile seizures should be determined using EP3KO infants, inhibition of mPGES-1 and/or EP3 receptors will be a valuable therapeutic option in treatment of temporal lobe epilepsy.

Disclosures: Y. Ikeda-Matsuo: None. **N. Tomori:** None. **S. Narumiya:** None. **T. Takahashi:** None. **M. Tanabe:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.26

Topic: B.08. Epilepsy

Support: H2020-MSCA-ITN project 722053 EU-GliaPhD
16GW0182 CONNEXIN, 01DN20001 CONNEX

Title: Reactive microglia are the major source of tumor necrosis factor alpha and contribute to astrocyte dysfunction and acute seizures in experimental temporal lobe epilepsy

Authors: *L. HENNING[1], H. ANTONY[1], A. BREUER[1], J. MÜLLER[1], G. SEIFERT[1], E. AUDINAT[2], P. SINGH[3], F. BROSSERON[4], M. T. HENEKA[5], C. STEINHÄUSER[1], P. BEDNER[1];
[1]Inst. of Cell. Neurosciences, Friedrich Wilhelms Univ. Bonn, Bonn, Germany; [2]Inst. of Functional Genomics, Univ. of Montpellier, CNRS, INSERM, Montpellier, France; [3]Plexxikon Inc., Berkeley, CA; [4]German Ctr. for Neurodegenerative Dis. (DZNE), Bonn, Germany; [5]Univ. du Luxembourg, Luxembourg, Luxembourg

Abstract: Extensive microglia reactivity has been well described in human and experimental temporal lobe epilepsy (TLE). To date, however, it is not clear whether and based on which molecular mechanisms microglia contribute to the development and progression of focal epilepsy. Astroglial gap junction coupled networks play an important role in regulating neuronal activity and loss of interastrocytic coupling causally contributes to TLE. Here, we show in the unilateral intracortical kainate (KA) mouse model of TLE that reactive microglia are primary producers of tumor necrosis factor (TNF) alpha and contribute to astrocyte dysfunction and serverity of status epilepticus (SE). Immunohistochemical analyses revealed pronounced and persistent microglia reactivity, which already started 4 h after KA-induced SE. Partial depletion of microglia using a coloncy stimulating factor 1 receptor inhibitor prevented early astrocyte uncoupling and attenuated the severity of SE, but increased the mortality of epileptic mice following surgery. Using microglia-specific inducible TNFalpha knockout mice, we identified microglia as the major source of TNFalpha during early epileptogenesis. Importantly, microglia-specific TNFalpha knockout prevented SE-induced gap junction uncoupling in astrocytes. Continuous EEG recordings revealed that during the first 4 weeks after SE induction, microglial TNFalpha did not significantly contribute to spontaneous generalized seizure activity. Moreover, the absence of microglial TNFalpha did not affect the development of hippocampal sclerosis but attenuated gliosis. Taken together, these data implicate reactive microglia in astrocytes dysfunction and network hyperexcitability after an epileptogenic insult.

Disclosures: L. Henning: None. **H. Antony:** None. **A. Breuer:** None. **J. Müller:** None. **G. Seifert:** None. **E. Audinat:** None. **P. Singh:** None. **F. Brosseron:** None. **M.T. Heneka:** None. **C. Steinhäuser:** None. **P. Bedner:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.27

Topic: B.08. Epilepsy

Support: NIH Grant 5R01NS036692

Title: Increased expression of chondroitin sulfate proteoglycan in dentate gyrus and amygdala contributes to seizures in a mouse model of infection-induced epilepsy

Authors: *D. C. PATEL[1], N. SWIFT[2], B. P. TEWARI[1], C. PRIM[1], L. CHAUNSALI[1], I. KIMBROUGH[1], H. SONTHEIMER[1];
[1]Dept. of Neurosci., Univ. of Virginia, Charlottesville, VA; [2]Dept. of Intrnl. Med., Wake Forest Univ. Sch. of Med., Winston-Salem, NC

Abstract: Extracellular matrix (ECM) is a glue-like component of the CNS that fills up small extracellular space surrounding all cellular structure. ECM contributes to the maintenance of ionic distribution, the regulation of synaptic plasticity, and the transport of neurotransmitters,

neuromodulators, nutrients and metabolites. The molecular mechanisms that disrupt the assembly and functions of the ECM can therefore contribute to neuronal network hyperexcitability and seizures. Chondroitin sulfate proteoglycan (CSPG) is a major constituent of the ECM, that along with other constituents, forms either amorphous interstitial matrix or well-organized lattice-like structures called perineuronal nets (PNNs) encapsulating mainly neuronal soma. Varying changes in the components of ECM including CSPG have been reported in human and in animal models of epilepsy, however, it is not entirely clear whether these changes are cause or consequences of seizures. In the present study, we investigated this question using a clinically relevant mouse model of acquired epilepsy caused by virus infection. Mice were treated with Theiler's murine encephalomyelitis virus (TMEV) and the brains samples were collected during acute infection period to evaluate the expression of CSPG by several biochemical methods. We found a distinct increase in the level of CSPG in the dentate gyrus and amygdala of mice that experienced seizures but not in seizure-resistant mice. In contrast, there was overall degradation of ECM in whole hippocampus. The PNNs in CA2 and CA3 regions, especially around dendritic processes, were structurally degraded, which was correlated with a concomitant increase in the activity of matrix metalloproteases (MMPs) in the region suggesting that MMPs may directly contribute to ECM degradation. TMEV-infected mice treated with minocycline, which reduces seizures and inhibits MMPs, significantly inhibited upregulation of CSPG in dentate gyrus and amygdala and suppressed the overall degradation of CSPG in hippocampus. Importantly, genetic and adeno-associated virus-mediated deletion of aggrecan in both dentate granule cells and amygdala significantly suppressed TMEV-induced seizures. Our data suggest a causal relationship between TMEV-induced seizures and enhanced deposition of CSPG in dentate gyrus and amygdala. Further studies are directed to investigate the physiological mechanisms whereby CSPG level in dentate gyrus and amygdala regulates network hyperexcitability.

Disclosures: D.C. Patel: None. **N. Swift:** None. **B.P. Tewari:** None. **C. Prim:** None. **L. Chaunsali:** None. **I. Kimbrough:** None. **H. Sontheimer:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.28

Topic: B.08. Epilepsy

Support: NINDS NS088776

Title: Ns-pten ko mice show age- and brain-specific changes in microglia activity.

Authors: *K. BLANDIN[1], D. A. NARVAIZ[1], P. D. WOMBLE[1], G. SULLENS[1], D. SANTANA-COELHO[1], E. KWOK[1], J. PILCHER[1], T. WILEY[1], G. O' NEILL[1], J. N. LUGO, Jr.[2];

[1]Psychology and Neurosci., Baylor Univ., Waco, TX; [2]Psychology and Neurosci., Baylor Univ., Mc Gregor, TX

Abstract: Rationale: Microglia act as a key component in maintaining homeostasis in the central nervous system. Seizures induce brain region dependent enhancements in microglia activation. Regional activation of microglia has been linked to higher seizure susceptibility and cognitive impairment, progressing with age. Neuronal subset specific phosphatase and tensin gene deleted on chromosome 10 (*Pten*) knockout (KO) mice display hyperactive mammalian target of rapamycin (mTOR) signaling in the hippocampus, cerebellum, and cortex followed by seizures that increase in severity with age.Methods: In the present study, we investigated region specific microglia activation in NS-*Pten* KO mice at multiple timepoints across the lifespan. Tissue collection of wildtypes (WT) and KO mice was done at week 5 and 15 weeks for Flow cytometry to analyze activated microglia as measured by MHC II in the hippocampus, cortex and cerebellum. Percent MHC II activated myeloid cells comparison was used for percent activation quantification. In a separate cohort of WT and KO mice, tissue collection for immunofluorescence was done at week 4 and 10 weeks of age to stain for activated microglia using Iba1 in the subsections of the dorsal hippocampus (dentate gyrus, CA1, CA2/3). Hippocampal region morphological alterations were recorded and Iba1 positive cells were quantified by region. Results: Chi squared analysis of myeloid cells in KO mice following flow cytometry at 5 and 15 weeks showed significantly greater activation compared to WT mice across regions. This significant increased activation was apparent in the cortex, hippocampus and cerebellum, ($p < 0.05$). At 15 weeks of age the KO mice continued to express increased activation over WT mice in the cortex and hippocampus ($p < 0.05$), but not the cerebellum. Two-way ANOVA analysis of the immunofluorescent imaging revealed that at 4 weeks of age, KO mice had significantly higher Iba1 activation over WT throughout the hippocampus, but not at the subregional level. At 10 weeks of age, KO mice expressed increased Iba1 activation in all hippocampal regions, as well as the total count for the entire structure. Conclusions: Within the NS *Pten* KO model, elevated levels of activated microglia are present in the hippocampus and cortex at 5 and 15 weeks of age. Specific analysis of the dorsal hippocampus revealed significant increases in microglia throughout the structure by 10 weeks of age. This study suggests future research on the impact of microglia in genetic models of epilepsy are needed.

Disclosures: K. Blandin: None. **D.A. Narvaiz:** None. **P.D. Womble:** None. **G. Sullens:** None. **D. Santana-Coelho:** None. **E. Kwok:** None. **J. Pilcher:** None. **T. Wiley:** None. **G. O' Neill:** None. **J.N. Lugo:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.29

Topic: B.08. Epilepsy

Support: NIH Intramural Research Program
NIH Intramural Research Training Award
NIH Medical Research Scholars Program

Title: Chronic Neuronal Activation Induces an Inflammatory Response and mTOR Activation in Rat Cortical Co-Cultures

Authors: *R. CARNEY[1], A. MORTAZAVI[2], M. ROCK[3], M. BACHANI[4], A. KSENDZOVSKY[4], K. ZAGHLOUL[1];
[1]Surgical Neurol. Br., NIH, Natl. Inst. of Neurolog. Disorders & Stroke (NINDS), Bethesda, MD; [2]Neurosurg., Univ. of Michigan, Ann Arbor, MI; [3]Georgetown Univ. Sch. of Med., Washington, DC; [4]Neurosurg., Univ. of Maryland, Baltimore, Baltimore, MD

Abstract: The consequences of chronic seizures remain difficult to study in the laboratory setting. Current methods such as animal kindling models and tissue from patients remain limited by available biochemical tools. Acute treatments of cell cultures with epileptogenic compounds have been performed and well characterized, but chronic, multiple-day treatments have not been widely explored. We have investigated a primary-cell culture model of neuronal activation to model chronic epilepsy. This model demonstrates key electrophysiological and inflammatory responses implicated in epilepsy. To accomplish this, we dissected cortices from neonatal rat pups which resulted in cultures yielding a mixture of cortical neurons, astrocytes, and microglia. To assess electrophysiology, we plated dissociated cortical cells on multi-electrode arrays (MEA). Cultures developed spontaneous neural activity by day in vitro (DIC) 7. To induce periods of neuronal hyperexcitability and to model chronic seizures, we treated the cells daily with 100 μM of 4-aminopyridine (4AP), a potassium channel inhibitor for up to 7 days. We quantified burst frequency across biological replicates and observed that 4AP induces a higher burst frequency compared to control cells on every day of treatment (n = 3). When looking across 7 days of treatment, 4AP treated cells had significantly higher burst frequencies at baseline starting at day 3 and continuing through day 7 compared to controls. To attenuate the response of cultures to 4AP, we treated the cells with a combination of 4AP and 10 nm of tetrodotoxin (TTX) for 1 hour each day. This treatment resulted in almost no firing during the 1-hour treatment period, and resulted in a baseline burst frequency comparable to that of controls (n = 3). We used this model to probe downstream mechanisms related to mTOR signaling and inflammation. After a single 1 hr 4AP treatment, western blot analysis showed elevated p-S6 which is indicative of mTOR activation. Using qPCR, we identified two cytokines TNF-α and IL1-β; whose RNA transcripts were significantly upregulated following 3 days of treatment (n = 3). mTOR activation, TNF-α and IL1-β; have all been observed following seizures in animal models and human epilepsy patients. Here, we propose a model of chronic neuronal hyperexcitability which recapitulates key downstream inflammatory findings. These results suggest this model's potential to study downstream effects of chronic neuronal activation. This model may be used to better understand molecular implications of chronic activation and to probe novel drug targets to help treat seizures.

Disclosures: R. Carney: None. **A. Mortazavi:** None. **M. Rock:** None. **M. Bachani:** None. **A. Ksendzovsky:** None. **K. Zaghloul:** None.

Poster

525. Epilepsy Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 525.30

Topic: B.08. Epilepsy

Support: R01NS107428
WT093205MA
WT104033AIA

Title: Glutamic-oxaloacetic Transaminase 2 (GOT2), novel variants substantiate its susceptibility for epilepsy and cognitive impairment

Authors: *A. GHAFFAR[1,2], M. A. USMANI[1,3], S. EFTHYMIOU[4], H. A. ARABIYAT[5], R. MAROOFIAN[4], H. VAN BOKHOVEN[6], H. HOULDEN[4], Z. M. AHMED[1], S. RIAZUDDIN[7], S. RIAZUDDIN[1];
[1]Otorhinolaryngology Head and Neck Surgery, Sch. of Medicine, Uni. of Maryland, Baltimore, MD; [2]Natl. Ctr. of Excellence in Mol. Biology, Univ. of the Punjab, Lahore, Pakistan; [3]Dept. of Mol. Biology, Shaheed Zulfiqar Ali Bhutto Med. Univ., Islamabad, Pakistan; [4]Dep. of Neuromuscular Disorders, UCL Inst. of Neurol., London, United Kingdom; [5]Ministry of Health, Al-Hussein Salt New Hosp., As-Salt, Jordan; [6]Dept. of Human Genetics, Donders Inst. for Brain, Cognition and Behaviour, Radboud Univ. Med. Ctr., Nijmegen, Netherlands; [7]Jinnah Burn and Reconstructive Surgery Center, Allama Iqbal Med. College, Univ. of Hlth. Sci., Lahore, Pakistan

Abstract: Epilepsy and cognitive impairment (CI) are common manifestations in neurodevelopmental disorders, which collectively affect >3% of world population. *GOT2*, encoding glutamic-oxaloacetic transaminase 2, biallelic and compound heterozygous variants are known to cause inborn errors of metabolism with heterogeneous phenotype including epilepsy and CI. GOT2 has transaminase activity for mitochondrion-based glycolysis and oxidative phosphorylation, and thus is essential for NAD+/NADH based malate-aspartate shuttle (MAS). GOT2 also has role in transaminate conversion of kynurenine into kynurenic acid (KYNA) in the brain. Here, through whole exome sequencing (WES) approach, we identify two homozygous predictive pathogenic variants, p.(Leu358Val) and p.(Lys309Asn), in two unrelated Asian families displaying epilepsy, CI and cephalopathies. Magnetic resonance Imaging (MRI) of the affected individuals show abnormal white matter regions, enlarged cisterna magna and thinning of corpus collasum. Both variants occur in the enzymatically active Aminotranseferase classI/II domain and are predicted to impact the catalytic activity of protein. Further, single cell-RNA expression data of developing telencephalon of human brain, revealed overlapping expression between *GOT2*, and genes involved in aspartate and glutamate amino acids synthesis (*MDH1, MDH2 and GLUD1*) which are critical neurotransmitters in synapse. We also observed high expression of these genes in excitatory neurons, radial glia and interneurons progenitor cells (IPCs). Previously, compound heterozygous and bi-allelic pathogenic variants in *GOT2* have been reported in four unrelated individuals of African origin, suffering with epilepsy and CI. Taken together, all affected individuals with variants in *GOT2* display severe to profound intellectual disability, microcephaly, psychomotor disabilities, feeding difficulties, epilepsy

along with cerebral atrophy and white matter abnormalities. In conclusion, our results increase the genetic and phenotypic spectrum variation associated with *GOT2* based inborn errors of metabolism and show its impact on growth and activity of the human brain.

Disclosures: A. Ghaffar: None. **M.A. Usmani:** None. **S. Efthymiou:** None. **H.A. Arabiyat:** None. **R. Maroofian:** None. **H. van Bokhoven:** None. **H. Houlden:** None. **Z.M. Ahmed:** None. **S. Riazuddin:** None. **S. Riazuddin:** None.

Poster

526. Epilepsy Mechanisms: Channels

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 526.01

Topic: B.08. Epilepsy

Support: MOST110-2628-B-A49-001

Title: De Novo Loss-of-function KCNA3 and KCNA6 Variants Cause Early Onset Developmental Epilepsies

Authors: *C. LO[1], M.-H. TSAI[5,7,6], Y.-J. WANG[8,9], E. HWANG[2,3,1,4];
[1]Inst. of Mol. Med. and Bioengineering, [2]Dept. of Biol. Sci. and Technol., [3]Inst. of Bioinformatics and Systems Biol., [4]Ctr. for Intelligent Drug Systems and Smart Bio-devices, Natl. Yang Ming Chiao Tung Univ., Hsinchu, Taiwan; [5]Dept. of Neurology, Div. of Epilepsy, [6]Dept. of Med. Research, Genomics and Proteomics Core Lab., Kaohsiung Chang Gung Mem. Hosp., Kaohsiung, Taiwan; [7]Col. of Med., Chang Gung Univ., Taoyuan, Taiwan; [8]Dept. of Senior Service Industry Mgmt., Minghsin Univ. of Sci. and Technol., Hsinchu, Taiwan; [9]Ctr. for Neuropsychiatric Res., Natl. Hlth. Res. Inst., Miaoli, Taiwan

Abstract: Epilepsy is characterized by abnormal brain activity in central nervous system which leads to seizure or unusual behaviors. It is well known that ion channels are essential for maintaining neuronal membrane potential and their mutations have been associated with epilepsies in humans. Potassium ion channels are responsible for regulating neuronal membrane potential and modulating the neuronal excitability; mutations in several shaker-type (Kv1) potassium channels, such as KCNA1 and KCNA2, have been shown to cause early onset epilepsies. In this study, we identified two *de novo* mutations in two additional Kv1 channels, one in KCNA3 and the other in KCNA6, that lead to early onset epilepsies. Both wild-type and mutant KCNA3/6 exhibit membrane association. Electrophysiological studies using the whole-cell patch-clamp technique detect loss-of-function effect in these variants, which could impair the repolarization in the membrane potential and lead to hyperexcitable neuronal activities. Our findings expand the list of Kv1 genes that associates with human developmental epileptic encephalopathy.

Disclosures: C. Lo: None. **M. Tsai:** None. **Y. Wang:** None. **E. Hwang:** None.

Poster

526. Epilepsy Mechanisms: Channels

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 526.02

Topic: B.08. Epilepsy

Support: DFG grant Le1030/16-1/2
Fond Nationale de la Recherche INTER/DFG/17/11583046
BMBF 01GM1907A/B/C/H
Foundation 'no epilep'
DFG Le1030/11-1/2
FNR INTER/ESF/10/02/CoGIE

Title: The KCNQ5 variant p.R359C implicated in genetic generalized epilepsies decreases phosphatidylinositol 4,5-bisphosphate binding and increases neuronal firing

Authors: *J. KRÜGER[1], J. SCHUBERT[1], J. KEGELE[1], G. SEEBOHM[2], P. YAN[1], M. KOKO[1], B. J. STEINHOFF[3], R. KRAUSE[4], P. MAY[4], H. LERCHE[1];
[1]Exptl. Epileptology, Univ. of Tuebingen, Tuebingen, Germany; [2]Cardiovasc. Med., Univ. of Muenster, Muenster, Germany; [3]Kehl Epilepsy Ctr., Kehl-Kork, Germany; [4]Luxembourg Ctr. for Systems Biomedicine, Univ. of Luxembourg, Belvaux, Luxembourg

Abstract: *KCNQ5,* a member of the highly conserved *KCNQ* gene family, encodes the α-subunits of the M-type, voltage-gated delayed rectifier potassium channel $K_V7.5$. These channels are important regulators of the neuronal M-current which is regulated not only by membrane voltage but also by binding of acetylcholine to muscarinic receptors. This triggers a signaling cascade that depletes phosphatidylinositol-4,5-bisphosphate (PIP_2) from the membrane and forces K_V7 channels to close reducing the M-current and resulting in increased neuronal firing. Pathogenic variants in this channel have been found in patients with developmental and epileptic encephalopathy (DEE) and/or intellectual disability (ID). Recently, we have described five additional pathogenic variants in patients with genetic generalized epilepsy (GGE). These patients predominantly had absence seizures and two of the variants were additionally associated with mild to moderate ID. Here, we set out to further investigate the effect of one of these variants, the p.R359C variant, on PIP_2 interaction and neuronal excitability. Patch-clamp recordings in Chinese hamster ovarian cells showed a dominant-negative complete loss-of-function (LOF) in current density even under $K_V7.3$ co-expression. Western blots showed no difference in expression of the variant as compared to the wild-type (WT) in whole cell lysates as well as in membrane biotinylation assays. We therefore concluded that the complete LOF was not caused by decreased membrane insertion but rather by an inability to open. Hence, PIP_2 levels were enhanced by co-transfection of a phosphatidylinositol-4-phosphate-5 kinase resulting in slightly elevated currents, yet only approximately 10% of WT current was reached. Depletion of PIP_2 by co-expressing a *Danio rerio* voltage-sensing phosphatase (Dr-VSP) resulted in an immediate and long-lasting current inhibition in the variant but not the WT. Furthermore,

phospholipid-overlay assays showed a significantly reduced binding affinity of the variant to PIP_2. Moreover, patch-clamp recordings in transfected primary hippocampal neuron cultures showed a significant increase in firing as compared to control neurons, while an overexpression of $K_V7.5$ WT channels resulted in significantly reduced firing. The effect of the p.R359C variant could be reversed by intracellular application of $ZnCl_2$. Our study identifies the p.R359 position as a PIP_2 binding site in $K_V7.5$ that is crucial for channel opening. Moreover, we demonstrate that the p.R359C variant results in increased neuronal firing, which can be reversed by $ZnCl_2$ application, opening new doors for treatment in patients.

Disclosures: **J. Krüger:** None. **J. Schubert:** None. **J. Kegele:** None. **G. Seebohm:** None. **P. Yan:** None. **M. Koko:** None. **B.J. Steinhoff:** None. **R. Krause:** None. **P. May:** None. **H. Lerche:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; German Research Foundation (DFG), Federal Ministry for Education and Research (BMBF), Else-Kröner Fresenius Foundation (EKFS), Bial, Boehringer Ingelheim. D. Fees for Non-CME Services Received Directly from Commercial Interest or their Agents (e.g., speakers' bureaus); Bial, Eisai, UCB/Zogenix, Desitin. F. Consulting Fees (e.g., advisory boards); Bial, Eisai, UCB/Zogenix, Arvelle/Angelini Pharma, IntraBio.

Poster

526. Epilepsy Mechanisms: Channels

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 526.03

Topic: B.08. Epilepsy

Support: NSF 2030348

Title: Role of a potassium ion channel mutant in neuronal development

Authors: *A. BORTOLAMI[1], W. YU[2], E. FORZISI[1], K. RITIK[1], I. ESTEVEZ[1], M.-R. RASIN[2], F. SESTI[2];
[1]Rutgers Univ., Piscataway, NJ; [2]Rutgers Univ. - Robert Wood Johnson Med. Sch., Piscataway, NJ

Abstract: Potassium ion channels regulate neuronal excitability and are capable of eliciting intracellular signal cascade in the central nervous system. Voltage-gated potassium ion channel subfamily B member 1 (KCNB1) is associated with integrins (IKCs) that is important for converting its electrical properties into a signal transduction that results in cell proliferation. Mutations of KCNB1 are associated with epileptic disorders. Particularly, a substitution mutation for Arginine to Histidine at position 312 in the KCNB1 gene ($KCNB1^{R312H}$) has been identified in children presenting early-onset epileptic encephalopathies. Children affected by this disorder present recurrent seizures and intellectual delays. To investigate this neurological condition, we

generated a CRISPR knock-in (KI) murine model harboring the KCNB1^{R312H} gene variant. Although KCNB1 is embryonically expressed, its role in neurodevelopment is unknown. Furthermore, since the KCNB1^{R312H}subunit of the IKCs complex may affect its signal transduction, we hypothesize the mutant channels to elicit an aberrant effect during neuronal development. We analyzed the KI mouse model at different developmental stages via immunohistochemistry, Golgi staining, western blots, and coimmunoprecipitation to analyze its role in neocortex development. Immunohistochemistry showed over-migration in upper cortical layers; Golgi staining revealed hyper arborization and lack in middle cortical layers; western blot and coimmunoprecipitation suggest variations in the macromolecular IKCs complex and its pathway. Our results support the hypothesis that defective IKCs, formed with the mutant KCNB1^{R312H}, can affect neurodevelopment. Our data reveals a previously unknown neurodevelopmental mechanism in which potassium channels affect the fundamental neuronal processes through mechanisms that do not directly depend on its current conducting properties.

Disclosures: **A. Bortolami:** None. **W. Yu:** None. **E. Forzisi:** None. **K. Ritik:** None. **I. Estevez:** None. **M. Rasin:** None. **F. Sesti:** None.

Poster

526. Epilepsy Mechanisms: Channels

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 526.04

Topic: B.08. Epilepsy

Title: The role of Kv4.2 in seizure susceptibility and neuronal degeneration

Authors: *M. AHERN, J.-H. HU, D. A. HOFFMAN;
NICHD/Eunice Kennedy Shriver Nat'l Inst. of Child Hlth. and Human Develop., Washington, DC

Abstract: Due to an imbalance between excitatory and inhibitory firing of neurons, seizures often lead to excitotoxicity and subsequently neurodegeneration. Found in high levels in the dendrites of the pyramidal neurons in the hippocampus, the voltage-gated potassium channel Kv4.2 plays a prominent role in regulating excitability. Thus, neurons without Kv4.2 are expected to be hyperexcitable and more susceptible to seizures. After 20mg/kg intraperitoneal (i.p.) injections of kainic acid (2mg/mL in saline solution) to both male adult wild type (WT) mice and Kv4.2-knockout mice (Kv4.2-KO), we evaluated how Kv4.2 impacts seizure outcomes three days and seven days post seizure induction. We recorded seizure severity by scoring the seizure using a modified Racine scale every five minutes for one hour. We confirmed that Kv4.2-KO (n=17) mice exhibited more severe seizures and longer in duration compared to the WT (n=20) mice. Given the inflammatory response to seizures, we measured glial fibrillary acidic protein (GFAP) expression, indicative of astrocyte activation. Both WT and Kv4.2-KO mice were found to have significantly increased levels of GFAP compared to controls at three days and seven days. Kv4.2-KO mice showed a trend toward elevated GFAP expression levels

compared to WT. Using basic Nissl staining, we counted the number of neurons presenting morphological changes in the pyramidal layer of CA1-CA3 regions in the hippocampus. We found there was no significant difference between WT and Kv4.2-KO at three or seven days post-seizure. These results confirm Kv4.2 contributes to severity and duration of seizures. Interestingly, despite their increased seizure susceptibility, initial assessment of cellular changes in Kv4.2-KO mice after seizure induction does not show increased inflammatory response or increased neurodegeneration.

Disclosures: **M. Ahern:** None. **J. Hu:** None. **D.A. Hoffman:** None.

Poster

526. Epilepsy Mechanisms: Channels

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 526.05

Topic: B.08. Epilepsy

Support: TKP20214

Title: Altered h-current in cortical interneurons of drug-resistant epileptic patients

Authors: M. TOTH[1], **S. SZOCS**[2], N. HENN-MIKE[2], A. AGOCS-LABODA[2], T. KOVACS[2], T. DOCZI[3], Z. HORVATH[3], J. JANSZKY[1], *C. VARGA[2];
[1]Dept. of Neurology, Med. school, [2]Dept. of Physiology, Med. school, Univ. of Pecs, [3]Dept. of Neurosurgery, Med. school, Univ. of Pecs, Univ. of Pecs, Pecs, Hungary

Abstract: Approximately 30% of the epileptic patients are drug-resistant. Most of this drug-resistant group has a well-defined, focal onset region. For these focal-onset, drug-resistant epilepsy patients the surgical resection might be the best therapeutic option for achieving seizure freedom. In many cases the pathological high-frequency oscillations (pHFO) are continuously present in the epileptogenic zone (EZ), however, the pHFOs do not always propagate to neighboring areas. The mechanism which blocks this continuously ongoing pHFOs from generating generalized epilepsy is not entirely known. In the present study, we compared the basic electrophysiological properties of GABAergic interneurons of surgically removed EZ and control (non-epileptic, subcortical tumor patient) cortical interneurons. Most GABAergic interneurons in the control cortical preparations showed prominent sag-potentials caused by h-current. In contrast, sag-potentials were less prominent or completely missing in epileptic interneurons. H-current in fast-spiker interneurons have been shown previously to be involved in the maintenance of repeated, high-frequency bursting of interneurons. We hypothesize that the epileptic interneurons are unable to maintain high-frequency bursting activity for longer period, which contributes to the propagation and generalization of pHFOs in epilepsy.Our research was funded by TKP20214. The research was performed in collaboration with the Nano-Bio-Imaging core facility at the Szentágothai Research Centre of the University of Pécs.

Disclosures: **M. Toth:** None. **S. Szocs:** None. **N. Henn-Mike:** None. **A. Agocs-Laboda:** None. **T. Kovacs:** None. **T. Doczi:** None. **Z. Horvath:** None. **J. Janszky:** None. **C. Varga:** None.

Poster

526. Epilepsy Mechanisms: Channels

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 526.06

Topic: B.08. Epilepsy

Support: Xenon Pharmaceuticals Inc.
Neurocrine Biosciences, Inc.

Title: Relationship between genetic variants and disease characteristics in patients with SCN8A developmental and epileptic encephalopathy (SCN8A-DEE) or SCN8A-related epilepsy

Authors: D. HAUBENBERGER[1], C. GRAYSON[2], A. CUTTS[2], C. LUZON[2], N. BUTTERFIELD[2], H. SAVOIE[3], M. F. HAMMER[4], J. SCHREIBER[5], S. N. PIMSTONE[2], C. HARDEN[2], *N. A. MINASSIAN[1], E. JEN[1], C. MCMICKEN[1], T. NGUYEN[1];
[1]Neurocrine Biosciences, Inc., San Diego, CA; [2]Xenon Pharmaceuticals, Inc., Burnaby, BC, Canada; [3]The Cute Syndrome Fndn., Troy, NY; [4]BIO5 Inst., Univ. of Arizona, Tucson, AZ; [5]Children's Natl. Hosp., Washington DC, DC

Abstract: SCN8A encodes the NaV1.6 channel, which is highly expressed in the central nervous system with a major role in regulating excitatory networks in the brain. SCN8A-DEE is a rare and severe genetic epilepsy syndrome characterized by early onset cognitive impairment, developmental delay, and intractable seizures. Variants in the SCN8A gene have also been reported in patients with a broader phenotypic spectrum with varying degrees of severity. A caregiver survey, solicited by an advocacy group (The Cute Syndrome Foundation) via an online questionnaire, was conducted to better understand variability in the clinical presentation of SCN8A-DEE and SCN8A-related epilepsy. A 36-question online survey was developed to obtain de-identified data from caregivers of children. Patterns of association were investigated between SCN8A genetic variants and disease characteristics, including seizure type, frequency, and progression. Of 70 SCN8A reported variants, G1475R (n=6), R850Q (n=6), and R1872W (n=4) were the most common. These variants were significantly correlated with absence seizures and infantile/epileptic spasms at onset, current seizures and seizure clusters, and seizure-free periods (Fisher's exact p-value <0.05). Among the 16 patients with these 3 variants, the most common current seizure types were generalized tonic-clonic (n=8) and absence (n=8). Among the 6 patients with the R850Q variant, myoclonic and partial/focal seizures were also common (both, n=4). Seizure frequency of 1 to >10x per day was reported for R850Q (n=5) and R1862W (n=1). Since seizure onset, 67% (4/6) of G1475R patients worsened over time, 83% (5/6) of R850Q patients had variable progression (periods of worsening and improvement), and 50% (2/4) of R1872W patients improved over time. SCN8A-related epilepsy is a disorder in which

phenotypes may differ across the spectrum of genotypes. Effective anti-seizure medications are still needed for this severely impacted patient population.

Disclosures: **D. Haubenberger:** A. Employment/Salary (full or part-time):; Neurocrine Biosciences, Inc. **C. Grayson:** A. Employment/Salary (full or part-time):; Xenon Pharmaceuticals, Inc. **A. Cutts:** A. Employment/Salary (full or part-time):; Xenon Pharmaceuticals, Inc. **C. Luzon:** A. Employment/Salary (full or part-time):; Xenon Pharmaceuticals, Inc. **N. Butterfield:** A. Employment/Salary (full or part-time):; Xenon Pharmaceuticals, Inc.. **H. Savoie:** None. **M.F. Hammer:** None. **J. Schreiber:** None. **S.N. Pimstone:** A. Employment/Salary (full or part-time):; Xenon Pharmaceuticals, Inc. **C. Harden:** A. Employment/Salary (full or part-time):; Xenon Pharmaceuticals, Inc. **N.A. Minassian:** A. Employment/Salary (full or part-time):; Neurocrine Biosciences, Inc. **E. Jen:** A. Employment/Salary (full or part-time):; Neurocrine Biosciences, Inc. **C. McMicken:** A. Employment/Salary (full or part-time):; Neurocrine Biosciences, Inc. **T. Nguyen:** A. Employment/Salary (full or part-time):; Neurocrine Biosciences, Inc..

Poster

526. Epilepsy Mechanisms: Channels

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 526.07

Topic: B.08. Epilepsy

Support: UCA-JEDI ANR-15-IDEX- 01
LabEx ICST ANR-11-LABX-0015-01

Title: Preictal perturbation of GABAergic neurons' activity leads to Low-Voltage-Fast onset seizures in Dravet syndrome patients and mice.

Authors: F. CAPITANO[1], M. KUCHENBUCH[3], J. LAVIGNE[2], R. NABBOUT[3], ***M. MANTEGAZZA**[2];
[1]Inst. of Mol. and Cell. Pharmacol., [2]Inst. of Mol. & Cell. Pharmacol. (IPMC), CNRS, Valbonne - Sophia Antipolis, France; [3]Pediatric Neurol., Hôpital Necker-Enfants Malades, Paris, France

Abstract: Understanding the neural dynamics leading to seizures is important for disclosing pathological mechanisms of epilepsy and developing therapeutic approaches. This can be particularly important for developmental and epileptic encephalopathies (DEEs), in which seizures are commonly drug resistant and may worsen cognitive and behavioral outcomes. We have analyzed here the electrographic activity recorded during convulsive seizures in patients and gene targeted mouse models of Dravet syndrome (DS), an archetypical DEE in which hypoexcitability of GABAergic neurons is considered to be the main pathological mechanism. We analyzed the onset features in EEG recordings of individuals with Dravet syndrome carrying a *SCN1A* pathogenic variant, obtained from the database of the Necker-Enfants Malades Hospital (Paris, France). We recorded electrocorticograms with epidural electrodes, hippocampal local

field potentials with depth electrodes and hippocampal single unit activities with tetrodes in *Scn1a*$^{+/-}$ knock-out and *Scn1a*$^{RH/+}$ knock in DS mice.
In both patients and mice, we surprisingly observed seizures with low voltage fast (LVF) onset for the majority of them, which has been described before in focal onset seizures and it is thought to be generated by increased activity of GABAergic neurons. Thus, LVF onset seizures are unexpected in a DEE in which activity of GABAergic neurons is reduced. By quantifying single unit neuronal activity and spectral features of the field potential signal, we have studied the conditions that lead to LVF onset seizures in Dravet syndrome mice. We observed that specific perturbations of the activity of putative fast-spiking interneurons in the pre-ictal period precede the increase of their activity, together with the entire neuronal network, at seizure onset. Moreover, we found early signatures of the preictal period in the spectral features of hippocampal and cortical field potential of DS mice and of DS patients' EEG.
Thus, generalized seizures in DS have features that are similar to those observed in other epilepsies for focal seizures triggered by hyperactivity of GABAergic neurons, but are characterized by reduced activity of these neurons in the preictal period.

Disclosures: F. Capitano: None. **M. Kuchenbuch:** None. **J. Lavigne:** None. **R. Nabbout:** None. **M. Mantegazza:** None.

Poster

526. Epilepsy Mechanisms: Channels

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 526.08

Topic: B.08. Epilepsy

Support: F31 NS120467
R01HL104101
R01HL137094

Title: Breathing abnormalities across development in a Scn1a haploinsufficient model of Dravet syndrome

Authors: *B. M. MILLA[1], D. K. MULKEY[2];
[1]Physiol. and Neurobio., Univ. of Connecticut, Storrs, CT; [2]Dept. Physiol. and Neurobio., Univ. of Connecticut Physiol. & Neurobio., Storrs Manfld, CT

Abstract: Dravet Syndrome (DS) is a severe form of epilepsy with a high rate of sudden unexpected death in epilepsy (SUDEP). Respiratory failure is a leading cause of SUDEP, despite this it is unclear how DS-associated genetic variants or seizure activity in general disrupts respiratory control. Most DS cases are caused by loss of function mutations in Scn1a, the gene that encodes a component of Nav1.1 channels that preferentially regulates activity of inhibitory neurons early in development. Consistent with this, we showed that conditional loss of Scn1a function in inhibitory neurons disrupted CO2/H+-dependent regulation of breathing (i.e.,

respiratory chemoreception) at the level of the retrotrapezoid nucleus (RTN) and caused seizures and premature death. However, recent evidence suggests expression of Scn1a by excitatory neurons also contributes to progression of DS symptoms. Therefore, the goal of this work is to determine whether and how respiratory function is perturbed at two developmental timepoints in Scn1a+/- mice. All mice are maintained on a 50:50 background of 129S1/SvlmJ::C57BL6/J. Breathing was measured under room air conditions and in response to high CO2 or hypoxia (10% O2) by whole-body plethysmography in neonatal (13 days postnatal) and weaning age (21 days postnatal) mice. We found that neonatal Scn1a+/- mice breathe normally in room air but show a blunted ventilatory response to 3% CO2. At the cellular level, RTN neurons in slices from neonatal Scn1a+/- mice show increased baseline activity and lower sensitivity to 10% CO2 compared to RTN neurons in slices from age matched littermate controls. At a later developmental time point that coincides with onset of mortality (3 weeks old), Scn1a+/- continued to exhibit a blunted ventilatory response to CO2. Notably, weaning age Scn1a+/- mice also showed a blunted ventilatory response to hypoxia and importantly this respiratory deficit positively correlated with mortality; mice that died by 24 days of age had a diminished hypoxic ventilatory response compared to Scn1a+/- that survived. These results show that loss of Scn1a function disrupts respiratory activity at the cellular and whole animal level, and breathing problems correlate with premature death in weaning age Scn1a+/- mice. It is worth noting that respiratory and mortality phenotype of Scn1a+/- mice are less severe compared to mice with Scn1a functional deficits only in inhibitory neurons, thus suggesting Scn1a in excitatory neurons contributes to features of DS.

Disclosures: B.M. Milla: None. **D.K. Mulkey:** None.

Poster

526. Epilepsy Mechanisms: Channels

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 526.09

Topic: B.08. Epilepsy

Support: NIH Grant R01NS094211
NIH Grant R03NS103111
NIH Grant R01NS110012
Institutional Training Grant 1T32NS09957801A1

Title: Temperature-induced increase of epileptic activity captured by machine learning in a mouse model of epilepsy

Authors: *A. I. BECK[1], D. KUNDA[4], A. M. BARD[5], F. K. KALUME[2], H. O. DE LA IGLESIA[3];
[1]Univ. of Washington, [2]Ctr. of Integrative Brain Res., [3]Biol., Univ. of Washington, Seattle, WA; [4]Natl. Inst. of Neurolog. Disorders and Stroke, Bethesda, MD; [5]Seattle Children's Res. Inst., Seattle, WA

Abstract: Epilepsy is a neurological disorder characterized by the presence of excessively synchronized neural activity in form of seizures and epileptiform discharge (interictal spikes; IS). In certain forms of epilepsy, epileptic activity increases in conjunction with core body temperature, such that both seizures and IS occur more frequently as an individual's body becomes hotter. Additionally, because increased IS frequency can be associated with seizure risk, real-time detection of IS could lead to seizure prediction and interventions to prevent seizures or their adverse outcomes. Thus, using a mouse model of epilepsy, we examined whether our machine-learning (ML) based method of autonomously detecting IS found a relationship between ML-predicted IS frequency and core body temperature. Neural activity was chronically recorded in mice using two electrocorticography electrodes and one electromyography electrode. We used data from a mouse model of Dravet syndrome (DS; heterozygous *Scn1a* gene deletion), along with wild-type controls. EEG data were collected from the Kalume lab and the De La Iglesia lab. All the data were normalized to a sampling rate of 400 Hz and min/max value of ±400 mV, and IS were manually scored in a subset of data. A total of 96 time- and frequency-based features were extracted in epochs of 5 seconds. The ML model was trained on data collected in the de la Iglesia lab and were randomly under-sampled to ensure balanced classes and used to train a single-layer Perceptron using L1 regularization to classify IS. The outcome was a binary classification; IS present/absent in the given epoch. The fully trained ML model had a mean training accuracy of 85.94. We used data collected from the Kalume lab to examine the relationship between IS frequency and core body temperature. Mice core body temperature was initially maintained at 27°C, and was afterwards gradually increased in a step fashion (0.2 degrees C every 2 min) until seizures were observed. As body temperature increased, frequency of observed seizures increased. Additionally, there was a significant correlation between ML-predicted IS frequency and temperature (r = 0.32, p = 6.90e-17), suggesting our ML-based method captures temperature-related increase in epileptic activity frequency in a mouse model of epilepsy. This algorithm could be used to detect IS frequency in real-time, and potentially forecast potential seizure onset.

Disclosures: A.I. Beck: None. **D. Kunda:** None. **A.M. Bard:** None. **F.K. Kalume:** None. **H.O. de la Iglesia:** None.

Poster

526. Epilepsy Mechanisms: Channels

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 526.10

Topic: B.08. Epilepsy

Title: A de novo variant (F325L) of GABRA1 bidirectionally alters GABA sensitivity at different ligand concentration

Authors: *W. CHEN[1], Y. BAI[1], E. CHAN[1], M. CHIU[2], M. DEMOS[2], L. LIU[1], Y. WANG[1]; [2]BC Children's Hospital, Div. of Neurology,Department of Pediatrics, [1]Univ. of British Columbia, Vancouver, BC, Canada

Abstract: The $GABA_A$ receptor ($GABA_AR$) is a chloride (Cl^-) channel receptor that primarily mediates neuronal inhibition, and thereby plays critical roles in maintaining excitation-inhibition (E/I) balance in the central nervous system (CNS). Mutations of the $GABA_ARs$ have been reportedly led to epileptic encephalopathy and neurodevelopmental disorders. Here, we report the identification of a novel de novo F325L (T973C) missense variant of the $GABA_AR$ alpha1 subunit from a pediatric patient diagnosed with focal epilepsy, global developmental delay and autism spectrum disorder. The F325 residue is a part of the pore-lining residues, indicating its important role in regulating channel pore gating. We used biochemical techniques, whole-cell and single channel patch-clamp recordings, and pharmacological characterizations in HEK293 cells overexpressing the wild-type and mutated human recombinant $GABA_ARs$ containing the $\alpha1/\beta2/\gamma2$ subunits. We found that this $\alpha1_{F325L}$ mutation produced a unique functional phenotype that is strikingly different from most of the mutational $GABA_AR$ variants previously identified from patients: it markedly increased the GABA-evoked whole-cell currents evoked with low concentrations of GABA, but decreased the maximum currents evoked with high concentrations of GABA. In addition, the $\alpha1_{F325L}$ mutation significantly increased the tonic current revealed in the presence of the GABAAR antagonist bicuculline. The gain of function at the low concentrations of GABA appeared to be mediated by decreasing the activation and prolonging the deactivation and desensitization of $GABA_ARs$. Moreover, this mutation was able to markedly improve the loss of function caused by the previously reported $\alpha1_{R214C}$ and $\alpha1_{A322D}$ mutations. Finally, this mutation did not overtly affect $GABA_AR$ expression in the cell and on the plasma membrane, suggesting the functional alterations were mainly resulted from the changes in the channel gating properties. This is the first observation that a single mutation of GABRA1 can cause an agonist-concentration dependent bidirectional modification of the $GABA_AR$ channel gating properties, which leads to focal epilepsy, global developmental delay and autism spectrum disorder.

Disclosures: W. Chen: None. **Y. Bai:** None. **E. Chan:** None. **M. Chiu:** None. **M. Demos:** None. **L. Liu:** None. **Y. Wang:** None.

Poster

526. Epilepsy Mechanisms: Channels

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 526.11

Topic: B.08. Epilepsy

Support: VA Merit I01BX004938
VA Merit I01BX002949
NIH R01NS126247
NIH R21NS102948
DoD CDMRP W81XWH-18-1-0598

Title: Histopathological markers of epilepsy in the hippocampus of CACNA2D2 knockout mice

Authors: *A. B. STAYNER[1], A. ISAKHAROV[2], K. A. BEESON[2], E. SCHNELL[1,3];
[1]Anesthesiol. and Perioperative Med., [2]Neurosci. Grad. Program, Oregon Hlth. & Sci. Univ., Portland, OR; [3]Portland VA Healthcare Syst., Portland, OR

Abstract: The voltage-gated calcium channel subunit gene CACNA2D2 controls calcium-dependent signaling in neurons, and loss of this subunit causes epilepsy and ataxia in both mice and humans. Although this gene is primarily associated with cerebellar Purkinje cell function, it is also expressed in the hippocampus. Homozygous CACNA2D2 mutant mice manifest electroencephalographic spike-wave discharges (SWDs) as well as generalized tonic-clonic seizures. SWDs are typically associated with aberrant thalamocortical activation, but due to the existence of generalized seizure events, we sought to determine whether these mice manifested signs of hippocampal involvement in seizure activity. We analyzed various histopathological correlates of epilepsy in the hippocampal dentate gyrus of juvenile (21-28 do) CACNA2D2 wildtype (WT) and knockout (KO) mice, using immunohistochemical staining and confocal microscopy. CACNA2D2 mice demonstrated increased sparse expression of the activity-dependent gene cFos within the dentate granule cell layer (GCL), as well as at the GCL-inner molecular layer (IML) border, consistent with the increased activation of granule cells and semi-lunar granule cells, respectively. In one KO mouse, there was widespread activation of nearly all granule cells, suggesting hippocampal involvement in recent seizure activity. We are currently analyzing other histopathological markers of epilepsy in these mice, including markers of altered neurogenesis (Ki67 and doublecortin), glial activation (GFAP AND Mac-2), and mossy fiber sprouting (ZnT3), as well as structural changes such as granule cell layer dispersion. Finally, we are analyzing tissue for changes in interneuron density, including changes in parvalbumin expression. Although it is unclear whether alterations in hippocampal circuitry are directly related to CACNA2D2 dysfunction or secondary consequences of recurrent seizures, our future work will analyze hippocampal electrophysiology at both earlier and later stages, to probe for potential functional changes in hippocampal network activity.

Disclosures: A.B. Stayner: None. **A. Isakharov:** None. **K.A. Beeson:** None. **E. Schnell:** None.

Poster

526. Epilepsy Mechanisms: Channels

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 526.12

Topic: B.08. Epilepsy

Support: NIH F31 NS124345-01
NIH R01 NS29709

Title: Transcriptional dysregulation of thalamic t-type calcium channel gene precedes seizure onset in childhood absence epilepsy mouse model tottering

Authors: *S. THOMPSON[1], A. SONIG[2], J. NOEBELS[1];
[1]Baylor Col. of Med., Baylor Col. of Med., Houston, TX; [2]Rice Univ., Houston, TX

Abstract: Childhood absence epilepsy (CAE), a common form of pediatric epilepsy, characterized by non-convulsive, generalized 3-5 Hz spike wave seizures (SWs). Loss of function mutations in *Cacna1a* that encodes P/Q calcium channels cause SWs in humans and the mouse model *tottering*. This model displays elevated thalamic T-currents prior to the onset of seizures. SWs are generated by the thalamocortical circuit that regulates important oscillations such as sleep spindles. However, abnormal activation due to elevated T-type calcium channel activity results in rebound bursts and SWs hundreds of times per day. The mechanism of how presynaptic P/Q channel deficits lead to elevated postsynaptic thalamic T-channel currents is poorly understood. To explore T-current remodeling in *tottering* mice, we performed transcriptional analysis of *Cacna1g*, the only T-channel α-subunit encoding gene expressed in thalamic relay nuclei. RNAscope fluorescent in situ hybridization technology (ACD Bio.) was utilized to target *Cacna1g* mRNA transcripts in sex-matched wildtype and homozygous *tottering* littermates. To compare expression levels before and after seizure onset, cohorts were taken at two timepoints (P14, P30). High magnification images (40x) of thalamic relay nuclei were quantified using HALO analysis software (Indica Labs). Expression of *Cacna1g* in thalamic relay cells of tottering mice prior to seizure onset (P14) showed a 2-fold increase in mRNA copy number/cell when compared to wildtype (WT= 8.6 Tg= 15.2 p-value: <0.0001). There is a significant difference in percentage of cells with low *Cacna1g* expression (WT= 27.5, Tg=15.8; p-value: 0.0006) and high *Cacna1g* expression (WT= 18.5 Tg=33.4; p-value:< 0.0001). Similarly, when the same regions were compared in adult (P30) *tottering* mice, there was a significant increase in mRNA copy number per cell (WT= 3.0 Tg= 5.4, p-value= 0.0005). Adult *tottering* mice continue to show a decrease in the percentage of cells with no *Cacna1g* expression (WT= 26.1, Tg=13.0; p-value: <0.0001) and increase in high *Cacna1g* expression (WT= 0.7 Tg=6.2; p-value: 0.0404) when compared to wildtype littermates. Our data are the first to identify transcriptional upregulation underlying elevated thalamic T-channel currents thought to drive absence seizures. Elevated T-type currents and transcription are present prior to the onset of seizures and persist in adult *tottering* mice, suggesting transcriptional upregulation may play a role in epileptogenesis.

Disclosures: S. Thompson: None. **A. Sonig:** None. **J. Noebels:** None.

Poster

526. Epilepsy Mechanisms: Channels

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 526.13

Title: WITHDRAWN

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.01

Topic: B.09. Glial Mechanisms

Title: Temporal dynamics of human astrocyte reactivity

Authors: *E. HILL, C. SOJKA, A. T. KING, S. A. SLOAN;
Emory Univ., Atlanta, GA

Abstract: Astrocytes respond to external or inflammatory stimuli, often produced by traumatic injury, ischemia, or neurological disease. These responses include functional, transcriptomic, and morphologic changes that together comprise a reactive phenotype. While reactive astrocytes can be induced via multiple intrinsic and extrinsic signals, exposure to microglial-secreted cytokines (Il-1α, TNF- α, and C1q) is a robust approach to trigger a reactive state in vitro. However, several important questions remain, including which genomic processes initiate and maintain the reactive state, and how the temporal duration of the reactive stimuli affects astrocytes genomically, transcriptionally, and functionally. Additionally, it remains unclear to what degree astrocyte reactivity is reversible, and whether this is dependent upon the length of the initial stimulus. To explore these questions, we leveraged the *in vitro* human cortical organoid (hCO) system, which recapitulates the developmental trajectory of human brain cell types, including the formation of quiescent, mature astrocytes. To validate the reactive astrocyte response to microglial cytokines in hCOs, we first exposed hCOs to these cytokines for 24hrs, performed bulk and single-cell RNA-sequencing, and observed robust transcriptomic changes reflective of a reactive state. These changes also reflect our observations in fetal human astrocytes exposed to the same cytokines. To test the effects of varying durations of cytokine exposure, we then exposed hCOs to cytokines for a time course spanning from one day to three months and performed paired ATAC and RNA-sequencing at each timepoint. These data reveal the existence of at least two distinct genomic and transcriptomic stages of reactivity — an acute phase (induced by cytokine exposure for 1-7 days), and a chronic phase (induced by cytokine exposure for 1-3 months). Both stages possess unique differentially accessible transcription factor binding motifs, such as BACH2 (acute) and PRDM1 (chronic), coupled with distinct differential gene expression profiles, which suggest that the reactive responses in astrocytes are temporally plastic. In ongoing experiments, we are now testing whether these acute and chronic states are reversible after cytokine withdrawal and which genomic loci are most dynamic. These experiments will elucidate how astrocytes respond to chronic stimulation, as well as the plasticity of the reactive astrocyte state.

Disclosures: **E. Hill:** None. **C. Sojka:** None. **A.T. King:** None. **S.A. Sloan:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.02

Topic: B.09. Glial Mechanisms

Support: NIH Grant 1R21NS119594-01
HJF 2021 Fellowship

Title: Astrocyte calcium signaling changes following nerve injury necessary for central sensitization

Authors: *M. A. PROKHORENKO[1,2], E. R. PETRUS[3], J. T. SMYTH[3];
[1]Uniformed Services Univ. of Hlth. Sci. Grad. Program In Neurosci., Bethesda, MD; [2]Henry M. Jackson Fndn. for the Advancement of Military Med., Bethesda, MD; [3]Uniformed Services Univ. of the Hlth. Sci., Bethesda, MD

Abstract: Chronic pain following an injury is a debilitating condition associated with serious comorbidities and opioid dependence. Understanding the transition from initial injury to neuropathic pain is essential for prevention and treatment. This transition is mediated by central sensitization (CS), which occurs via amplification of nociceptive signaling by the central nervous system and involves changes to neurocircuitry pathways. CS research and therapeutics have focused on neuronal circuitry, whereas the role of astrocytes is poorly defined. Astrocytes that normally maintain and modulate neuronal synapses become reactive and exhibit aberrant Ca^{2+} signals after injury and under neuropathic pain. However, how these Ca^{2+} signals are generated and how they modulate astrocyte function in CS are unknown. Importantly, CS involves significant upregulation of glutamate release at nociceptive synapses, and glutamate acts on astrocytes via metabotropic glutamate receptors to stimulate Ca^{2+} release from endoplasmic reticulum (ER) Ca^{2+} stores via inositol 1,4,5-tripshosphate receptors (IP_3Rs). We are testing the hypothesis that central sensitization requires astrocyte IP_3R-mediated Ca^{2+} release from ER Ca^{2+} stores and activation of store-operated Ca^{2+} entry (SOCE) by combining powerful genetic tools with a robust model of CS in *Drosophila melanogaster*. We assay CS in flies by amputating one leg and monitoring the flies' responses to subnoxious temperatures seven days following injury. Injured animals exhibit increased jumping behavior at the subnoxious temperature indicative of thermal allodynia. Using a "Ca^{2+} memory" indicator known as Transcriptional Reporter of Intracellular Calcium (TRIC), we found that astrocytes in the ventral nerve cord of injured animals exhibited significantly enhanced Ca^{2+} signaling activity 3-4 days following injury compared to uninjured controls. We also found that astrocyte-specific suppression of IP_3R as well as other SOCE components attenuated nociceptive jumping behavior at one week following nerve injury, These results implicate IP_3R and SOCE-mediated Ca^{2+} signaling as key components in regulating astrocyte activity during pain sensitization. We will combine these these findings with single-nucleus RNA sequencing analysis to identify differentially regulated signaling cascades in astrocytes that may further contribute to CS. Our results will bring new understanding to the role of astrocyte signaling in pain sensitization and may suggest novel therapeutic targets for the prevention or treatment of chronic pain.

Disclosures: M.A. Prokhorenko: None. **E.R. Petrus:** None. **J.T. Smyth:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.03

Topic: B.09. Glial Mechanisms

Support: NIH Grant AG068091
BrightFocus Foundation Grant A2019363S
Sanofi iAward
Swid Professorship

Title: Reactive oxygen species from mitochondrial complex III promote astrocytic STAT3 signaling and dementia-related pathology

Authors: *D. BARNETT, F. PALAGUACHI, T. S. ZIMMER, S. M. MEADOWS, A. G. ORR, A. L. ORR;
Feil Family Brain and Mind Res. Inst., Cornell University: Weill Cornell Med. Col., New York, NY

Abstract: Mitochondrial reactive oxygen species (mtROS) are implicated in diverse aging-associated neurological disorders, including Alzheimer's disease, frontotemporal dementia, and Parkinson's disease. Complex III of the respiratory chain has a high capacity for mtROS production and generates mtROS towards the cytosol, poising it to regulate intracellular signaling and disease mechanisms. However, the exact triggers, sources, and downstream effects of mtROS are unclear, in part because previously available tools for manipulating mtROS were not site-selective or disrupted mitochondrial respiration. Recently identified small molecules which suppress complex III site Q_o electron leak (S3QELs, "sequels") enable site-selective blockade of mtROS production by complex IIIQ_o without interfering with other mitochondrial processes. Here, we used S3QELs and other complementary approaches to manipulate complex IIIQ_o ROS in astrocytes, which produce abundant mtROS and contribute to neuropathological processes, in part via JAK-STAT3 signaling. Using biochemical and molecular assays, we found that disease-related factors such as oligomeric amyloid-β and interleukin-1α increased astrocytic ROS production and STAT3 activation and that these changes were significantly blunted by blockade of IIIQ_o ROS using structurally distinct S3QEL analogs. S3QELs also reduced the induction of astrocytic genes associated with neurodegeneration. In astrocytic-neuronal co-cultures, but not in neurons cultured alone, S3QELs reduced neuronal dysfunction and cell death, suggesting that IIIQ_o ROS in astrocytes but not neurons promote neural impairments. In transgenic mice expressing mutant P301S human tau, oral administration of S3QEL reduced glial alterations and tauopathy and delayed premature mortality. Together, our findings suggest that IIIQ_o ROS promote disease-related astrocytic changes and aberrant astrocytic-neuronal interactions, and that S3QELs may be a novel therapeutic approach for neurodegenerative disorders.

Disclosures: D. Barnett: None. **F. Palaguachi:** None. **T.S. Zimmer:** None. **S.M. Meadows:** None. **A.G. Orr:** Other; Co-inventor on patents for treating neurological disease and other

conditions with suppressors of mitochondrial reactive oxygen species production. **A.L. Orr:** Other; Co-inventor on patents for treating neurological disease and other conditions with suppressors of mitochondrial reactive oxygen species production.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.04

Topic: B.09. Glial Mechanisms

Support: the Intramural Research Program of the National Institute of Health

Title: Morphometric analysis of glia cells in mice engineered to express human GNPTAB stuttering mutation

Authors: *A. ADECK, M. MILLWATER, S. SHEIKHBAHAEI;
Natl. Inst. of Neurolog. Disorders and Stroke, NIH, Bethesda, MD

Abstract: Stuttering, a common neurodevelopmental speech disorder, affects nearly 1% of the US adult population and is characterized by repeated interruptions in the smooth flow of speech, silent blocks, repetition, and prolongation of words. Recently it was shown that mice engineered to express a mild mutation in the *Gnptab* gene demonstrate stuttering-like vocal deficits similar to those observed in humans with the similar mutation. Although the underlying mechanisms have yet to be elucidated, it is postulated that astrocytes might have a key role in the development of stuttering disorder. In this study, we used *Gnptab*-mutant mice to study the effect of this mutation on structural characteristics of astrocytes. To do so, we performed 3D-semi-automatic reconstructions of over 400 astrocytes from 14 different brain regions associated with vocalization and used metrics such as sholl and convex-hall analysis to quantify morphometric properties of astrocytes in *Gnptab*-mutant and control mice. Our data demonstrate that in *Gnptab*-mutant mice, the structural morphology of astrocytes residing in the vocal production circuits are different from those in the control animals. For instance, astrocytes in *Gnptab*-mutant mice show less complexity in primary motor cortex (194991 ± 30508 in *Gnptab*-mutant vs. 540102 ± 89125 in control mice; $p = 0.002$), anterior cingulate cortex (55541 ± 5135 in *Gnptab*-mutant vs. 553933 ± 95849 in control mice; $p < 0.001$) and central amygdala (99456 ± 19212 in *Gnptab*-mutant vs. 341430 ± 57754 in control mice; $p < 0.001$) ($n = 10$ astrocytes per group, *Mann-Whitney U* rank test). On the other hand, we did not find a difference in structural complexity of astrocytes in regions that are not involved in vocalization. Furthermore, in our preliminary data, we found no difference in morphology of microglia in most regions analyzed between *Gnptab*-mutant and control mice. Our data further provide evidence that astrocytes may have a key role in pathophysiology of stuttering disorder and suggest that astrocytes may be the main glial cells affected by the mutation in *GNPTAB* gene that is linked to the stuttering disorder.

Disclosures: **A. Adeck:** None. **M. Millwater:** None. **S. Sheikhbahaei:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.05

Topic: B.09. Glial Mechanisms

Support: R01DK102918
RF1AG059778
Jackson Laboratory Startup Funds to KMSO

Title: Effect of high-fat diet on hypothalamic astrocytes

Authors: *T. OUELLETTE, A. KORGAN, H. SAMUEL, K. M. S. O'CONNELL;
The Jackson Lab., Bar Harbor, ME

Abstract: The continued rise of obesity and associated comorbidities in the United States highlight the critical lack of effective therapeutic interventions to maintain appropriate body weight. Current strategies fail to account for central nervous system (CNS) regulation of food intake, including diet induced hypothalamic dysregulation which affects behavior and further promotes the obese state. Changes to the CNS caused by poor diet and obesity may explain the behavioral, social, or mental difficulties of maintaining weight loss. High-fat diet (HFD) has a profound effect on the hypothalamus, including a loss of synaptic inhibition and loss of modulation by peripheral factors like leptin. This results in AgRP neurons becoming functionally disconnected from the systems that should regulate their activity and instead adopt an autonomous pattern of high output, driving food intake and sensations of hunger despite metabolic satiety. Aside from neuronal cell types, glia are known to influence several behaviors like sleep and wake cycles via modulation of synaptic signaling in the CNS; importantly, these functions are affected by high fat diet. In addition to synaptic regulation, this cell type influences blood brain barrier permeability which is a critical process for hypothalamic sensation of circulating peripheral signals. Astrocytes in the hypothalamus have only recently been investigated for their role in modulating synaptic activity during normal physiological conditions but the field is lacking in our understanding the role of hypothalamic astrocytes in pathologies like obesity. In this study, we are characterizing astrocyte recruitment, morphology, expression, and function in the arcuate nucleus of the hypothalamus (ARH) of mice that are fed a high fat diet. Here, we use genetically resilient (WSB/EiJ) and susceptible (C57Bl/6J and NZO/HtJl) mice to investigate astrogliosis and its role in facilitating changes in neuronal excitability, synaptic plasticity (and decoupling or dysregulation), and circuit connectivity of hypothalamic neurons following high-fat diet consumption. These changes in hypothalamic astrocyte activity may potentiate diet induced obesity by altering food intake behaviors and weight gain.

Disclosures: **T. Ouellette:** None. **A. Korgan:** None. **H. Samuel:** None. **K.M.S. O'Connell:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.06

Topic: B.09. Glial Mechanisms

Support: R01 NS084817
R01 DA044552
R01 DA044552-03S1
DA033966
NS060632
MH122241

Title: Psychostimulant abuse and cART worsen HIV-induced astrocyte dysfunction in HIV^+ human brains

Authors: *A. DONNER[1], L. CHEN[2], M. ASH[1], J. SCHNEIDER[1], L. AL-HARTHI[1], X.-T. HU[3];
[2]Dept. of Microbial Pathogens and Immunity, [3]Microbial Pathogens and Immunity, [1]Rush Univ. Med. Ctr., Chicago, IL

Abstract: People living with HIV/AIDS (PLWH) have an improved life expectancy due to combined antiretroviral therapy (cART); but the prevalence of HIV-associated neurocognitive disorders (HAND) persists. The mechanism underlying HAND is unknown; but it is likely related to HIV- and cART-induced neurotoxicity in the brain regions regulating neurocognition. HIV induces neurotoxicity, causing dysregulation, injury, and in severe cases, death of neurons in these brain regions; which is worsened by astrocyte dysfunction. Among infected/affected astrocytes, HIV not only dysregulates cytokines/chemokines, but also disturbs K^+ buffering (mediated by inwardly rectifying $K_{ir}4.1$ channels), glutamate uptake (mediated by excitatory amino acid transporters, GluT-1), and cell-to-cell communication (mediated by connexin 43, Cx43, a gap-junction and hemichannel protein); all promote neurotoxicity. Further study is needed to reveal how and to what extent HIV actually alters astrocytes in the human brains, which will add to the field as previous studies focusing on assessing HIV-induced astrocyte dysfunction mainly used animal or cell culture models. We previously evaluated HIV-induced changes in the protein/mRNA levels of Kir4.1 channels, GluT-1s and Cx43 in the prefrontal cortex (PFC, a key but understudied regulator of cognition) and cerebellum (unaffiliated with cognition) from post-mortem HIV+ human brains. We found that the protein levels of $K_{ir}4.1$ channels and GluT-1s were significantly decreased ($p<0.01$ and $p<0.05$, respectively); while $K_{ir}4.1$ channel mRNA levels were significantly increased ($p<0.05$), along with a trend of increase in GluT-1 and decrease in Cx43 mRNA, in the PFC of HIV^+ human brains compared to

HIV^- controls. Meanwhile, we found no significant change in the expression of such proteins/mRNAs in the cerebellum of HIV^+ human brains, suggesting that HIV-induced brain dysfunction is region-specific. Encouraged by these novel findings from human brains, we expanded the project to examine the consequential effects of HIV infection on neurons/astrocytes and their interaction in the brains of deceased individuals who were infected by HIV (HIV^+), diagnosed with HAND, while with or without chronic cART and psychostimulant abuse (cocaine or methamphetamine). HIV^- and drug-free healthy human brains were used as controls. We expect that cART and psychostimulant-induced neurotoxicity will worsen HIV-induced changes in neurons/astrocytes and their interaction, by jointly dysregulating the protein/mRNA expression of $K_{ir}4.1$ channels, GluT-1s and Cx43s to a greater extent, in addition to the consequential effects of HIV-1.

Disclosures: **A. Donner:** None. **L. Chen:** None. **M. Ash:** None. **J. Schneider:** None. **L. Al-Harthi:** None. **X. Hu:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.07

Topic: B.09. Glial Mechanisms

Support: IBS-R001-D2
2018M3C7A1056894
NRF-2020M3E5D9079742
KIST-2E30954
KIST-2E30962

Title: Astrocytic urea cycle detoxifies Aβ-derived ammonia while impairing memory in Alzheimers Disease

Authors: ***Y. JU**[1,2], M. BHALLA[2], S. HYEON[3], J. OH[4], S. YOO[4], U. CHAE[5], I.-J. CHO[5], J. KWON[2], W. KOH[2], J. LIM[2], Y. M. PARK[2], J. LEE[6], H. LEE[4], H. RYU[3], C. LEE[2];
[1]Korea Inst. of Sci. and Technol., Korea Inst. of Sci. and Technol., Seoul, Korea, Republic of; [2]Inst. for Basic Sci., Daejeon, Korea, Republic of; [4]Mol. Recognition Res. Ctr., [5]Brain Sci. Inst., [3]Korea Institie of Sci. and Technol., Seoul, Korea, Republic of; [6]Boston Univ. Sch. of Med., Boston, MA

Abstract: Alzheimer's disease (AD) is one of the foremost neurodegenerative diseases, characterized by beta-amyloid (Aβ) plaques and significant progressive memory loss. In AD, astrocytes are proposed to take up and clear Aβ plaques. However, how Aβ induces pathogenesis and memory impairment in AD remains elusive. We report that normal astrocytes show non-cyclic urea metabolism, whereas Aβ-treated astrocytes show switched-on urea cycle with upregulated enzymes and accumulated entering-metabolite aspartate, starting-substrate ammonia,

end-product urea, and side-product putrescine. Gene-silencing of astrocytic ornithine decarboxylase-1 (ODC1), facilitating ornithine-to-putrescine conversion, boosts urea cycle and eliminates aberrant putrescine and its toxic by-products ammonia, H_2O_2, and its end-product GABA to recover from reactive astrogliosis and memory impairment in AD. Our findings implicate that astrocytic urea cycle exerts opposing roles of beneficial Aβ detoxification and detrimental memory impairment in AD. We propose ODC1-inhibition as a promising therapeutic strategy for AD to facilitate removal of toxic molecules and prevent memory loss.

Disclosures: **Y. Ju:** A. Employment/Salary (full or part-time):; Institute for Science and Technology. **M. Bhalla:** None. **S. Hyeon:** None. **J. Oh:** None. **S. Yoo:** None. **U. Chae:** None. **I. Cho:** None. **J. Kwon:** None. **W. Koh:** None. **J. Lim:** None. **Y.M. Park:** None. **J. Lee:** None. **H. Lee:** None. **H. Ryu:** None. **C. Lee:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.08

Topic: B.09. Glial Mechanisms

Support: IBS-R001-D2

Title: Hypothalamic GABRA5-positive neurons control obesity via astrocytic GABA

Authors: ***M. SA**[1], E.-S. YOO[2], W. KOH[1], M. G. PARK[1], H.-J. JANG[3], Y. YANG[3], J. LIM[1], W. WON[1], J. KWON[1], M. BHALLA[1], H. AN[1], Y. SEONG[1], S. LEE[4], K. PARK[5], P.-G. SUH[3], J.-W. SOHN[2], C. J. LEE[1];
[1]Ctr. for Cognition and Sociality, Inst. for Basic Sci. (IBS), Daejeon, Korea, Republic of; [2]Biol. Sci., Korea Advanced Inst. of Sci. and Technol., Daejeon, Korea, Republic of; [3]Ulsan Natl. Inst. of Sci. and Technol. (UNIST), Ulsan, Korea, Republic of; [4]Res. Animal Resource Ctr., [5]Convergence Res. Ctr. for Diagnosis, Treatment and Care Syst. of Dementia, Korea Inst. of Sci. and Technol. (KIST), Seoul, Korea, Republic of

Abstract: The lateral hypothalamic area (LHA) regulates food intake and energy expenditure. Although LHA neurons innervate adipose tissues, the identity of neurons that regulate fat is undefined. Here we identify that GABRA5-positive neurons in LHA ($GABRA5^{LHA}$) polysynaptically project to brown and white adipose tissues in the periphery. $GABRA5^{LHA}$ are a distinct subpopulation of GABAergic neurons and show decreased pacemaker firing in diet-induced obesity (DIO) mouse model. Chemogenetic inhibition of $GABRA5^{LHA}$ suppresses energy expenditure and increases weight gain, whereas gene-silencing of GABRA5 in LHA decreases weight gain. In DIO mouse model, $GABRA5^{LHA}$ are tonically inhibited by nearby reactive astrocytes releasing GABA, which is synthesized by MAOB. Gene-silencing of astrocytic MAOB in LHA reduces weight gain significantly without affecting food intake, which is recapitulated by administration of a MAOB inhibitor, KDS2010. We propose that firing of

GABRA5^{LHA} facilitates energy expenditure and selective inhibition of astrocytic GABA is a molecular target for treating obesity.

Disclosures: **M. Sa:** None. **E. Yoo:** None. **W. Koh:** None. **M.G. Park:** None. **H. Jang:** None. **Y. Yang:** None. **J. Lim:** None. **W. Won:** None. **J. Kwon:** None. **M. Bhalla:** None. **H. An:** None. **Y. Seong:** None. **S. Lee:** None. **K. Park:** None. **P. Suh:** None. **J. Sohn:** None. **C.J. Lee:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.09

Topic: B.09. Glial Mechanisms

Title: Using antisense oligonucleotides to target the neurotoxic astrocyte state

Authors: *J. DUGAL, C. HONG, B. AHN, H. KORDASIEWICZ, F. KAMME;
Ionis Pharmaceuticals, Inc., Carlsbad, CA

Abstract: Using antisense oligonucleotides to target the neurotoxic astrocyte state
Jordan DuGal[1] Christine Hong[1], Benjamin Ahn[1], Holly Kordasiewicz[1], Fredrik Kamme[1] [1]Ionis Pharmaceuticals, Carlsbad CA 92010
Astrocytes are known to take on different phenotypes depending on context. The role of neurotoxic astrocytes in neurodegenerative disease and neuroinflammation is becoming more evident as demonstrated throughout literature. Reverting this neurotoxic state back to a homeostatic state using antisense oligonucleotide (ASO) technology could be beneficial in disease contexts. In this study, we established an in vitro neurotoxic astrocyte model in which we recapitulated two gene modules associated with the neurotoxic state and showed knockdown of module-associated targets using ASOs.
We optimized an in vitro neurotoxic astrocyte model by treating astrocytes in vitro with various cytokines and assessing expression levels of module markers using qPCR and transcriptome profiling. Our model recapitulated the expected upregulation of neurotoxic astrocyte marker C3, as well as increased expression of neurotoxicity-associated gene modules. We next evaluated ASO uptake by performing a Malat-1 ASO dose titration, in which we observed a dose-responsive reduction in Malat-1 expression. Finally, we treated neurotoxic astrocytes with ASOs against module-associated targets. Transcriptome profiling showed ASO treatment suppressed the designated target gene, compared to an inert control ASO.
Here we show that ASOs can suppress target genes in an in vitro model of neurotoxic astrocytes. These experiments demonstrate the potential for ASOs as a therapeutic to modulate astrocyte states for benefit in neurodegenerative diseases.

Disclosures: **J. DuGal:** None. **C. Hong:** None. **B. Ahn:** None. **H. Kordasiewicz:** None. **F. Kamme:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.10

Topic: B.09. Glial Mechanisms

Support: R01DA04155

Title: Using astrocyte-specific RNASeq to analyze differential expression of cytoskeletal genes in the nucleus accumbens across cocaine self-administration and abstinence

Authors: *J. P. FRANKLIN[1], K. J. REISSNER[2];
[1]UNC Neurosci. Ctr., [2]Psychology and Neurosci., Univ. of North Carolina at Chapel Hill, Chapel Hill, NC

Abstract: Cocaine abuse is a significant public health concern across the United States, as the number of cocaine-related deaths in the United States has almost tripled since 2013. Although relapse rates remain high, an FDA-approved treatment for Cocaine Use Disorder is lacking. Hence, there is a considerable need for investigations into the mechanisms that drive relapse vulnerability. Recent advances indicate that cocaine-induced structural adaptations in astrocytes may contribute to relapse vulnerability. Astrocytes are the most abundant glial cell in the brain and regulate diverse critical functions, including synaptic transmission and plasticity. Research in the Reissner lab has revealed that astrocytes in the nucleus accumbens are extensively structurally impaired following long-access (LgA, 6h/day) cocaine self-administration and prolonged (45 d) abstinence, as there is a ~40% decrease in astrocyte volume, surface area, and synaptic colocalization (Kim et al., bioRxiv 2022.04.06.487393). However, the mechanisms driving these observations are unknown. Prior literature has shown that astrocyte-abundant cytoskeletal proteins contribute to maintenance of astrocyte structure and are involved in drug-seeking behaviors. Hence, we hypothesize that cocaine-induced downregulation of astrocyte cytoskeletal dynamics contributes to the observed astrocyte structural decreases, decreased synaptic interactions, and increased drug seeking observed across abstinence. This hypothesis will be tested by investigating changes in astrocyte cytoskeletal dynamics following long-access cocaine self-administration and extended abstinence. We have validated expression of astrocyte-specific AAV5-GfaABC1D-Rpl22-HA (Addgene #111811) in rat nucleus accumbens astrocytes, and confirmed isolation of astrocyte-specific mRNAs using RNASeq. We have also prepared mRNAs from nucleus accumbens following LgA cocaine vs. saline self-administration, at withdrawal days 1 and 45, for RNASeq analyses to examine differences in the astrocyte translatome. In addition to bioinformatic analysis of differential gene expression, we will specifically test the hypothesis that astrocyte cytoskeletal gene expression is decreased following cocaine self-administration and abstinence. These results will provide a greater understanding of how cocaine abuse leads to dysfunctions in astrocyte structure and function, and how these may contribute to relapse vulnerability.

Disclosures: **J.P. Franklin:** None. **K.J. Reissner:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.11

Topic: B.09. Glial Mechanisms

Support: The Intramural Research Program of the National Institutes of Health, NINDS

Title: Disrupted iron homeostasis in mice engineered with a mutation associated with stuttering

Authors: *M. MILLWATER[1], A. ADECK[2], S. SHEIKHBAHAEI[3];
[2]Natl. Inst. of Neurolog. Disorders and Stroke, [3]NINDS, [1]NIH, Bethesda, MD

Abstract: Stuttering is a neurodevelopmental disorder characterized by involuntary disruptions in speech fluency and is linked to white matter abnormalities or dopamine hyperactivity in the basal ganglia. Recently, stuttering has been shown to link to point mutations in a gene involved in the lysosomal enzyme-targeting pathway (i.e., GNPTAB), though it remains unclear how such a mutation might relate to the stuttering phenotype. In this study, we used mice engineered with a mutation in GNPTAB gene found in humans who stutter, and found increased iron deposition in the basal ganglia of these mice. Using Perl's staining method, we stained for iron in age-matched Gnptab-mutant and control mice and analyzed iron deposition with ImageJ-Fiji software based on intensity of the chemical stain and percent area of deposition. There was an increase in iron deposition in the medial lateral (10.30 ± 1.785 in Gnptab-mutant vs. 1.252 ± 1.785 in control), dorsal lateral (10.99 ± 1.304 in Gnptab-mutant vs. 1.831 ± 1.304 in control), and central striatum (9.241 ± 1.569 n Gnptab-mutant vs. 3.525 ± 1.569 in control) of Gnptab-mutant mice ($p = 0.002$, $p = 0.001$, $p = 0.011$; $n = 4$ animals per group; Unpaired t test). Further, these iron deposits localized predominantly with regional astrocytes when Perl's staining was combined with an astrocyte-specific marker S100ß. Astrocytes, star-shaped glial cells, were found to be less complex in the Gnptab-mutant mice (704539 ± 91059 in Gnptab-mutant vs. 144010 ± 91059 in control $p < 0.0001$; $n = 10$ astrocytes per group). There was no cellular loss noted across cell populations, as determined by quantification of cell bodies on ImageJ-Fiji. These findings support the hypothesis that iron homeostasis is altered in Gnptab-mutant mice, and that regional astrocytic morphology differences may have implications for the traditional circuit-modulatory role of these glial cells. Here we hypothesize a relationship between a missense mutation in a cellular housekeeping mouse *Gnptab* gene, iron homeostasis, astrocytes, and ultimately the stuttering phenotype that has long remained elusive.

Disclosures: **M. Millwater:** None. **A. Adeck:** None. **S. Sheikhbahaei:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.12

Topic: B.09. Glial Mechanisms

Support: NIH grant R03AG063264
NIH grant R21NS109918
NIH grant R01DA048815
NIH grant R01DA048815-03S1

Title: A longitudinal study of a subset of post-inflammatory reactive astrocytes using a novel genetic tool

Authors: W. AGNEW-SVOBODA[1], ***T. UBINA**[1], Z. FIGUEROA[3], Y.-C. WONG[4], E. VIZCARRA[4], B. ROEBINI[5], E. WILSON[6], T. A. FIACCO[7], M. RICCOMAGNO[2];
[1]Molecular, Cell, and Systems Biol., [2]Molecular, Cell and Systems Biol., Univ. of California, Riverside, Riverside, CA; [3]Neurosci. Grad. Program, UCR, Riverside, CA; [5]Molecular, Cell, and Systems Biol., [4]Univ. of California, Riverside, CA; [6]Univ. of California, Riverside Biomed. Sci. Grad. Program, Riverside, CA; [7]Cell Biol. & Neurosci., UC Riverside, Riverside, CA

Abstract: Astrocytes are vital support cells, partnering with neurons to ensure proper brain function and play a critical role in the brain's innate immune response. In brain injury, infection, and disease, astrocytes reprogram into a reactive state which alters many of their most fundamental processes with beneficial or detrimental outcomes. In the case of acute inflammation, markers of reactivity that astrocytes display soon after the initial insult are no longer observed 1 month after inflammation was induced. A long-standing question in the field is whether downregulation of reactive astrocyte (RA) markers during resolution of inflammation is because these astrocytes revert back to a nonreactive state or die and are replaced. This question has proven difficult to answer mainly because existing genetic tools using traditional astrocyte promoters cannot distinguish between healthy vs. reactive astrocytes. Here we describe the generation and characterization of a novel inducible genetic tool that can be used to specifically target, label, and manipulate a subset of reactive astrocytes in the diseased or injured brain. We provide evidence for the utility of this approach to study RAs in several models of brain inflammation. The biggest advantage of this transgenic model is that it can be used to perform longitudinal analysis of reactive astrocyte morphology, function and gene expression. Analysis of these transgenic mouse brains during and after the resolution of acute inflammation revealed that RAs that are labeled soon after acute inflammation survive for at least one month. Furthermore, most morphological features and expression of markers associated with astrogliosis revert back to baseline after 1 month in the labeled cells, while one morphological hallmark of reactivity remains unchanged after 1 month. Our data indicate that the previously observed downregulation of RA markers one month after inflammation is likely due to changes in gene expression and not because of programmed cell death. Overall, our findings suggest that cellular changes associated with astrogliosis after acute inflammation can be temporary and largely reversible.

Disclosures: **W. Agnew-Svoboda:** None. **T. Ubina:** None. **Z. Figueroa:** None. **Y. Wong:** None. **E. Vizcarra:** None. **B. Roebini:** None. **E. Wilson:** None. **T.A. Fiacco:** None. **M. Riccomagno:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.13

Topic: B.09. Glial Mechanisms

Support: Pritzker Neuropsychiatric Disorders Research Consortium Fund LLC

Title: Astrocyte dysfunction in bipolar disorder and schizophrenia differs from major depressive disorder

Authors: *A. MEDINA[1], M. H. HAGENAUER[1], D. M. KROLEWSKI[1], E. HUGHES[1], L. C. THEW FORRESTER[1], M. WASELUS[1], D. M. WALSH[2], E. RICHARDSON[1], C. A. TURNER[1], P. CARTAGENA[3], R. C. THOMPSON[1], M. P. VAWTER[4], B. BUNNEY[5], A. SEQUEIRA[5], R. MYERS[6], J. D. BARCHAS[7], F. LEE[8], A. F. SCHATZBERG[9], W. E. BUNNEY[10], H. AKIL[1], S. J. WATSON[1];
[1]Univ. of Michigan, Univ. of Michigan, Ann Arbor, MI; [2]Univ. of California - Irvine, SOM, Univ. of California - Irvine, SOM, Irvine, CA; [3]Univ. Of California Irvine, Irvine, CA; [4]UC Irvine Schl Med., UC Irvine Schl Med., Irvine, CA; [5]UCI, UCI, Irvine, CA; [6]Hudson Alpha, Huntsville, AL; [8]Weill Cornell Med. Col., [7]Weill Cornell Med. Col., New York, NY; [9]Stanford Univ., Stanford, CA; [10]Univ. of California Irvine Dept. of Psychiatry and Human Behavior, Univ. of California Irvine Dept. of Psychiatry and Human Behavior, Irvine, CA

Abstract: Astrocytes are fundamental in maintaining and modulating brain function at the cellular level. Their activity regulates different aspects of neurotransmission, and dysfunction of the glial syncytium may underlie the pathophysiology of mental illness. Previous studies from our group showed that several aspects of the glial function are affected by Major Depressive Disorder (MDD), most of them related to the modulation of glutamate neurotransmission and the resetting of the synaptic environment after its excitatory activity, functions that are mediated by astrocytes. Those results showed the downregulation of genes involved in glutamate re-uptake, post-excitatory water and potassium siphoning, and the gap junction elements necessary to maintain the syncytial function of astrocytes. This study explored the effect of two other diagnoses, Bipolar Disorder (BP) and Schizophrenia (SZ), on the astrocyte network. We hypothesize that astrocyte function may also be affected in these two diseases, however, since the etiological theories for BP and SZ differ from those described for MDD, there could also be differences in the glial mechanisms associated with these disorders. Methods: Human brain samples from the frontopolar cortex (Brodmann area 10, BA10) were obtained through the University of California, Irvine (UCI)-Pritzker Brain Bank. The subjects included in the schizophrenia and bipolar disorder groups met diagnostic criteria from the Diagnostics and

Statistical Manual of Mental Disorders (DSM-V). The control group showed no evidence of psychiatric or neurological disorders. Covariates that could affect gene expression were accounted for including gender, age, and postmortem interval. All subjects used in the study had cerebellar tissue pH above 6.5 and agonal factor scores (AFS) of zero. After performing RNA extraction and adequate quality control, we used TaqMan® Gene Expression Custom Array Cards to perform qPCR analysis of genes related to astrocyte function. The results were then bolstered by performing a meta-analysis of publicly released BA10 microarray data to find points of convergence with our qPCR results. Results: our study showed alterations of genes related to astrocyte function in BP and SZ compared to controls. However, the altered molecules did not overlap with the list of candidate genes based on previously reported findings in MDD. There was a strong correlation between the BP and SZ results, suggesting pathophysiological commonalities between the two diagnoses.

Disclosures: A. Medina: None. **M.H. Hagenauer:** None. **D.M. Krolewski:** None. **E. Hughes:** None. **L.C. Thew Forrester:** None. **M. Waselus:** None. **D.M. Walsh:** None. **E. Richardson:** None. **C.A. Turner:** None. **P. Cartagena:** None. **R.C. Thompson:** None. **M.P. Vawter:** None. **B. Bunney:** None. **A. Sequeira:** None. **R. Myers:** None. **J.D. Barchas:** None. **F. Lee:** None. **A.F. Schatzberg:** None. **W.E. Bunney:** None. **H. Akil:** None. **S.J. Watson:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.14

Topic: B.09. Glial Mechanisms

Support: International Graduate School in Molecular Medicine Ulm (IGradU)

Title: Toll-like receptor activation up-regulates the cystine/glutamate antiporter system x_c^- in cortical murine astrocytes via NFkB and through auto- and paracrine mechanisms dependent on TNFα and IL1α/β

Authors: T.-D. VOSS[1], F. APPELT[2], P. MAHER[3], ***J. LEWERENZ**[4];
[1]Clin. Chem., Univ. of Heidelberg, Ulm, Germany; [2]Ulm Univ., Ulm, Germany; [3]Salk Inst. for Biol. Studies, La Jolla, CA; [4]Univ. of Ulm, Univ. of Ulm, Ulm, Germany

Abstract: Background: The cystine/glutamate antiporter system x_c^- imports cystine into cells while exporting glutamate. It is composed of two subunits with xCT being specific to this antiporter. In the healthy brain, xCT is mainly expressed in astrocytes. However, upon inflammatory activation, it is prominently upregulated in microglia. Thus, in the context of neuroinflammation, release of glutamate via system x_c^- might induce neuronal excitotoxicity. **Aim:** The aim of this study was to characterize the pathways that lead to upregulation of xCT mRNA and system x_c^- activity upon inflammatory activation of astrocytes. **Methods:** Murine primary microglia-free cortical astrocytes were treated with tumor necrosis factor α (TNFα) or

the toll-like receptor (TLR) 3 and 4 agonists bacterial lipopolysaccharide (LPS) and polyinosinic:polycytidylic acid (Poly I:C), respectively. System x_c^- activity was measured as homocysteate-inhibitable ^{3}H-glutamate uptake. For immunoblotting, astrocytic membranes were used for xCT, cytosolic and nuclear extracts for (phospho)p65. For luciferase reporter promoter assays, astrocytes were transiently transfected using constructs containing xCT promoter fragments of different lengths. Anti-TNFα antibodies and an interleukin 1 (Il1) receptor antagonist (RA) were used to block the activity of both cytokines released after stimulation with LPS and PolyI:C. Realtime reverse transcriptase PCR was used to quantify mRNA expression. **Results:** LPS, PolyI:C as well as TNFα increased the mRNA and protein levels of xCT as well as system x_c^- activity. LPS and PolyI:C caused a more robust induction than TNFα. LPS and PolyI:C, but not TNFα, increased both TNFα as well as Il1α/β mRNA levels. Co-incubation of astrocytes with either TNFα- neutralising antibodies or Il1-RA reduced TLR3/4-mediated induction of system x_c^- as did both inhibitors when added to conditioned medium from activated microglia. xCT promoter luciferase reporter assays showed similar patterns for all three stimuli although none induced classical transcripts downstream of Nrf2 or ATF4. However, all three inflammatory stimuli induced nuclear p65 phosphorylation. Furthermore, the IκB kinase 1/2 inhibitor BMS-345541 blocked induction of system x_c^- activity by LPS and PolyI:C as well as TNFα. **Conclusion:** Upon TLR3/4 activation, system x_c^- is upregulated via induction of xCT mRNA via NFκB but not Nrf2 or ATF4. In addition, TLR3/4 activation induces TNFα and IL1α/β, which in turn amplify the effects of TLR3/4 activation via auto- and paracrine mechanisms.

Disclosures: T. Voss: None. **F. Appelt:** None. **P. Maher:** None. **J. Lewerenz:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.15

Topic: B.09. Glial Mechanisms

Support: R01AA016959
R01AA016959-11S1

Title: Upregulation of Vimentin Reveals Timing of Astrocyte Reactivity in Female Rats Following Binge Ethanol Administration

Authors: *S. P. GUERIN, K. NIXON;
Pharmacol. & Toxicology, The Univ. of Texas at Austin Col. of Pharm., Austin, TX

Abstract: Individuals with alcohol use disorders (AUD), of which an increasing number are women, frequently experience cognitive deficits related to alcohol-induced structural changes in the brain. Astrocytes may play a role in mediating these structural changes, and in some ethanol administration models adopt a reactive phenotype through altered expression of intermediate

filaments like GFAP and vimentin. In a study with male rats in a four-day binge model of AUD, striking increases in vimentin were found to peak in many brain regions a week following cessation of ethanol administration. However, temporal and regional characteristics of vimentin upregulation are still unknown in female rats. Therefore, adult female Sprague-Dawley rats (n=8 per group) were administered ethanol in a four-day binge administration paradigm of alcohol dependence. Rats received either an ethanol diet (25% w/v in vanilla Ensure Plus) or an isocaloric control diet (vanilla Ensure Plus and dextrose) three times per day for four days. Ethanol dose was adjusted based on behavioral indicators of intoxication, scored from 1 to 5, which averaged 1.4 ± 0.1 (ataxia) resulting in a mean ethanol dose of 9.6 ± 1.4 g/kg/day and a mean blood ethanol concentration of 373.1 ± 97 mg/dl, all similar to past studies in males. Withdrawal behavior observations took place 18 hours following the last ethanol dose and averaged 1.2 ± 1.0 on a 4-point scale, with a maximum of 2.7 ± 1.0, indicating most rats experienced withdrawal tremors. Animals were sacrificed via transcardial perfusion 2, 7, or 14 days after their last dose of ethanol, and brains were removed, post-fixed, and sectioned for immunohistochemistry. Cortical regions were quantified for vimentin immunoreactivity and reported as area fraction of immunoreactivity. The time course of vimentin immunoreactivity was similar between females and that previously reported in males, with dramatic vimentin expression increased at 7 days of abstinence, and little to no vimentin increase at either the 2- or 14-day timepoints. Like reported in males, vimentin immunoreactivity was noted in the insular, piriform, and perirhinal/entorhinal cortices. Vimentin immunoreactivity was also found in the somatosensory cortex and the basolateral amygdala, two regions that had not been reported in males. These data suggest that while the time-course of vimentin immunoreactivity is similar in males and females, regional variations between sexes may exist.

Disclosures: **S.P. Guerin:** None. **K. Nixon:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.16

Title: WITHDRAWN

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.17

Topic: B.09. Glial Mechanisms

Support: NRF Grant 2017R1D1A1B05028221

Title: L-serine-dependent tamoxifen toxicity to cultured human retinal Müller cells

Authors: *J. CHOI[1], J.-Y. KOH[1,2], Y. YOON[3];
[1]Neural Injury Res. Center, Biomed. Res. Ctr., Asan Inst. for Life Sciences, Asan Med. Ctr., SEOUL, Korea, Republic of; [2]Neurol., Asan Med. Center, Univ. of Ulsan Col. of Med., Seoul, Korea, Republic of; [3]Ophthalmology, Asan Med. Center, Univ. of Ulsan Col. of Med., SEOUL, Korea, Republic of

Abstract: Tamoxifen (TAM) given as an antiestrogen agent in breast cancer often causes retinopathy. Of interest, TAM retinopathy shares some morphological characteristics with idiopathic macular telangiectasia type 2 (MacTel2). An increasing body of evidence indicates that dysfunction and/or loss of Müller cells likely plays a key role in the pathogenesis of MacTel2. Interestingly, mutations in the gene of an L-serine synthetic enzyme (3-Phosphoglycerate dehydrogenase; PHGDH) have been causally linked to a subset of MacTel2 (Eade et al, Nat Metab. 2021; Gantner ML et al, NEJM, 2019; Scerri TS et al, Nat Genet. 2017). These findings raise a possibility that serine-dependent Müller cell dysfunction may contribute also to TAM retinopathy. Hence, in the present study, we examined 1) whether Müller cells in vitro and in vivo are more sensitive to tamoxifen toxicity than other retinal cells, and 2) whether TAM toxicity to Müller cells is linked to an abnormality in L-serine metabolism. Twenty-four hours after TAM treatment (1-10 μM), cultured Müller cells showed a significant decrease in cell viability (19.57 ± 2.34 %) as assessed by the MTT assay, as compared to cultured retinal pigment epithelial (RPE) cells (ARPE-19) or photoreceptors (661w). Concomitantly TAM treatment markedly reduced L-serine concentrations in Müller cells. Intriguingly, supplementation of L-seine (0.5 - 5 mM) blocked the TAM cytotoxicity in a concentration-dependent manner. In C57BL/6 mice, intravitreal TAM treatment (0.1 μg/μl, 3 μl/eye) altered the expression of vimentin and glutamine synthesis (GS), markers of Müller cells, and increased the number of TUNEL positive cells in the inner nuclear layer (INL) at 5 days after the treatment. In serine-treated retinal tissue, however, all the above changes induced by TAM treatment were substantially ameliorated. Our results suggest that Müller cells may be the main target of TAM toxicity, which may involve aberrant L-serine metabolism as in a subset of MacTel2. Further studies may be needed to determine which step in the L-serine metabolism is altered with TAM treatment.

Disclosures: J. Choi: None. **J. Koh:** None. **Y. Yoon:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.18

Topic: B.09. Glial Mechanisms

Support: P30 AG066514
U01AG046170
RF1AG058469
RF1AG059319
R01AG061894
U01AG046170
Neurodegenerative Diseases Consortium MD Anderson

Title: An integrative multimodal sequencing approach uncovers regionally-restricted reactive astrocyte sub-states common to neuroinflammation and Alzheimer's disease

Authors: *P. HASEL[1], E. L. CASTRANIO[5], R. D. KIM[1], S. GANDY[5,6], M. E. EHRLICH[5,7,8], S. A. LIDDELOW[1,2,3,4];
[1]Neurosci. Inst., [2]Dept. of Neurosci. & Physiol., [3]Dept. of Ophthalmology, [4]Parekh Ctr. for Neurol., NYU Grossman Sch. of Med., New York, NY; [5]Dept. of Neurol., [6]Dept. of Psychiatry and Alzheimer's Dis. Res. Ctr., [7]Dept. of Genet. and Genomic Sci., [8]Dept. of Pediatrics, Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Astrocytes undergo a 'reactive' transformation in acute and chronic diseases of the brain, leading to changes in gene expression and function. Recent advances in multimodal, integrative sequencing has highlighted how heterogenous these reactive transformations can be. Using single cell RNA-seq and spatial transcriptomics (ST) we investigated the response of astrocytes to acute inflammation and chronic disease mouse models. We report that astrocytes fall into distinct subtypes that occupy specific positions in the brain. These subtypes can undergo unique reactive transformations during acute inflammation induced by the endotoxin lipopolysaccharide (LPS) that are likely controlled by the location of the astrocyte and the type of initiating insult. We find an interferon-induced astrocyte super-responder that emerges early in this reactive transformation and occupies strategic positions in the brain, including the surface of the brain and the interventricular foramen. The interferon-responsive reactive subtype is present in a myriad of acute and chronic neurodegenerative diseases including mouse models of Alzheimer's disease (AD), demyelination, and acute stab wound injury. By generating extensive ST on APP/PS1 brain sections we report that plaques cause a local shift in gene expression that is modulated by microglia, and we propose novel markers for plaque-specific reactivity. We find that interferon-induced astrocytes are selectively found around plaques. Employing sub-spot level differential gene expression and spatial ligand-receptor analyses we uncover a whole range of plaque-restricted putative multi-cell type interactions that suggest an extensive upregulation of inflammatory, phagocytic and lysosomal activity around the plaque. Ultimately, profiling region-restricted reactive transformations of astrocytes in chronic neurodegeneration will facilitate the discovery of therapeutic targets to modulate the disease trajectory.

Disclosures: P. Hasel: None. **E.L. Castranio:** None. **R.D. Kim:** None. **S. Gandy:** None. **M.E. Ehrlich:** None. **S.A. Liddelow:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); AstronauTx Ltd.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.19

Topic: B.09. Glial Mechanisms

Support: NIH Grant P30 AG08051
NIH Grant T32AG052909
Blas Frangione Foundation
Neurodegenerative Diseases Consortium from MD Anderson
Alzheimer's Research UK
Gifford Family Neuroimmune Consortium as part of the Cure Alzheimer's Fund
Alzheimer's Association

Title: Astrocytes undergo subtype-specific gene expression changes in Alzheimer's disease

Authors: *M. R. O'DEA[1], J. S. SADICK[1], P. HASEL[1], T. DYKSTRA[1], A. FAUSTIN[2], S. A. LIDDELOW[3];
[1]Neurosci. & Physiol., [2]Dept. of Pathology, [3]Neurosci. Inst., NYU Grossman Sch. of Med., New York, NY

Abstract: Neuronal dysfunction and degeneration in Alzheimer's disease (AD) is increasingly recognized as a non-cell intrinsic process. Rather, this dysfunction is accompanied by changes in surrounding cell types, including glial cells like astrocytes. While previous studies indicate astrocytes can influence neuronal dysfunction and degeneration, these potential contributions remain understudied. Single-cell RNA sequencing provides a powerful tool for exploring these contributions; however, defining transcriptional changes in astrocytes in Alzheimer's disease has remained challenging due to low capture rates and high intersubject variability. To improve understanding of glial heterogeneity in AD, we used single-nucleus RNA sequencing (snRNA-seq) to profile over 80,000 transcriptomes from post-mortem prefrontal cortex from AD and age-matched non-symptomatic (NS) donors with apolipoprotein (*APOE*) ε2/3 genotype. Using fluorescence-assisted nuclei sorting to isolate LHX2+/NeuN- nuclei, we achieved higher proportions of astrocytes than previous studies, allowing for higher-powered investigation of astrocyte transcriptional changes. We identified heterogeneous astrocyte subtypes in both NS and AD samples based on gene expression, and discovered both common and subtype-specific gene expression changes in AD patients compared to NS patients. GO analysis of these transcriptional changes indicates cluster-specific gain- and loss-of-function alterations in astrocytes in AD, highlighting potential pathways influencing disease progression. We further profiled these human astrocyte subtypes by exploring their anatomical positions using existing sequencing-based human and mouse spatial transcriptomic datasets. By examining modules of cluster-specific marker genes, we localize the astrocyte subtypes from our snRNA-seq data to distinct cortical regions, finding astrocyte gene signatures largely separate between white matter and gray matter. Using these spatial datasets, we also found a subset of the astrocyte transcriptional changes we identified in AD were also induced in a mouse model of inflammation, suggesting these changes are attributable to inflammation in AD. This work describes heterogeneity in astrocytes in AD using snRNA-seq, identifies AD-associated transcriptional changes with

potential relevance for disease progression, and explores the putative spatial restriction of astrocyte subtypes using existing spatial transcriptomics datasets, providing a new resource for examining glial contributions to Alzheimer's disease.

Disclosures: **M.R. O'Dea:** None. **J.S. Sadick:** None. **P. Hasel:** None. **T. Dykstra:** None. **A. Faustin:** None. **S.A. Liddelow:** Other; S.A.L. is a founder of AstronauTx Ltd and a member of its scientific advisory board.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.20

Topic: B.09. Glial Mechanisms

Support: National Science Foundation Graduate Research Fellowship 2019273004
Anonymous donors
NYU Grossman School of Medicine
NIH/NEI R01EY033353

Title: Molecular profiling of ischemic injury-induced reactive astrocytes

Authors: *R. D. KIM[1], P. W. FRAZEL[1], P. HASEL[1], U. A. RUFEN-BLANCHETTE[1], A. X. GUO[1], A. E. MARCHILDON[1], S. A. LIDDELOW[1,2,3,4];
[1]Neurosci. Inst., [2]Dept. of Neurosci. & Physiol., [3]Dept. of Ophthalmology, [4]Parekh Ctr. for Interdisciplinary Neurol., NYU Sch. of Med., New York, NY

Abstract: Astrocytes undergo robust gene expression changes in response to a variety of perturbations such as infection, disease, and acute insults, including ischemic injury. How these changes are affected by time and sex, as well as how heterogeneous and spatially distinct various reactive astrocyte populations are, remain unclear. In this study, we performed single nuclei RNAseq of ~122,000 forebrain astrocytes isolated from Aldh1l1-EGFP/Rpl10a mice at 1, 3, and 14 days after ischemic injury induced by Rose Bengal photothrombosis. We found that injury induces a widespread and temporally diverse response across many astrocyte subtypes. We also identified clusters unique to injury-induced reactive astrocytes, including an interferon-responsive population that is rapidly induced at 1 day and persists up to 14 days post-injury; this population, in addition to expressing many interferon-response genes (e.g. *Igtp*, *Iigp1*, *Ifit3*, among others), appears to be Stat1/Stat2 dependent according to gene regulatory network analysis. Another unique injury-induced reactive astrocyte cluster expresses proliferative markers (e.g. *Mki67, Top2a, Pola1*, etc.) and is present only at 3 days post-injury. Spatial transcriptomics highlight that these reactive astrocyte populations are spatially restricted and reside in locations that are likely functionally important in the stabilization and resolution stages following injury. Together, these datasets provide a powerful resource for probing injury-induced reactive astrocyte heterogeneity and can be used to guide functional interrogation of

biologically meaningful reactive astrocyte substates to understand their pro- and anti-reparative functions following acute injuries like stroke.

Disclosures: **R.D. Kim:** None. **P.W. Frazel:** None. **P. Hasel:** None. **U.A. Rufen-Blanchette:** None. **A.X. Guo:** None. **A.E. Marchildon:** None. **S.A. Liddelow:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); AstronauTx Ltd..

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.21

Topic: B.09. Glial Mechanisms

Support: NIH grant AG061775

Title: Ab40 and Ab42 adopt distinct fibril structures that differentially stimulate astrocytes and microglia

Authors: ***X. ZHU**[1], J. SCHRADER[1], B. IRIZARRY[2], S. SMITH[2], W. E. VAN NOSTRAND[1]; [1]Univ. of Rhode Island, Univ. of Rhode Island, Kingston, RI; [2]Stony Brook Univ., Long Island, NY

Abstract: Fibrillar amyloid β-protein (Aβ) deposits in brain, which are primarily composed of Aβ40 or Aβ42 peptides, are key pathological features of Alzheimer's disease and related disorders. Structural studies on mature Aβ40 fibrils formed in solution have suggested they adopt a two stranded (U-shaped) structure whereas studies on Aβ42 fibrils typically show a three-stranded structure. Fibrillar amyloid deposits in brain promote activation of astrocytes and microglia although the underlying mechanisms are still not clear. Here, we treated primary astrocyte and microglia cells with Aβ40 or Aβ42 fibrils, and bulk RNA sequencing was performed. According to the Pearson's correlation result, astrocytes and microglia cells undergo a great number of Aβ peptides-induced changes in gene expression and Aβ42 and Aβ40 peptides showed similar effects on glia cells compared with the control group. To elucidate the gene changes after Aβ peptides treatment, we conducted the differential gene expression analysis to compare Aβ42 and Aβ40 glia cells. The expression data were set with limits of ≥5-fold increase in expression and $p < 0.05$. A greater number of genes were differentially expressed by Aβ42 treated glia cells (186 and 1909 genes in astrocyte and microglia, respectively) compared with the Aβ40 treated glia cells (129 and 238 genes in in astrocyte and microglia, respectively). Immunohistochemical analysis validated our RNAseq data by performing in an AD rat model with parenchymal fibrillar Aβ42 deposits confirming the expression of MMP9, MMP12, PAI-1 and C1r in plaque-associated microglia and iNOS, GBP2 and C3D in plaque-associated astrocytes. To better understand Aβ peptides-induced gene changes in glia cells, we performed Ingenuity Pathway Analysis. These analyses further highlighted that Aβ42 treatment up-

regulated cellular activation pathways and immune response pathways in glia cells, including protein ubiquitination, sirtuin signaling and tumor microenvironment pathways in astrocyte, and granulocyte adhesion and diapedesis, wound healing and IL17 signaling pathways in microglia. Interestingly, we found that astrocytes treated with Aβ peptides up-regulated a larger number of A1 reactive genes compared with A2 reactive genes, and microglia treated with Aβ peptides up-regulated more M1 state genes than M2 state genes. Together, these studies show that the distinct structures of Aβ40 and Aβ42 fibrils differentially stimulate the gene expression profile of glia cells and that Aβ42 fibrils are more potent stimulators that could provide a detailed insight in the pathogenesis of AD and related amyloid-depositing disorders.

Disclosures: X. Zhu: None. **J. Schrader:** None. **B. Irizarry:** None. **S. Smith:** None. **W.E. Van Nostrand:** None.

Poster

527. Reactive Astrocytes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 527.22

Topic: B.09. Glial Mechanisms

Support: 1P20GM121176-01

Title: Antipsychotics differentially alters exosome-secreted and inflammation-related microRNA in neurons and glia

Authors: *S. K. AMOAH[1,2], B. RODRIGUEZ[3], L. JANTZIE[4], N. MELLIOS[5];
[1]Anat. and Cell Biol., Univ. of Cape Coast, CAPE COAST, Ghana; [2]Neurosciences, [3]Dept. of Neurosciences, Univ. of New Mexico, Albuquerque, NM; [4]Neonatology Division, Pediatrics, Johns Hopkins Sch. of Med., Baltimore, MD; [5]Dept. of Neurosciences, Sch. of Med., Albuquerque, NM

Abstract: Exosome mediated transport is an exciting mode of transport that influences neuronal and glial genes via their microRNA (miRNA) cargo as a novel. This emerging role of exosome-secreted miRNAs in the regulation of glutamate gene expression and their relevance for psychosis-associated conditions. Using mature miRNA profiling and quantitative PCR (qPCR) in the orbitofrontal cortex we uncovered that miR-223, an exosome-secreted miRNA that targets glutamate receptors, was increased at the mature miRNA level in the OFC of schizophrenia (SCZ) and bipolar disorder (BD) patients with positive history of psychosis at the time of death and was inversely associated with deficits in the expression of its targets glutamate ionotropic receptor NMDA-type subunit 2B (GRIN2B) and glutamate ionotropic receptor AMPA-type subunit 2 (GRIA2). Furthermore, changes in miR-223 levels in the OFC were positively and negatively correlated with inflammatory and GABAergic gene expression, respectively. Moreover, miR-223 is enriched in astrocytes and secreted via exosomes, and antipsychotics olanzapine and haloperidol had differential effects on the cellular and exosomal localization

fractions. Furthermore, addition of astrocytic exosomes in neuronal cultures resulted in a significant increase in miR-223 expression and a notable reduction in Grin2b and Gria2 mRNA levels, which was strongly inversely associated with miR-223 expression. Lastly, inhibition of astrocytic miR-223 abrogated the exosomal-mediated reduction in neuronal Grin2b expression. Ongoing experiments are investigating the effects of other pharmaceutical agents in vivo.

Disclosures: S.K. Amoah: None. **B. Rodriguez:** None. **L. Jantzie:** None. **N. Mellios:** A. Employment/Salary (full or part-time):; Autophagy Inflammation and Metabolism Center of Biomedical Research Excellence, University of New Mexico Health Sciences Center, Albuquerque, NM, USA. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); N.M. have a financial interest as a co-founder of Circular Genomics Inc. and are inventors of patents related to the use of circRNAs for brain disease diagnostics.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.01

Topic: C.01. Brain Wellness and Aging

Support: DoD Grant GW160151
DoD Grant GW170023
CURE Grant 4100083087
CURE Grant 4100072545
NIH Grant 1R01NS115977
NIH Grant R01NS28785
NIH Grant R01NS118117

Title: Gulf War veteran derived cerebral organoids display multifaceted pathological defects in Gulf War Illness

Authors: *K. CASE[1], P. L. YATES[1], X. SUN[1], K. SULLIVAN[2], P. W. BAAS[1], L. QIANG[1]; [1]Neurobio. and Anat., Drexel Univ. Col. of Med., Philadelphia, PA; [2]Boston Univ. Sch. of Publ. Hlth., Boston Univ. Sch. of Publ. Hlth., Boston, MA

Abstract: Approximately one third of the veterans who fought in the 1991 Gulf War suffer from Gulf War Illness (GWI), which comprises a plethora of symptoms, including cognitive deficits. Mounting evidence indicates that GWI was caused by exposure to subthreshold levels of organophosphate pesticides and nerve agents coupled with physical stressors of the battlefield. Seeking to overcome shortcomings of rodent models for GWI, we recently established a bank of human induced pluripotent stem cells from veterans suffering from GWI and from similarly exposed veterans who remained healthy. With the presumption that these cells reset the clock prior to the veterans being exposed, we treated neurons differentiated from these cells with a GW

toxicant regimen consisting of the organophosphate diisopropyl fluorophosphate (a sarin analog) and cortisol (to mimic battlefield stress), which we adapted from previous work on rodent models. Having documented effects of the toxicant regimen on various cellular features and functions, we have now sought to differentiate the cells into three-dimensional multi-cellular organoids that mimic the structure of the human brain. For this, we established cerebral organoid cultures from two veterans, one with GWI and one healthy control. Treating the organoids with the GW toxicant regimen resulted in an increase in total tau with enhanced tau phosphorylation, decreased microtubule stability, enhanced astrocyte activation, and altered neurogenesis. Some of these alterations were more pronounced in the organoids derived from the veteran with GWI compared to those derived from the healthy control veteran. The observed cellular effects are consistent with the cognitive decline suffered by the veterans. Taken together, these results provide the GWI field with a powerful new model for elucidating the cellular etiology of the disease and for testing potential therapies. In addition, they set the stage for a personalized medicine approach for individualized treatment of veterans with GWI, which is important because different soldiers were exposed to different toxicant exposures and because each soldier came into the battlefield with different genetic and epigenetic dispositions for neurodegenerative diseases.

Disclosures: **K. Case:** None. **P.L. Yates:** None. **X. Sun:** None. **K. Sullivan:** None. **P.W. Baas:** None. **L. Qiang:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.02

Topic: C.01. Brain Wellness and Aging

Support: NIH T32 Grant AG000096
NIH T32 Grant AG073088
NIH P30 Grant AG066519

Title: Deep learning characterization of the choroid plexus spatial pathome reveals differences with age and sex, but not Alzheimer's disease

Authors: M. J. NEEL[1], B. A. JOHNSON[1], P. D. CHANG[2], *E. S. MONUKI[1];
[1]Pathology and Lab. Med., [2]Radiological Sci., UC Irvine Sch. of Med., Irvine, CA

Abstract: The choroid plexus (ChP) plays an important role in brain homeostasis, but is relatively understudied. In particular, many pathologies of the ChP and how they vary with age and disease lack thorough characterization. Characterizing the pathologies of ChP can help us understand ChP dysfunction in aging and disease and help validate disease models using stem cell-derived ChP cells. Recently, deep learning (DL) technology has seen increasing use in biomedical research, offering new opportunities to characterize these pathologies at a speed,

scale, and consistency not previously feasible. Using DL models, we characterized three ChP pathologies: Biondi bodies (BBs), lipid vesicles (LVs), and fibrosis in autopsy samples from over a hundred individuals and analyzed how these pathologies change with age, sex, and Alzheimer's disease (AD). We developed custom DL models based on ResNet, RetinaNet, and UNet architectures to quantify each pathology from either thioflavin S or hematoxylin & eosin-stained whole slides images to assess prevalence and spatial distributions. We found that DL model-generated data were able to replicate manual data. BBs and fibrosis were found to increase with age, albeit with different temporal relationships, and to appear before the earliest AD-related biomarkers. These pathologies were also found to have sex differences, with females having less pathology. Interestingly, none of the pathologies had AD-related differences. LVs and fibrosis were found to be decreased in the fourth ventricle vs lateral ventricle ChP. Within ventricular regions, all pathologies had clustered, non-random distributions. Lastly, fibrosis impacted other pathologies, with fibrotic regions having fewer BBs and LVs. Our results reveal that many ChP pathologies appear relatively early in life, possibly contributing to brain neurodegeneration later in life, and that ChP pathologies spatially interact with each other.

Disclosures: **M.J. Neel:** None. **B.A. Johnson:** None. **P.D. Chang:** None. **E.S. Monuki:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.03

Topic: C.01. Brain Wellness and Aging

Support: NIH/NIA R15AG073947
Creighton Summer Undergraduate Research Fellowships

Title: Impairment of choroid plexus epithelial cells upon age-related genetic deficiency.

Authors: *T. SANCHEZ, G. D. KING;
Biol., Creighton Univ., Omaha, NE

Abstract: Aging is influenced by both genetic and environmental factors. Genetically, brain aging is underpinned by changes to only a small percentage of expressed genes. Defining the earliest distinctions between normal brain aging and neurodegenerative disease requires an understanding of how age-related regulation of genes affects the brain. Utilizing a model of premature brain aging driven by deficiency of a naturally age-downregulated gene (klotho-deficiency), we reported that rapid onset of memory impairment occurs alongside changes to neuronal structure and function. Herein, we sought to determine if stochastic changes to neurons might be caused by dysfunction of choroid plexus epithelial cells. Age-related changes to choroid plexus epithelia are sufficient to induce age-like dysfunction. We characterized the choroid plexus epithelia of model mice before and after the onset of memory impairment. Using immunohistochemistry and Western blot, we measured protein expression and localization as

metrics of choroid plexus epithelial cell health. Analysis was grouped by choroid plexus functional activity either measuring proteins important in cerebrospinal fluid production or blood:cerebrospinal fluid barrier function (n=6+/group, males and females included, compared to age-matched wild-type controls). Further electron microscopic analysis considered overall structure of cells and extracellular matrix. Most model phenotypes are only measured after the onset of memory impairment (~7 weeks of age). Assessment of the choroid plexus at this age revealed choroid plexus-specific decreased expression of all transporter proteins measured but only minor changes to junction or extracellular matrix proteins. Structurally, cell number was not different, but volume and appearance of choroid plexus epithelial cells were more similar to advanced age than age-matched controls. Additionally, when we analyzed younger brains, we measured decreased transporter protein expression in most but not all the transporters found changed at the older age. Molecular phenotypes at this age, before the onset of memory impairment, have not previously been reported. Altogether, our data suggest that dysfunction of the choroid plexus occurs before any neuronal structural or functional change is measured. Age-related changes to protein expression can drive the slow failure of supporting cells, like the choroid plexus epithelia. Support cell maintenance may provide a novel target to predict brain health and prevent neurodegenerative disease.

Disclosures: **T. Sanchez:** None. **G.D. King:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.04

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant AG064895
NIH Grant AG076235

Title: Loss of neuronal HDAC9 impairs cognitive function and synaptic plasticity

Authors: Y. LEI[1], Y. BAI[1], Y. CHEN[1], H. ZHANG[1], M. GUO[1], N. WEINTRAUB[2], X.-Y. LU[1]; [1]Dept. of Neurosci. & Regenerative Med., [2]Vascular Biol. Ctr., Med. Col. of Georgia at Augusta Univ., Augusta, GA

Abstract: Alzheimer's disease (AD) is an age-related neurodegenerative disorder that causes memory loss and cognitive decline. Epigenetic mechanisms involving histone deacetylases (HDACs) play an important role in age-related neurodegeneration. Among the 11 zinc-dependent HDACs, HDAC9 is the most abundant isoform in the brain. However, its function in the central nervous system remains largely unknown. The goal of this study was to determine whether HDAC9 plays a role in brain aging and age-related cognitive decline. We found that HDAC9 mRNA and protein were expressed exclusively in neurons, and expression levels of HDAC9 decreased with aging in the hippocampus. Global HDAC9 knockout mice exhibited impaired

spatial working memory and object recognition memory in the spontaneous alternation in Y-maze test and novel object recognition test, respectively. In the three-chamber social test, HDAC9 knockout mice showed impaired social recognition memory. Furthermore, loss of HDAC9 resulted in impaired hippocampal long-term potentiation. Selective deletion of HDAC9 in the hippocampal CA1 of adult HDAC9flox/flox mice was sufficient to induce cognitive deficits. These results suggest that age-dependent downregulation of neuronal HDAC9 underlies age-related cognitive decline and synaptic dysfunction.

Disclosures: **Y. Lei:** None. **Y. Bai:** None. **Y. Chen:** None. **H. Zhang:** None. **M. Guo:** None. **N. Weintraub:** None. **X. Lu:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.05

Topic: C.01. Brain Wellness and Aging

Support: Garrison Institute on Aging/TTUHSC
Newby Family

Title: The role of the highly amyloidogenic cystatin-related epididymal spermatogenic (CRES) and CRES subgroup members in sex-specific learning and memory

Authors: *A. GOMEZ, P. GROZDANOV, A. HEWETSON, J. D. BAILOO, G. A. CORNWALL;
Texas Tech. Univ. Hlth. Sci. Ctr., Lubbock, TX

Abstract: Amyloids are highly ordered cross β-sheet aggregates that are typically associated with disease states such as neurodegeneration and diabetes. However, growing evidence shows that some amyloids carry out biological roles including as structural scaffolds and signaling complexes and are categorized as functional amyloids. We previously demonstrated an extracellular amyloid matrix surrounds sperm in the normal mouse epididymal lumen and has proposed functions in sperm protection and maturation. The epididymal amyloid matrix is composed of several members of the highly amyloidogenic reproductive CRES subgroup of cystatin cysteine protease inhibitors suggesting it is a complex functional amyloid. In the mammalian brain extracellular matrix (ECM) specialized mesh-like structures called perineuronal nets (PNNs) surround select subsets of neurons and are important participants in neuron-neuron communication and plasticity which makes them key players in memory formation and maintenance. We hypothesize that CRES and CRES subgroup members are also found in the brain ECM as part of the PNN structure and are important participants in learning and memory processes. Preliminary RT-PCR showed CRES and other subgroup members are present in the hippocampus and other brain regions from adult male mice. Western blot analysis of sequentially extracted adult mouse hippocampus show monomer and higher molecular weight,

possibly aggregated, forms of CRES and other subgroup members are enriched in the urea soluble/PNN fraction compared to other populations of the brain ECM. Furthermore cognitive-behavioral studies between adult wild-type (WT) mice and a global CRES knockout (KO) mouse model highlight that male, but not female, KO mice display impairments in learning in a 2-choice water-escape task as well as behavioral inflexibility during reversal (they perseverate on the previously learned escape location) when the location of the platform to escape is changed. These findings suggest CRES and CRES subgroup members are part of the normal mouse brain ECM and are necessary for memory processes in potential association with PNNs.

Disclosures: **A. Gomez:** None. **P. Grozdanov:** None. **A. Hewetson:** None. **J.D. Bailoo:** None. **G.A. Cornwall:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.06

Topic: C.01. Brain Wellness and Aging

Support: Orville Edward Egbert, M.D. Endowment fund

Title: Aging-related changes in cholinergic neurons of the olfactory pathway

Authors: *P. ABLANEDO MORALES[1], P. SABANDAL[3], K.-A. HAN[2];
[2]Biol. Sci., [1]Univ. of Texas at El Paso, El Paso, TX; [3]Biol. Sci., Univ. of Texas, El Paso, El Paso, TX

Abstract: Acetylcholine signaling in the brain is important for multiple neural processes such as learning, memory, sleep and wake, and stress response. Learning, memory, and sleep regulation decrease with aging, and its underlying mechanism is not well understood. We hypothesize that the aforementioned decrease in cognitive functions is due to dysfunctional cholinergic activity that is aging sensitive. To test this hypothesis, we examined changes in acetylcholine neurons in the well-studied *Drosophila* olfactory circuit across four different ages: 4 days (young), 2 weeks (middle), 4 weeks (early old), and 8 weeks (old). We measured differences in the expression of choline acetyltransferase (ChAT; the rate limiting enzyme for acetylcholine biosynthesis) and axonal and synaptic markers in the first- (olfactory sensory neurons), second- (projection neurons), and third-order (mushroom body neurons) neurons of the olfactory pathway. Our preliminary results show increased marker immunoreactivity at the axons of first order neurons for both males and females at the early old age. Furthermore, ChAT immunoreactivity was increased in males but decreased in females at the early old age. In the second order neurons, however, marker and ChAT immunoreactivities were increased in both males and females at the early old age. These results suggest that there may be a decline in cholinergic signaling with aging, which may trigger a compensatory mechanism, resulting in increases in ChAT and axonal markers. Furthermore, changes in ChAT and axonal markers occur early in the olfactory

pathway and the changes are sexually dimorphic in *Drosophila*. The insight obtained from this study will help us better understand the changes in the cholinergic system that are responsible for aging-related cognitive decline and how they differ between males and females.

Disclosures: P. Ablanedo morales: None. **P. Sabandal:** None. **K. Han:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.07

Topic: C.01. Brain Wellness and Aging

Support: DOD Research & Education Program
US Army Research Office xTech HBCU Program
UNC-Pembroke Faculty Research Grant

Title: Altered proteostasis and corresponding synaptopathy are commonalities across dementia risk-enhancing brain vulnerabilities including seizure-related neuronal activity, toxin exposures, traumatic injuries, and blast-mediated neurocompromise

Authors: *B. A. BAHR[1], M. F. ALMEIDA[2,3];
[1]Univ. of North Carolina - Pembroke, Pembroke, NC; [2]UNC Pembroke, Pembroke, NC; [3]Univ. of North Carolina Wilmington, Wilmington, NC

Abstract: As the brain ages, intricate mechanisms underlying memory encoding become challenged, often with gradual development of mild cognitive impairment (MCI) indicators. Much evidence points to age-related compromise of protein clearance mechanisms as disrupting neuronal integrity (Farizatto et al. 2017 PLoS ONE 12:e0182895; Tecalco-Cruz et al. 2022 Mol Cell Biochem 477:915), often including synaptic pathology which is the best correlate of Alzheimer's disease (AD) and related dementias. Altered proteostasis and the corresponding synaptopathy are commonalities among dementia risk-enhancing brain vulnerabilities, including seizure-related excitotoxic neuroactivity (Karanian et al. 2007 JPET 322:1059; Caba et al. 2021 J Cell Mol Med 25:9011), toxin exposures (Farizatto et al. 2019 Sci Rpts 9:6532), neurotrauma, and military blast exposure (Almeida et al. 2021 Brain Pathol 31:e12936), all of which trigger events of or susceptibility to proteinopathy and cognitive dysfunction. Our comparative analyses also found protein accumulation pathology and synaptic decline to be associated events across a broad spectrum of experimental models including i) subtoxic exposure to excitotoxins, ii) induction of modest to mild seizures in which synaptic marker reductions correlated with seizure severity, and iii) exposure to glutamatergic and cholinergic neurotoxins. The resultant reductions in the neuropilar staining of important synaptic markers were similar to those produced by proteostatic stress mediated by failures of the ubiquitin-proteasome and/or autophagy-lysosomal systems - both systems being known for gradual compromise with age. The different vulnerabilities elicit the general features of a pathogenic cascade comprised of altered protein

chemistries, intracellular formation of abnormal configurations and deposits, disrupted tubulin dynamics, microtubule destabilization, transport failure, and progressive synaptopathy. Our recent work also used a rodent model of MCI (Hwang et al. 2019 Inter J Mol Sci 20:4432), addressing whether promoting proteostasis reduces age-related risks to cognitive health. Young adult (3 months) and MCI Fischer rats (12-14 months) were assessed to confirm cognitive decline in the middle-aged rats. Early results indicate that a specific extract treatment that boosts protein clearing pathways caused a two-fold enhancement of learning behavior in a passive avoidance paradigm as compared to MCI rats fed the control food. To conclude, gradual collapse of proteostasis is the likely root of the MCI to dementia continuum that is driven by those risk factors contributing to AD-type synaptopathy.

Disclosures: B.A. Bahr: None. **M.F. Almeida:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.08

Topic: C.01. Brain Wellness and Aging

Support: R01AG050721
RF1AG054000

Title: Exophers are components of mammalian neurobiology in health and disease

Authors: *I. SIDDIQUE[1], J. DI[1], C. K. WILLIAMS[1,2], D. MARKOVIC[3], H. V. VINTERS[1,2], S. TAWAKLEY[1], C. ZANG[1], A. VAISHAMPAYAN[1], K. MCCAULEY[1], G. BITAN[1,4,5]; [1]Dept. of Neurology, David Geffen Sch. of Med., [2]Dept. of Pathology and Lab. Med., [3]Dept. of Medicine, Div. of Gen. Intrnl. Medicine, and Hlth. Services Research,, [4]Brain Res. Inst., [5]Mol. Biol. Inst., UCLA, Los Angeles, CA

Abstract: The failure of cellular clearance mechanisms results in the accumulation of toxic protein aggregates in neurodegenerative disorders. The ubiquitin-proteasome system and autophagy-lysosomal pathway are the two main clearance mechanisms in eukaryotic cells. Recently, a third mechanism, expulsion of large extrusions, called exophers, has been discovered in *C. elegans*. Exophers are large, membrane-enclosed extrusions in which damaged organelles and protein aggregates are removed from the cytoplasm, presumably for phagocytosis and degradation by immune or immune-like cells. Exophers are initially connected to the parent cell via a nanotube and eventually disconnect. We discovered for the first time that exophers exist in mammalian neurons during analysis of mouse brain sections stained for hyperphosphorylated tau. We also observed this phenomenon in primary neurons from tauopathy mice and human neuroblastoma cells. Subsequently, we discovered exophers also in human brain, demonstrating that this phenomenon is conserved from nematodes to humans. In line with the proposed role of exophers, we observed disease-related protein aggregates, e.g., Aβ42 and tau, colocalized with

exophers in Alzheimer's disease and other tauopathy brains and in primary neurons from tauopathy mouse models. The number of exophers increased as an adaptive response under proteostatic stress or as a response to oxidative or osmotic stress. Moreover, we found that tau-positive exophers tend to have larger sizes than tau-negative exophers. However, we also found rare, innate exophers in healthy human brain and primary neurons from wild-type mice, suggesting that in addition to their adaptive role in removal of undesired cellular materials, under normal conditions exophers may be involved in sharing resources between neighboring cells. In line with this hypothesis, SH-SY5Y neuronal cultures showed exophers in contact with neighboring cells or with exophers of neighboring cells, and exophers connected by nanotubes to two or even three cells. Moreover, we found that exophers may contain exosomes, suggesting that they may participate in cell-to-cell short range communication possibly via targeted transfer of exosomes. In the context of neurodegenerative diseases, our data suggest that exophers may be a double-edge sword - contributing to the degradation of toxic aggregated proteins, but possibly also to their spread throughout the brain.

Disclosures: I. Siddique: None. **J. Di:** None. **C.K. Williams:** None. **D. Markovic:** None. **H.V. Vinters:** None. **S. Tawakley:** None. **C. Zang:** None. **A. Vaishampayan:** None. **K. McCauley:** None. **G. Bitan:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.09

Topic: C.01. Brain Wellness and Aging

Title: Ap-4 regulates neuronal lysosome composition, function, and transport via regulating export of critical lysosome receptor proteins at the trans-golgi network

Authors: *S. GOWRISHANKAR[1], P. MAJUMDER[2], D. EDMISON[2], C. RODGER[3], E. REID[3];
[1]Univ. of Illinois at Chicago, Chicago, IL; [2]UIC, Chicago, IL; [3]CIMR, Cambridge, United Kingdom

Abstract: The adaptor protein complex-4 or AP-4 is known to mediate autophagosome maturation through regulating sorting of transmembrane cargo such as ATG9A at the Golgi. There is a need to understand AP-4 function in neurons, as mutations in any of its four subunits cause a complex form of hereditary spastic paraplegia (HSP) with intellectual disability. While AP-4 has been implicated in regulating trafficking and distribution of cargo such as ATG9A and APP, little is known about its effect on neuronal lysosomal protein traffic, lysosome biogenesis and function. In this study, we demonstrate that in human iPSC-derived neurons AP-4 regulates lysosome composition, function and transport via regulating export of critical lysosomal receptors, including Sortilin 1, from the trans-Golgi network to endo-lysosomes. Additionally, loss of AP-4 causes endo-lysosomes to stall and build up in axonal swellings potentially through

reduced recruitment of retrograde transport machinery to the organelle. These findings of axonal lysosome build-up are highly reminiscent of those observed in Alzheimer's disease as well as in neurons modelling the most common form of HSP, caused by *spastin* mutations. Our findings implicate AP-4 as a critical regulator of neuronal lysosome biogenesis and altered lysosome function and axonal endo-lysosome transport as an underlying defect in AP-4 deficient HSP.

Disclosures: S. Gowrishankar: None. **P. Majumder:** None. **D. Edmison:** None. **C. Rodger:** None. **E. Reid:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.10

Topic: C.01. Brain Wellness and Aging

Support: 1 RF1AG068292

Title: The G-quadruplex DNA ligand pyridostatin modulates transcription in neurons

Authors: *R. DIAZ ESCARCEGA[1], A. URAYAMA[1], S. P. MARRELLI[1], D. MONCHAUD[2], L. D. MCCULLOUGH[1], A. TSVETKOV[1,3];
[1]Neurol., Univ. of Texas, McGovern Med. Sch., Houston, TX; [2]Inst. de Chimie Moléculaire, ICMUB CNRS UMR 6302, UBFC, 21078 Dijon, France, Dijon, France; [3]UTHealth Consortium on Aging, the Univ. of Texas McGovern Med. School,, Houston, TX

Abstract: Non-canonical base pairing between four guanines (G) within single-stranded G-rich sequences leads to the formation of G-quartet, and self-stacking of G-quartets results in the formation of a columnar four-stranded DNA structure known as G-quadruplexes (G4s or G4-DNA). G4-DNA regulates a broad variety of DNA-dependent processes including transcription, replication, and telomere function. We previously showed that, in primary cultured neurons, small molecule G-quadruplex ligands downregulate transcription of the Brca1 gene and the autophagy gene Atg7. Here, we performed genome-wide gene expression analysis (RNAseq) to identify genes modulated by a G4-DNA ligand, pyridostatin (PDS). PDS promotes the stabilization of G4 structures, thus allowing us to define genes directly or indirectly responsive to G4 regulation. Our results demonstrate that 3,188 genes are differentially expressed in neurons treated with PDS out of a total of 17,140 genes with measured expression. 1573 genes are downregulated and 1445 genes are upregulated, which provides invaluable insights into the dysregulated pathways, notably involving a network of genes regulating p53 signaling, axon guidance, and MAPK signaling. Our data showing the high number of genes responsive to PDS treatment (18.6%) highlights the potential and magnitude for similar transcriptional regulation by endogenous G4-DNA ligands. Taken together, our analyses suggest that G4-DNA may be an epigenetic-like mechanism that regulates gene expression in neuronal cells.

Disclosures: **R. Diaz Escarcega:** None. **A. Urayama:** None. **S.P. Marrelli:** None. **D. Monchaud:** None. **L.D. McCullough:** None. **A. Tsvetkov:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.11

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant 5R01MH123748-02
NIH Grant 1P50MH122379-01A1

Title: Impaired autophagy in high passaged human embryonic kidney 293 (HEK) cells

Authors: *S. SALVATORE, H. SHU, A. BENZ, C. F. ZORUMSKI, S. MENNERICK; Psychiatry, Washington Univ. Sch. of Med., St. Louis, MO

Abstract: Neuropsychiatric and neurodegenerative disorders are correlated with cellular stress, and the process of macroautophagy (herein referred to as autophagy) may represent an important protective pathway to maintain cellular homeostasis and functionality. Given recent evidence that some novel psychiatric treatments, such as the neuroactive steroid (NAS) allopregnanolone (AlloP, Brexanolone), may target autophagy, we designed a stably transfected HEK cell model to assay NAS effects on autophagy. We used pcDNA3-GFP-LC3-RFP-LC3Δ3, a fluorescent ratiometric autophagy assay. GFP/RFP levels are inversely correlated with autophagic flux; GFP-LC3 is targeted for autophagic degradation while RFP-LC3Δ3 is a cytosolic control. During assay development, we discovered decreasing autophagy induction with successive experiments. Live-cell imaging 24 hours post treatment revealed that 500 nM Torin1, a potent autophagy inducer, and 1 μM AlloP, increased GFP/RFP fluorescence ratios relative to control in 3 consecutive experiments. We hypothesized that changes associated with cell passage number may be responsible for diminished autophagy induction over time. To test our hypothesis, we compared autophagy induction in low (<P10) and high passage (P15+) HEK cells using Torin1. As hypothesized, pharmacological autophagy induction, measured as a GFP/RFP ratio percentage of control, was impaired in higher passage cells (84.3 ± 4.8%) compared with lower passage cells (55.1 ± 3.6%, $p = 0.0007$, unpaired t-test, $n = 3$ per condition in 2 independent replicates). Moreover, the impaired GFP/RFP decrease in higher passage cells was driven by higher GFP fluorescence (25.3 ± 3.2 Torin1 GFP units in high passage cells vs. 18.1 ± 1.4 in low passage cells, $n = 3$ per condition in 2 independent replicates), indicative of less GFP quenching/degradation. Levels of basal autophagy were also altered with passage number (2.2 ± 0.1 control GFP/RFP in passage 18 vs. 1.8 ± 0.1 in passage 5, $p = 0.0197$, two-way ANOVA with Sidak's multiple comparisons, $n = 3$ per condition). We conclude that passage number in immortalized HEK cells affects autophagy and opens possibilities of studying the transcriptional/epigenetic factors regulating autophagic flux, including lysosomal pH or protease dysfunction. Our results could be consistent with previous literature showing that autophagy is

decreased with age; however, cellular senescence, implicated in prior work, is likely not involved given the immortalized nature of the HEK cell line.

Disclosures: **S. Salvatore:** None. **H. Shu:** None. **A. Benz:** None. **C.F. Zorumski:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Sage Therapeutics. F. Consulting Fees (e.g., advisory boards); Sage Therapeutics. **S. Mennerick:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.12

Topic: C.01. Brain Wellness and Aging

Support: YK: NIH Grant R01NS108407
YK: NIH Grant RF1MH12460501
PJD: NIH Grant R01NS078168
PJD: NIH Grant R01NS079737
QZ: AHA Career Development Award 935961

Title: Structural and morphological remodeling of the mural and vascular network in the aged mouse brain

Authors: *H. C. BENNETT[1], D. J. VANSELOW[2], Q. ZHANG[3], P. J. DREW[4], Y. KIM[1]; [1]Neural and Behavioral Sci., [2]Pathology, Penn State Univ., Hershey, PA; [3]Ctr. for Neural Engineering, Dept. of Engin. Sci. and Mechanics, The Pennsylvania State Univ., University Park, PA; [4]Ctr. for Neural Engineering, Dept. of Engin. Sci. and Mechanics, Dept. of Neurosurg., The Pennsylvania Univ., University Park, PA

Abstract: Cerebrovascular dysfunction contributes to both normal aging and neurodegenerative disease. Mural cells, including both vascular smooth muscle cells (vSMCs) and pericytes, are critical vasculature components that dynamically regulate blood flow. Pericytes are also implicated in the pathophysiology of Alzheimer's disease. Despite the links between mural cells, cerebrovascular dysfunction, and aging, it is unclear how mural populations across brain regions are impacted by aging.Here, we first asked whether normal aging was associated with reduced capillary pericyte population density. We utilized serial two-photon tomography (STPT) and cell type mapping with young (2 month) and aged (18 month) Pdgfrb-Cre;Ai14 male and female mice to examine capillary pericyte population density changes across all brain regions (n=10 per age). To investigate changes in mural cell types and the vasculature in the same brain, we utilized iDISCO-based whole brain clearing and immunolabeling paired with Light Sheet Fluorescence microscopy (LSFM) as depicted in Figure 1. This strategy enabled us to comprehensively and simultaneously label the whole vasculature, vSMCs, and pericytes (Fig 1a,d), to interrogate changes associated with advanced age, by comparing young (2 month) and

aged (24 month) C57BL/6 mice (n=5 per age).Our cell mapping results in Pdgfrb-Cre;Ai14 mice demonstrate that normal aging is associated with reduced capillary pericyte density in the basal forebrain and differential changes across isocortical layers, which was further validated in immunolabeling of C57BL/6 mice. Moreover, in C57BL/6 mice, normal aging is associated with striking structural changes in vSMCs, particularly in the arterioles of the lateral association areas of the isocortex (Fig 1c), and at the large arteries of the Circle of Willis. The results of this study suggest that mural cell populations and cerebrovasculature are heavily impacted by normal aging, yet the effects vary vastly across brain regions.

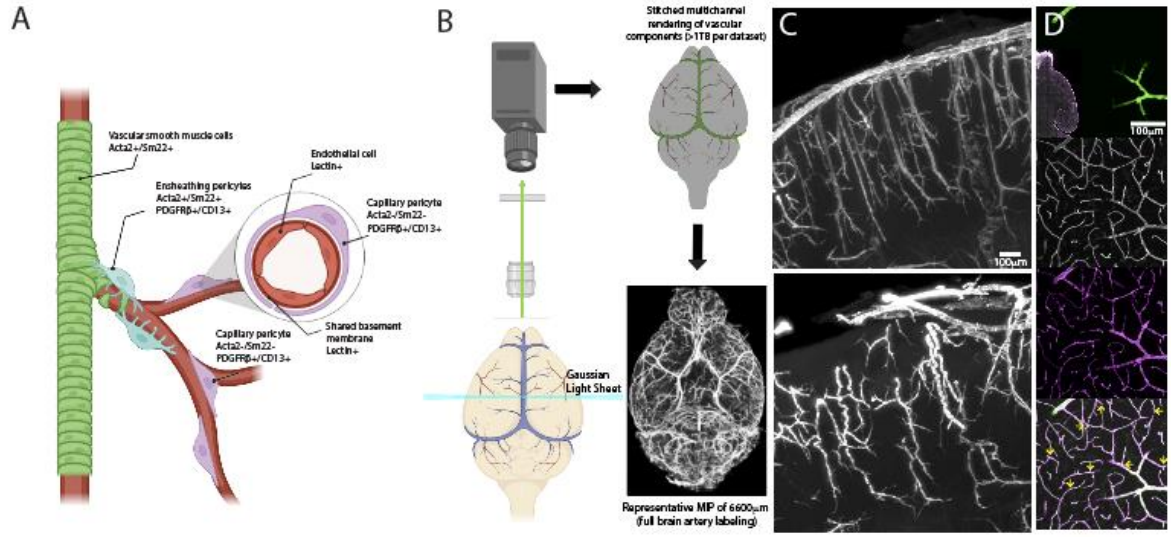


Figure 1: Methods used to investigate changes in mural cell types and vasculature in normal aging.

Disclosures: H.C. Bennett: None. **D.J. Vanselow:** None. **Q. Zhang:** None. **P.J. Drew:** None. **Y. Kim:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.13

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant R01AG071787

Title: Effect of rejuvenating factors on mouse cortical dendritic spine dynamics

Authors: *H. LEE, J. LU, Y. ZUO;
Molecular, Cell and Developmental Biol., Univ. of California Santa Cruz, Santa Cruz, CA

Abstract: Aging-related cognitive decline and neurodegeneration are prevalent with increased life expectancy. It is suggested that aging-related elevation in neuroinflammation contributes to the impairment of synaptic circuits, leading to cognitive deficits. Previous studies have shown that the plasma level of oxytocin (OT) decreases with aging, whereas TGF-β signaling, which activates activin-like kinase 5 (Alk5), increases in the aged brain. Systemic delivery of a cocktail composed of OT and an Alk5 inhibitor (OTA5i) rapidly and robustly enhances hippocampal neurogenesis, reduces neuroinflammation, and improves cognitive performance of aged mice in sensory discrimination and novel object recognition. However, how OTA5i affects the structural dynamics of synapses, which may underlie the cognitive improvement, remains unclear. In this work, we used in vivo two-photon microscopy to examine the effects of OTA5i treatment on the turnover (i.e., formation and elimination) of dendritic spines (postsynaptic sites of most excitatory synapses) of layer 5 pyramidal neurons in the somatosensory cortex of adult Thy1-YFP-H mice. We injected OTA5i or vehicle intraperitoneally for 7 days and imaged the same set of dendritic segments before the treatment course and one day after the last injection. We found that while spine elimination was unaffected by OTA5i treatment, spine formation was significantly elevated. We are currently extending our study to aged mice, in order to correlate the synaptic changes with behavioral improvements.

Disclosures: H. Lee: None. **J. Lu:** None. **Y. Zuo:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.14

Topic: C.01. Brain Wellness and Aging

Support: Yulgilbar Foundation PhD Top Up Scholarship
JR and JO Wicking Trust

Title: Parvalbumin interneurons and perineuronal nets in aging and neurodegeneration

Authors: *E. A. BUCHER, Y. V. DOUST, J. F. MCMANUS, O. G. HOLLOWAY, J. M. COLLINS, A. E. KING, J. C. VICKERS, J. M. ZIEBELL, M. T. K. KIRKCALDIE; Wicking Dementia Res. And Educ. Ctr., Univ. of Tasmania, Hobart, Australia

Abstract: A fine balance of excitation and inhibition is required for cognitive processing, likely mediated by fast-spiking parvalbumin interneurons (PV-INs) and their enveloping perineuronal nets (PNNs), which respectively coordinate inhibitory synchrony and regulate plasticity. There is evidence to suggest that these structures are altered in conditions which disrupt cognition and behaviour, though these changes are not thoroughly understood. In this study, we explored the structural responses of PNNs and PV-INs in models of neurodegeneration that physically perturb the neuropil, including amyloidosis and diffuse traumatic brain injury (TBI). We fluorescently labelled PV-INs, PNNs, and amyloid-β in coronal sections of mouse brains. Images were

segmented using ImageSURF in FIJI to quantitate the extent of these labels in regions of cortex and hippocampus. Linear mixed-effects models incorporating age/timepoint, region, and sex or genotype were constructed to analyse each stain. In male APPswe/PSEN1dE9 (APP/PS1) and C57Bl/6J mice, ages 6 and 24 months (n = 2-4) we observed a main effect of age on PNN percentage area, with both genotypes demonstrating an overall loss from 6 to 24 months of age, $p = 0.029$, most notably in primary visual cortex. No changes in PV were noted. Next, we examined the gross response of PV-INs and PNNs to trauma. In 3-month-old male and female C57Bl/6J mice, a midline fluid percussion injury model was used to replicate a moderate TBI. Brains were collected at 3 hours and 1-, 3-, or 7-days post-injury and compared to naïve animals (n = 3 per sex and timepoint). A time × sex × region interaction was observed for the percentage area of PV ($p = 0.003$), driven by changes in the dentate gyrus (DG), where males demonstrated significantly more PV at days 1 and 3 post-injury. Significant time × sex, and time × region interactions were observed in PNN coverage, indicating that PV expression and the PNN are altered in the days following TBI, particularly in DG and CA1, but typically return to naïve levels by 7-days post-injury. Together, these results demonstrate a range of responses of PV-INs and PNNs in the context of physical perturbations of the cortex and hippocampus, which would be expected to have functional consequences for inhibition and plasticity. In the future, these techniques will be applied in a model of functional perturbation, exploring the effects of altered sleep and plasticity in APP/PS1 mice to examine how these structures might change when faced with both physiological and physical perturbations.

Disclosures: **E.A. Bucher:** None. **Y.V. Doust:** None. **J.F. McManus:** None. **O.G. Holloway:** None. **J.M. Collins:** None. **A.E. King:** None. **J.C. Vickers:** None. **J.M. Ziebell:** None. **M.T.K. Kirkcaldie:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.15

Title: WITHDRAWN

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.16

Topic: C.01. Brain Wellness and Aging

Support: NIH/NIA R01AG019719
NIH/NIA R37AG028730
Glenn Foundation for Medical Research
KNRF 2012R1A6A3A03040476

Title: Dna break-induced loss of epigenetic information as a cause of neuronal aging

Authors: *J.-H. YANG[1], P. T. GRIFFIN[1], J. A. AMORIM[1], M. HAYANO[2], L. A. RAJMAN[1], A. R. PFENNING[3], D. A. SINCLAIR[1];
[1]Harvard Med. Sch., Harvard Med. Sch., Boston, MA; [2]Keio Univ., Tokyo, Japan; [3]Computat. Biol. Dept., Carnegie Mellon Univ., Pittsburgh, PA

Abstract: There are numerous hallmarks of aging in mammals, but no unifying cause has been identified. In budding yeast, aging is associated with a loss of epigenetic information that occurs in response to genome instability, particularly DNA double-strand breaks (DSBs). Mammals also undergo predictable epigenetic changes with age, including alterations to DNA methylation patterns that serve as epigenetic "age" clocks, but what drives these changes is unknown. Using a transgenic mouse system called "ICE" (for inducible changes to the epigenome), we show that a tissue's response to non-mutagenic DSBs reorganizes the epigenome and accelerates molecular, physiological, and cognitive changes normally seen in older mice, including the advancement of the epigenetic clock. These findings implicate DSB-induced epigenetic drift as a conserved cause of aging from yeast to mammals.

Disclosures: J. Yang: None. **P.T. Griffin:** None. **J.A. Amorim:** None. **M. Hayano:** None. **L.A. Rajman:** None. **A.R. Pfenning:** None. **D.A. Sinclair:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); https://sinclair.hms.harvard.edu/david-sinclairs-affiliations. F. Consulting Fees (e.g., advisory boards); https://sinclair.hms.harvard.edu/david-sinclairs-affiliations. Other; https://sinclair.hms.harvard.edu/david-sinclairs-affiliations.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.17

Topic: C.01. Brain Wellness and Aging

Support: MRC DTP iCASE ST11722

Title: Generation of a progerin-driven accelerated ageing model in human neural stem cells to uncover novel geroneuroprotectors

Authors: *D. ROCK[1], M.-D. RUEPP[1,2], S. THURET[1];
[1]Basic and Clin. Neurosciences, [2]UK Dementia Res. Inst., King's Col. London, London, United Kingdom

Abstract: Adult hippocampal neurogenesis (AHN) is associated with aspects of learning, memory and cognition and its dysfunction is linked to ageing and neurodegenerative disease. Long-lived quiescent neural stem cells (qNSCs) are susceptible to the accumulation of damage, leading to stem cell dysfunction and declines in neurogenesis. Intrinsic hallmarks of ageing found in old qNSCs are partially responsible for this decreased capacity to activate. Progerin, a mutant variant of lamin A responsible for Hutchinson-Gilford Progeria Syndrome, can recapitulate some key features of ageing in neurons and NSC. Although small molecules have been demonstrated to boost neurogenesis and cognition in model organisms, it is still unclear whether these findings apply to humans. Therefore, we established a human hippocampal progenitor cell line (HPC0A07/03C) that conditionally expresses mature progerin cDNA to be utilised as an AHN-relevant drug screening platform. We show that progerin-expressing cells have a decreased rate of proliferation and lower percentage of ki67 expression as compared to non-expressing cells ($ki67^{+}prog^{+}$ 50.3% 5.83, $ki67^{+}prog^{-}$ 60.8% 6.47, paired T-test, $P < 1 \times 10^{-4}$). Additionally, we analysed in vivo and in vitro transcriptomic data measured in young (<40 years) and old (>60 years) human hippocampal tissue, controlling for sex, post-mortem interval and batch effects in statistical analyses, as well as a dataset of progerin-induced aged human neurons. Using connectivity mapping (CMap) to measure the degree of matching/anti-matching of gene pairs between gene signatures, we scored each dataset against a library of drug-induced gene signatures and cross-referenced the results. We uncovered 1,271 shared drug hits, which showed enrichment for lifespan-extending compounds reported in the longevity compound database, DrugAge (52/1,271, Fisher's exact test, $P < 1.2 \times 10^{-7}$). There were 7 consistently high-scoring compounds ($T > 90$) across the ageing datasets, of which two - AS-601245 and albendazole - had previously been demonstrated to be neuroprotective in rodents, validating the approach. Future work will seek to understand the mechanisms of deficient proliferation in progerin-NSCs, assessing hallmarks of ageing related to proteostasis, senescence, mitochondrial function and epigenetic regulation. Drugs identified from CMap will be tested for their ability to rescue growth impairments in progerin-NSCs and improve underlying cellular features of NSC ageing. Promising drugs discovered through this work will be worthy of study in more complex in vitro and in vivo ageing models.

Disclosures: **D. Rock:** None. **M. Ruepp:** None. **S. Thuret:** None.

Poster

528. Brain Aging: Molecular and Cellular Changes

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 528.18

Topic: C.01. Brain Wellness and Aging

Support: Tau Consortium (Rainwater charitable foundation)

Title: Human Histopathologic Brain Age Estimation via Deep Multiple Instance Learning

Authors: *G. A. MARX[1], A. T. MCKENZIE[2], J. KAUFFMAN[3], D. G. KOENIGSBERG[2], C. MCMILLAN[4], K. FARRELL[2], J. F. CRARY[5];
[1]Icahn Sch. of Med., New York, NY; [2]Icahn Sch. of Med. at Mount Sinai, New York, NY; [3]Icahn Sch. of Med. at Mount Sinai, Long Island City, NY; [4]Univ. of Pennsylvania, Philadelphia, PA; [5]Pathology, Icahn Sch. of Med. At Mount Sinai, New York, NY

Abstract: The discordance between cellular and chronologic aging is useful for understanding diseases in the brain and biology at large. One established method for analyzing the factors that contribute to brain aging is to train machine learning models that predict an individual's age based on an MRI image of their brain. While this approach has yielded important insights, it is inherently constrained by the information provided by an MRI. However, age-dependent pathologic change has the potential to be assessed at greater detail histologically. Histopathological whole slide images provide more granular information regarding cellular structure, injury, dysfunction, and morphology. Recent technological advances in whole slide image digitization has paved the way for large scale analysis of histologic data via artificially intelligent based computer vision techniques. Here, we leveraged a large novel collection of uniformly processed digitized human post-mortem brain tissue sections to create a histological brain age estimation model. We further investigated the effect of cognitive impairment and exogenous stress on the model. This was accomplished by developing a context-aware attention-based deep multiple instance learning model on 702 human brain tissues sections (age range 50-110 yr) from the hippocampus stained with Luxol Fast Blue counterstained with hematoxylin and eosin (LH&E) on a brain age estimation task. This model estimated brain age within a mean absolute error of 5.2 years. Learned attention weights corresponded to neuroanatomical regions known to be vulnerable to age-related change. We found that histopathologic brain age acceleration had significant associations with cognitive impairment, MMSE, p-tau burden, and cerebrovascular disease. These associations were not found when using epigenetic-based measures of age acceleration. These data indicate that estimated histopathologic brain age can be used as a reliable pathologic correlate to identify factors that contribute to accelerated or decelerated brain aging.

Disclosures: G.A. Marx: None. **A.T. McKenzie:** None. **J. Kauffman:** None. **D.G. Koenigsberg:** None. **C. McMillan:** None. **K. Farrell:** None. **J.F. Crary:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.01

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

Support: 319103-4

Title: Ameliorative Effect of Wasp Venom on Alzheimer's Phenotypes in 5xFAD Transgenic Mice

Authors: Y. JEONG[1], Y. KIM[2], C. JANG[3], H. YUN[1], J. CHAE[2], J. LIM[4], J. LIM[1], H. KIM[5], ***J. OH**[6], J.-S. KIM[1];
[1]Sch. of Food Sci. and Biotech., [2]Dept. of Integrative Biol., [3]Inst. of Agr. Sci. and Technol., Kyungpook Natl. Univ., Daegu, Korea, Republic of; [4]Dept. of Nuclear Med., Keimyung Univ. Dongsan Hosp., Daegu, Korea, Republic of; [5]Dept. of Korean Med. Develop., Natl. Inst. for Korean Med. Develop., Daegu, Korea, Republic of; [6]New Drug Develop. Ctr., Daegu-Gyeongbuk Med. Innovation Fndn., Daegu, Korea, Republic of

Abstract: Alzheimer's disease (AD) is the most prevalent neurodegenerative disease. AD is often characterized by progressive and irreversible impairment of cognitive functions such as learning, memory, reasoning and perception and by the eventual loss of language and motor skills with gross atrophy of the brain. However, its etiology still remains poorly understood, and thus therapeutic interventions are limited. Our previous study revealed that wasp venom (WV, obtained from *Vespa velutina*) exerted anti-neuroinflammatory and neuroprotective effects in lipopolysaccharide-treated mice. Herein, we examined whether WV administration may ameliorate the major hallmarks of AD pathology which manifest in 5xFAD transgenic mice. The mice at 6 months of age were intraperitoneally treated with WV at a dose of either 250 or 400 μg/kg BW once a week for a total of 14 weeks. We found that WV administration (i) improved behavioral deficits as assessed by passive avoidance, Morris water maze, and Y-maze tasks, (ii) decreased the inflammation-associated protein levels in the cerebral cortex, (iii) decreased the levels of oxidative stress markers (8-hydroxy-2'-deoxyguanosine in the serum and malondialdehyde in the liver and brain), and (iv) lowered the level of C-terminal fragment of Aβ precursor protein (an enzymatic product of β-secretase) in the hippocampal tissue. Moreover, histological abnormalities and Aβ plaques observed in the hippocampal area of 5xFAD mouse brain were considerably reduced with WV administration. Interestingly, our preliminary data demonstrated that the effectiveness was attributed to a tryptophan derivative, serotonin, which was found to be a component of the WV. Overall, these findings suggest that a long-term administration of WV may possibly alleviate AD symptoms and delay its progression.

Disclosures: Y. Jeong: None. **Y. Kim:** None. **C. Jang:** None. **H. Yun:** None. **J. Chae:** None. **J. Lim:** None. **J. Lim:** None. **H. Kim:** None. **J. Oh:** None. **J. Kim:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.02

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

Support: NIH Grant 1R21NS054162-01
Joseph Drown Foundation Grant

Title: Circulating memory CD8 T cells that induce AD-like pathology in mice eliminate Doublecortin $(DCX)^+$neurons independent of interferon-gamma production

Authors: *C. WHEELER[1,2,3,4], A. PANWAR[4], A. RENTSENDORJ[4], M. C. JHUN[4], N. GULL[5], K. L. BLACK[4], M. KORONYO-HAMAOUI[6], D. K. IRVIN[7];
[1]Res. & Develop., T-Neuro Pharma Inc, Aptos, CA; [2]StemVax Therapeut., Los Angeles, CA; [3]Board of Directors, Intl. Brain Mapping Fndn, Society for Brain Mapping & Therapeut., Pacific Palisades, CA; [4]Neurosurg., [5]Regenerative Med. Inst., [6]Neurosurgery, Maxine Dunitz Neurosurgical Res. Inst., Cedars-Sinai Med. Ctr., Los Angeles, CA; [7]Res. and Develop., NovAccess Global & StemVax Therapeut., Los Angeles, CA

Abstract: Objective: We quantified Doublecortin $(DCX)^+$ neurons in the subventricular zone (SVZ), corpus callosum (CC), and cortex in ^{hi}T mice, a model of sporadic Alzheimer's disease (AD) induced by i.v. injection of wild-type or functionally deficient CD8 T cells into nude (B6.Foxn1) hosts. Rationale: ^{hi}T mice develop all major hallmarks of AD with aging, including amyloid plaques, neurofibrillary tangles, robust neurodegeneration and cognitive decline, when induced by WT-CD8 T cell injection. When induced by injection of CD8 T cells deficient in interferon-gamma (IfnγKO-CD8), ^{hi}T mice develop more limited plaque and tangle pathology with aging, without neurodegeneration or cognitive decline, resembling early-stage AD. To examine a potential early mechanism in their neuropathology, we examined CD8 T cells and DCX^+ neurons in C57BL/6 mouse brain, as well as in brains of aged (15 mos) ^{hi}T mice induced with WT-CD8 or IfnγKO-CD8 T cells. Method: Mouse brains were stained with anti-CD8 (clone 53-6.72; 1:100) and anti-DCX antibodies (polyclonal sc-8066; 1:2000) for C57BL/6, and with anti-DCX in ^{hi}T mice and nude controls (PBS-injected), followed by anti-rat and/or anti-goat secondary and DAPI staining (Alexa Flour-488, -647; 1:200). Nude brain staining was blinded with respect to experimental groups. Coronal half-brain sections including left ventricles were imaged at 10x objective magnification ($n \geq 8$ biological replicates), and CC imaged at 5x ($n \geq 2$ biological replicates). DCX signals were quantified by induction group after unblinding. Result: CD8 T cells were observed in close proximity to DCX^+ neurons in the SVZ rostral migratory stream of C57BL/6 mice, but were dramatically decreased in SVZ and throughout coronal brain sections in ^{hi}T mice induced with WT-CD8 and IfnγKO-CD8 T cells (Avg$\pm$SEM: 1.15$\pm$0.16, 0.44$\pm$0.02, and 0.37$\pm$0.10 in PBS, IfnγKO-CD8, and WT-CD8 injected groups, respectively; $P <$ 0.005 for T cell vs. PBS group by 2-tailed T-test). DCX signals in corpus callosum generally reflected these values (Avg$\pm$SEM: 0.63$\pm$0.20, 0.43$\pm$0.14, and 0.25$\pm$0.17 in PBS, IfnγKO-CD8, and WT-CD8 groups, respectively), but were not significant ($P \geq$ 0. 05 by 2-tailed T-test). Conclusion: Our findings suggest that age-related CD8 T cells modulate AD-like features in the ^{hi}T mouse model by Ifnγ-mediated acceleration of neuropathology, as well as Ifnγ-independent elimination of DCX^+ neurons, the latter of which may occur prior to widespread neurodegeneration and cognitive decline. As such, they point to inhibition of age-related T cell activity, as well as enhancement of neurogenesis, as potential targets for intervention in AD-like neurodegeneration.

Disclosures: C. Wheeler: A. Employment/Salary (full or part-time):; T-Neuro Pharma Inc. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); T-Neuro Pharma Inc. F. Consulting Fees (e.g., advisory boards); StemVax Therapeutics. **A. Panwar:** None. **A. Rentsendorj:** None. **M.C. Jhun:** None. **N. Gull:** None. **K.L. Black:** None. **M. Koronyo-Hamaoui:** None. **D.K. Irvin:** A. Employment/Salary (full or part-time):; NovAccess Global. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); NovAccess Global.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.03

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

Support: NIH AG 061746
Larry L. Hillblom postdoctoral Fellowship #2021-A-020-FEL

Title: C5ar1 antagonist alters microglial cells and influences microglial synaptic pruning in a mouse model of alzheimer's disease

Authors: *A. GOMEZ-ARBOLEDAS, K. CARVALHO, G. BALDERRAMA GUTIERREZ, P. SELVAN, S.-H. CHU, H. LIANG, A. MORTAZAVI, A. J. TENNER;
Univ. of California Irvine, Irvine, CA

Abstract: Alzheimer’s disease (AD) is a neurodegenerative disorder that causes cognitive decline and memory loss. The etiology underlying AD is complex and multi-factorial, including inflammation and complement activation as essential contributors. Local activation of the complement system by fAβ can generate C5a, that binds to its receptor C5aR1 on microglial cells and contributes to neuroinflammation. Previous results from our lab have shown a beneficial effect using either genetic ablation (C5aR1KO) or pharmacological inhibition of C5aR1 in AD mouse models. To further examine the effect of PMX205 (a C5aR1 antagonist) on microglial gene expression and gliosis in a mouse model of AD, Tg2576 mice (and WT littermates) were treated with PMX205 in the drinking water for 12 weeks (12-15 mo of age). Mice were perfused, and brains were harvested and further processed for scRNA-seq and IHC analysis. Confocal images as well as super-resolution images were further analyzed by Imaris, where 3D reconstruction analysis and quantifications were carried out to test differences in microglial phagocytosis and synaptic density. By using single cell RNAseq, we recovered 10 different populations of microglial cells. Our results revealed significant changes in the proportions of microglial clusters when AD animals were treated with PMX205. Clusters 13 and 16 are mainly present in the AD mice, when compared to WT littermates, and they both express a relevant DAM signature. Interestingly, we also identify a subpopulation of microglial cells, cluster 8, that is associated with synaptic pruning. Furthermore, cluster 8 is reduced in the hippocampus of Tg2576 mice treated with PMX205, suggesting a beneficial role of C5aR1 antagonism. Additionally, using super-resolution microscopy we observed a significant loss of VGlut1 puncta in the CA3 hippocampal region in the Tg2576 mice when compared to WT littermates, that was rescued when PMX205 was administer to the mice. Moreover, 3D analysis of microglial cell synaptic engulfment showed a significant reduction of synaptic pruning when comparing Tg2576-PMX205 with Tg2576-vehicle, which correlates with the rescue in VGlut1 puncta observed by super-resolution. Overall, our results demonstrate a neuroprotective and beneficial effect of blocking C5a-C5aR1 signaling in the Tg2576 mouse model of Alzheimer’s

disease. Thus, modulation of C5a-C5aR1 could be a valuable therapeutic strategy for Alzheimer's disease patients.

Disclosures: A. Gomez-Arboledas: None. **K. Carvalho:** None. **G. Balderrama Gutierrez:** None. **P. Selvan:** None. **S. Chu:** None. **H. Liang:** None. **A. Mortazavi:** None. **A.J. Tenner:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.04

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

Support: NIH R01AG072896

Title: Effect of chronic alcohol dosing on alzheimer's disease pathology in a mouse model of amyloidosis

Authors: *D. V. CHANDRASHEKAR[1], N. JAGADEESAN[1], T. ABDULLAH[1], R. T. CHANG[1], J. YANG[1], R. MURPHY[1], A. DREW[1], R. TRINH[1], O. TAPIA[1], M. CHOI[2], D. HAN[3], R. K. SUMBRIA[1];
[1]Dept. of Biomed. and Pharmaceut. Sci., Chapman Univ., Irvine, CA; [2]Dept. of Neurosci., Claremont McKenna Col., Claremont, CA; [3]Sch. of Pharm., Keck Grad. Inst., Claremont, CA

Abstract: Chronic alcohol intake is considered a modifiable risk factor for Alzheimer's disease (AD). However, studies show that low to moderate alcohol consumption may protect against while high alcohol consumption may potentiate AD pathology. Herein, we studied the effects of mild but chronic alcohol dosing on AD pathology markers using the Tg2576 AD mice subjected to the National Institute on Alcohol Abuse and Alcoholism (NIAAA) alcohol feeding regimen. Approximately 13-month-old male and female Tg2576 (AD), or wild-type mice were fed with Liber-De Carli (LDC) diet with 5% ethanol or control diet for six weeks (n=11-13/group). Exploration (open-field test) and spatial reference memory (Y-maze test) were assessed 6-weeks after treatment. Terminal blood was collected to measure alcohol levels and markers of liver injury. Brains were harvested for β-amyloid, neuroinflammation (microglia and TNF-α), β-amyloid synthesis and transport proteins (APP and LRP1), and neuronal health (PSD95) using immunoassays. Chronic alcohol feeding significantly increased ($p<0.05$) the mortality in the alcohol-fed AD mice. Blood alcohol levels were ~10-12mg/dL (0.01%) ~5h after alcohol feeding, which is equivalent to mild effects of alcohol observed in humans. Liver injury markers largely remained unaltered, indicating an absence of liver injury. Alcohol feeding increased the latency to the novel arm ($p<0.05$) and a reduced selection of the novel arm as the first arm choice ($p<0.001$) in AD mice. Alcohol feeding did not change brain human β-amyloid levels, however, it increased ($p<0.01$) endogenous mouse β-amyloid (1-42) in AD mice. Microglia-positive area and number were significantly reduced ($p<0.05$) in alcohol-fed AD male mice, suggestive of

microglial dystrophy. No change in brain levels of APP, LRP1, TNF-α, and PSD95 was observed. Overall, the LDC diet with 5% ethanol reduced spatial reference memory, increased endogenous β-amyloid (1-42), and reduced microglia-positive area, suggesting that chronic mild alcohol intake results in modest but significant effects in the Tg2576 mice.

Disclosures: D.V. Chandrashekar: None. **N. Jagadeesan:** None. **T. Abdullah:** None. **R.T. Chang:** None. **J. Yang:** None. **R. Murphy:** None. **A. Drew:** None. **R. Trinh:** None. **O. Tapia:** None. **M. Choi:** None. **D. Han:** None. **R.K. Sumbria:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.05

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

Support: Swedish Research Council Grant #2018-03033
Swedish Research Council Grant #2019-01125

Title: Intraneuronal amyloid-beta and associated neuroinflammatory alterations in early Alzheimer’s disease

Authors: *M. G. GARCIA, T. T. ROOS, S. BACHILLER, G. K. GOURAS, T. DEIERBORG; Exptl. Med. Sci., Lund Univ., Lund, Sweden

Abstract: The amyloid cascade hypothesis of Alzheimer’s disease (AD) posits that extracellular amyloid-beta (Aβ) plaques are the trigger for downstream, detrimental events, including intracellular tau tangles, extensive neuroinflammation, and cognitive decline. However, increasing evidence suggests that neuroinflammatory and synaptic alterations are events that precede even plaque formation. Furthermore, accumulation of Aβ intraneuronally (iAβ), particularly in synapses, has been observed in the brains of patients with early AD pathogenesis, suggesting that this aberrant accumulation may form part of the groundwork for later, more well-known pathological events, such as plaque formation. Based on these findings, we hypothesize that there is an interplay between neuron- and microglia-related alterations that can be linked to the appearance of aggregated iAβ and contribute to pathogenesis. To study early neuron-microglia alterations related to iAβ, we first assessed a model using the well-separated anatomical connection between the subiculum (SUB) and mammillary bodies (MB) in young, pre-plaque 5xFAD AD-model mice. Using immunohistochemistry, we observed punctate and wispy plaque-like aggregated iAβ in SUB and MB, respectively. Despite the presence of aggregated Aβ, no gross microglial morphological alterations, such as those associated with reactive microglia around plaques, were observed. By injecting a viral vector in SUB, we were able to induce GFP expression in neurons and confirm that the neurons with somatodendritic, punctate iAβ in SUB were related to the wispy plaque-like Aβ in MB via their axon terminals. Therefore, with this model, we will be able to consider differential effects of somatodendritic

versus axonal iAβ on neuron-microglia interactions. Further work, including spatial proteomics, will be done to analyze markers that are associated with neuronal and microglial health and function. Overall, our aim is to better understand these iAβ-vulnerable environments and the role of neuron-microglia interactions within to shed light on AD pathogenesis. Ultimately, understanding the early events in AD may be key for the success of future disease-modifying therapeutics, which are sorely needed.

Disclosures: M.G. Garcia: None. **T.T. Roos:** None. **S. Bachiller:** None. **G.K. Gouras:** None. **T. Deierborg:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.06

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

Support: NIH Grant R01AA027097
NIH Grant R56AG058849

Title: Neuroinflammation alters oxygen pressure and blood flow in a mouse model of Alzheimer's disease

Authors: *C. LIU[1], M. ALFADHEL[1,2], A. CÁRDENAS-RIVERA[1], M. A. YASEEN[1]; [1]Bioengineering, Northeastern Univ., Boston, MA; [2]Athinoula A Martinos center for Biomed. Imaging, Massachusetts Gen. Hospital/Harvard Med. Sch., Boston, MA

Abstract: Neuroinflammation alters oxygen pressure and blood flow in a mouse modelof Alzheimer's diseaseAuthors*Chang Liu, Alfredo Cardenas-Rivera, Mohammad Abbas Yaseen; Department of Bioengineering,Northeastern University, Boston, MA**DisclosuresChang Liu**: None. **Alfredo Cardenas-Rivera**: None. **Mohammad Abbas Yaseen**: None.**Abstract** Chronic inflammation is one of the most distinctive characteristics of Alzheimer's disease (AD). The effect of chronic inflammation on AD brain function are debatable as different studies show conflicting results. Little is known about how brain hemodynamics and oxygenation are affected by AD inflammation. Here, we use two-photon phosphorescence lifetime imaging with an oxygen-sensitive dye "Oxyphor 2P" to measure the changes of brain intravascular oxygen pressure (pO2) before and after endotoxin-induced neuroinflammation in an awake mouse model of AD. Capillary red blood cell flux(RBC flux) was measured through two-photon phosphorescence intensity microscopy. 2-photon microscopy enables us to investigate the influence of AD inflammation in cerebral blood flow and oxygenation with microscale-level spatial resolution and provides more insight into understanding inflammation's contribution to the preclinical stages of AD progression. To induce chronic inflammation, we inject lipopolysaccharide (LPS) intraperitoneally, daily for two weeks, in female APPswe:PS1dE9mice and age-matched wild-type (WT) controls (n=10 each). Intravascular pO2 and RBC flux were

measured in the somatosensory cortex up to 500 μm depth on Day 0, 8 and 15 during LPS injection. Our results show significant decreases of capillary pO2 in cortical layer I to V after 1 week of LPS injection in both cohorts. pO2 changes in AD mice are more pronounced than the changes in WT controls, which suggest AD mice brains are more vulnerable to chronic inflammation due to the presence of AD-related pathology. Two-weeks of LPS injection, capillary pO2s in both AD and WT mice recovered slightly, although still less than the baseline-level, indicating that both types of mice can adapt their brain oxygen supply to persistent inflammation. We also found notable inflammation-induced changes of RBC flux along cortical layers in AD mice. Our findings suggest that inflammation further aggravates AD-related dysregulation of cerebral blood flow and oxygenation.

Disclosures: C. Liu: None. **M. Alfadhel:** None. **A. Cárdenas-Rivera:** None. **M.A. Yaseen:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.07

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

Support: ADCS grant

Title: Transplantation of wild type hematopoietic stem and progenitor cells rescue Alzheimers disease in a mouse model and highlights the central role of microglia in disease pathogenesis

Authors: *P. MISHRA[1], A. SILVA[2], J. SHARMA[1], J. NGUYEN[1], D. PIZZO[1], D. SAHOO[1], S. CHERQUI[1];
[1]UCSD, San Diego, CA; [2]Univ. of California, Sna Diego, CA

Abstract: Transplantation of wild-type hematopoietic stem and progenitor cells rescue Alzheimer disease in a mouse model and highlights the central role of microglia in disease pathogenesis
Priyanka Mishra[1], Alexander Silva[1], Jay Sharma[1], Jacqueline Nguyen[1], Donald P Pizzo[2], Debashis Sahoo[1,3,4] and Stephanie Cherqui[1]*
[1]Department of Pediatrics, University of California, San Diego, La Jolla, California, USA.[2]Department of Pathology, University of California, San Diego, La Jolla, California, USA.[3]Department of Computer Science and Engineering, University of California, San Diego, La Jolla, California, USA.[4]Moores Comprehensive Cancer Center, University of California, San Diego, La Jolla, California, USA.
Abstract:
Alzheimer's disease (AD) is the most prevalent cause of dementia but still no effective treatment exists. Microglia have been implicated in AD, but their role is still matter of debate. Our study represents direct evidence that microglia play a key role in disease progression, and that

replacing diseased *APP*/*PS1* microglia via single wild-type (WT) hematopoietic stem and progenitor cell (HSPC) transplantation rescue AD in the 5xFAD mice. Cell therapy led to complete rescue of the neurocognitive impairment, and significant decrease of Aβ plaque burden in the hippocampus and cortex of the 5xFAD mice. Proliferation and activation of microglia were apparent in the 5xFAD mice untreated or transplanted with *APP*/*PS1* HSPCs, whereas distinct reduction in number and activation of microglia was observed in WT HSPC-transplanted mice. Further, transcriptomic analysis revealed significant decrease of "disease-associated microglia" in the cortex and "neurodegeneration associated endothelial cells" in the hippocampus, and T cell associated genes in both hippocampus and cortex of the WT HSPC-transplanted 5xFAD mice compared to diseased controls. Therefore, this work strongly highlights the important role of immune cells in the pathogenesis of AD, and suggest that HSPC gene therapy to correct known familial mutations in AD could be a promising therapeutic approach for treating AD.

Disclosures: P. Mishra: None. **A. Silva:** None. **J. Sharma:** None. **J. Nguyen:** None. **D. Pizzo:** None. **D. Sahoo:** None. **S. Cherqui:** F. Consulting Fees (e.g., advisory boards); Papillon Therapeutics Inc, Cystinosis Research Foundation, AVROBIO.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.08

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

Support: NIH Grant NS104282-01A1

Title: Depression precedes cognitive impairment that can be improved by targeting meningeal B cells in the 5xFAD model of Alzheimer's disease

Authors: *J. IANNUCCI[1], S. BEEVERS[1], K. PULIDO[1], V. ARISMENDI[1], A. R. TAYLOR[1], R. JIMENEZ[1], S. SADHWANI[1], L. IBARRA[1], R. P. TOBIN[3], M. K. NEWELL-ROGERS[2], L. A. SHAPIRO[1];
[1]Neurosci. and Exptl. Therapeut., [2]Med. Physiol., Texas A&M Hlth. Sci. Ctr., Bryan, TX; [3]Dept. of Surgery, Div. of Surgical Oncology, Univ. of Colorado Anschutz Med. Campus, Aurora, CO

Abstract: Background: Depression often occurs prior to Alzheimer's disease (AD), and depression and AD have been linked to immune mechanisms. A recent study identified a pathological role for pro-inflammatory B cells in AD, with B cell depletion reducing Aβ plaque accumulation, neuroinflammation, and behavioral deficits in three transgenic AD models. Altered B cell subsets in the periphery were identified in individuals with depression and a specific role for B cells in depression-associated behavior in mice was reported. We've identified a pathogenic role for CLIP+ B cells. CLIP, Major Histocompatibility Complex class II (MHCII) invariant peptide, is key for the transition from an innate to an adaptive immune response. Using

a Competitive Antagonist Peptide (CAP) against CLIP, we have shown that CLIP+ B cell depletion has functional and neuropathological benefits. We hypothesized that depression-associated behavior changes would precede cognitive impairment in the 5xFAD model of AD and these impairments will be improved by CAP. **Methods:** 11-week-old WT and 5xFAD mice underwent baseline depression testing, including social interaction and burrowing test. 3-month-old mice were treated once with either CAP or vehicle followed by monthly depression testing for 5 months. At 6 months post-treatment, hippocampus-associated cognitive performance was assessed with pattern separation test (PST) and y-maze, followed by meningeal isolation and flow cytometric analysis. **Results:** Depression-associated changes were observed in 5xFAD mice as early as 11-weeks-old, persisting up to 8 months of age. CAP treatment in 5xFAD mice protected against age-associated decline in social interaction and PST impairments, but it did not rescue Y-maze ability. Meninges from 9-month 5xFAD mice had significantly more B and CLIP+ B cells compared to WT, and CLIP+ B cells were significantly reduced by CAP injection at 3 months of age. **Conclusion**: This study builds upon previous studies demonstrating a pathological role for B cells in depression and AD. We demonstrate depression-associated behavior changes in 5xFAD mice by 11 weeks of age that persist until at least 8 months of age. These changes are prior to the previously reported onset of cognitive impairment, suggesting that depression-associated behavior precedes cognitive impairment in this AD model. Considering the persistent depression, this novel phenotype in 5xFAD mice facilitates longitudinal assessment of mechanistic targets. The results also suggest that CLIP+ B cells increase their trafficking to the meninges in 5xFAD mice and reducing this B cell subset is associated with improved social interaction and PST performance.

Disclosures: J. Iannucci: None. **S. Beevers:** None. **K. Pulido:** None. **V. Arismendi:** None. **A.R. Taylor:** None. **R. Jimenez:** None. **S. Sadhwani:** None. **L. Ibarra:** None. **R.P. Tobin:** None. **M.K. Newell-Rogers:** None. **L.A. Shapiro:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.09

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

Support: NIH U54AG065187

Title: In vivo characterization of antibodies directed against TREAT-AD target proteins in a mouse model of AD pathology

Authors: *S. DOOLEN[1], R. AYOUBI[2], C. LAFLAMME[2], R. BATARBET[3], E. ZOELLER[3], S.-P. G. WILLIAMS[1], S. J. SUKOFF RIZZO[1];
[1]Med., Univ. of Pittsburgh, Pittsburgh, PA; [2]McGill Univ., Montreal, QC, Canada; [3]Emory Univ. Sch. of Med., Atlanta, GA

Abstract: The overarching goals of the Target Enablement to Accelerate Therapy Development for Alzheimer's disease (TREAT-AD) program is to improve, diversify and reinvigorate the AD drug development pipeline by accelerating the characterization and experimental validation of next generation therapeutic targets. To this end, a major goal in support of the Emory-Sage-Structural Genomics Consortium (SGC) TREAT-AD Center is to develop and identify high-quality tools to test target or mechanistic hypotheses. As commercial research antibodies become available and validated in commercially available knock-out cell lines, it is essential to extend the characterization of these antibodies in animal models. This allows not only for confirmation of the presence and/or changes in the protein level of target proteins but will also potentially serve as a reagent for future *in vivo* target engagement studies. Here we have used the 5xFAD mouse to characterize target proteins. While the 5xFAD is a well-characterized and highly published transgenic model that manifests Aβ plaque deposition as early as 4-6 months of age, we have not previously characterized the expression of new target proteins being pursued by the TREAT-AD center. Here we have characterized the expression of Moesin, CD44, Midkine and SFRP1 in the 5xFAD mouse model. YCharOS (https://ycharos.com/) is an open-science organization which has industrialized an antibody characterization platform formalized at the Montreal Neurological Institute. Together with YCharOS, we have characterized commercial antibodies against Moesin, CD44, Midkine and SFRP1 using corresponding human knockout cell lines. Here, we have used the best performing antibodies on protein extracts from cortex of 9 month aged 5xFAD mouse brain homogenates and compared to age-matched C57BL/6J controls. Anti-Moesin ab52490 reacted in mouse brain homogenate with a predicted molecular weight of 68 kD. Moesin protein expression was 1.96 times higher in 5xFAD compared to WT. Anti-CD44 ab189524 reacted with a band at the predicted size of 82 kD. CD44 protein expression was 2.89 times higher in 5xFAD compared to WT. Anti-Midkine AF7769 reacted with a band ~16 kD and a 24.7 times greater expression in 5xFAD compared to WT. Anti-SFRP1 ab267466 reacted with a band at 35 kD as predicted. SFRP1 protein expression was 5.96 times greater in 5xFAD compared to WT. Our data suggest these proteins are involved in AD-associated pathology and are a potential source of promising therapeutic targets and biomarkers for AD that can be directly interrogated for translational in vivo target engagement studies with potential novel therapeutics in the 5XFAD mouse model.

Disclosures: S. Doolen: None. **R. Ayoubi:** None. **C. Laflamme:** None. **R. Batarbet:** None. **E. Zoeller:** None. **S.G. Williams:** None. **S.J. Sukoff Rizzo:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.10

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

Support: NIH Grant T32GM135095

Title: Inhibition of G9a modulates molecular and behavioral pathogenesis in Alzheimer's Disease

Authors: *R. N. SHEEHY[1], L. XIE[2], Y. LUO[1], X. CHEN[2], J. SONG[1,3];
[1]Pharmacol., [2]Biochem. and Biophysics, [3]Neurosci. Ctr., Univ. of North Carolina at Chapel Hill, Chapel Hill, NC

Abstract: Alzheimer's Disease (AD) is a neurodegenerative disease that characterized by aberrant processing of amyloid precursor protein (APP) to form neuroinflammatory Aβ plaques and progressive cognitive failure. G9a (EHMT2), a histone/lysine methyltransferase, has recently been implicated in AD. Recent work showed that short-term enzymatic inhibition of G9a for 3 days rescued synaptic and some cognitive functions in AD mice without an effect on Aβ plaques. Despite the established role of G9a in acute treatment of AD, the long-term effects of G9a inhibition remain unknown. Recent data from our lab implicated increased activity of G9a in chronically inflamed microglia during AD. We hypothesized long-term inhibition of G9a would significantly ameliorate microglial activation in AD and thus modulate plaque deposition and further improve behavior. To test this hypothesis, we utilized 5xFAD mice, an Aβ AD mouse model, and treated these mice with a novel brain penetrant G9a inhibitor or vehicle control for 6 weeks. After inhibitor treatment, mice underwent behavioral testing and tissue was collected for both immunofluorescence and proteomic analysis. Inhibitor-treated mice displayed significantly altered synaptic markers, neuroinflammatory markers, plaque deposition, and behavioral changes. These results implicate a prominent role for G9a and its downstream effectors for multiple facets of AD pathogenesis. Overall, our data suggest that targeting G9a to treat AD has putative therapeutic value.

Disclosures: R.N. Sheehy: None. **L. Xie:** None. **Y. Luo:** None. **X. Chen:** None. **J. Song:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.11

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

Support: NIH R01 AG060148(AT)
NIH R21 AG61746
NIH T32 AG000096
NIH MARC Grant T34GM136498

Title: Loss of Perineuronal Nets in Alzheimer's Disease Mouse Models are not mediated by C5a-C5aR1 signaling

Authors: *T. PETRISKO[1], M. A. GARCIA[1], A. J. TENNER[2];
[1]Mol. Biol. and Biochem., Univ. of California, Irvine, Irvine, CA; [2]Neurobio. and Behavior, Univ. California Irvine, Irvine, CA

Abstract: Perineuronal nets (PNNs), an extracellular matrix structure, primarily surround parvalbumin (PV) inhibitory neurons. Together, PVs and PNNs are critical for proper long-term memory. Loss of PNNs and PV neurons has been demonstrated in both patient's and animal models of Alzheimer's Disease (AD). Furthermore, this decrease has been revealed to be microglial dependent in mice. The complement system, a critical component of the innate immune system, is activated in the context of AD pathology, and genetic ablation or pharmacologic inhibition of the complement receptor, C5aR1, has been shown to provide protection from gliosis in animal models. The objective of this study was to determine if PV interneurons and PV neurons surrounded by perineuronal nets (PV^+ PNNs) are reduced in the Arctic (Arc) and Tg2576 mouse models of AD and if these reductions are rescued by inhibition of C5a-C5aR1 signaling. Brain sections from 10-month Arc and Arc C5aR1 knockout mice and 15-month Tg2576 mice treated with the C5aR1 inhibitor PMX205 for 12-weeks were stained for PV and PNNs, imaged at 10X and quantified using IMARIS. While no change in the number of PV neurons was observed, Arc mice showed a significant reduction in the percentage of PNN^+ PV neurons compared to WT mice in both the hippocampus (62.5%) and cortex (33%). Arc C5aR1KO animals also demonstrated a significant reduction in percentage of PNN^+ PV neurons in both regions (70%, 35%, respectively) when compared to WT C5aR1KO. No difference was observed between Arc and Arc C5aR1KO mice. No significant loss of PV neurons nor the percentage of PNN+ PV neurons were observed in Tg2576 mice relative to WT regardless of PMX205 treatment. These results indicate that C5a-C5aR1 signaling does not contribute to PNN loss in the Arc mice model. Our results also indicate there are differences among the mouse models in damage to PNNs, providing opportunities to dissect pathways that lead to PNN loss.

Disclosures: T. Petrisko: None. **M.A. Garcia:** None. **A.J. Tenner:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.12

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

Support: NIH/NIA R01AG057555
NIH/NCATS # TL1-TR-002386
City University of New York (Neuroscience Collaborative program, Graduate Center)

Title: Bt-11, an immunomodulatory drug that binds to lanthionine synthetase c-like 2, ameliorates cognitive deficits and hippocampal pathology in a transgenic rat model of alzheimer's disease

Authors: *E. BIRNBAUM[1,4,5], P. A. SERRANO[2], L. XIE[3], C. HENCHCLIFFE[7,6], M. FIGUEIREDO-PEREIRA[1], P. ROCKWELL[1];
[1]Biol., Hunter Col., NY, NY; [2]Psychology, Hunter Col., New York, NY; [3]Computer Sci., Hunter Col., NY, NY; [4]Biol., CUNY Grad. Ctr., Ny, NY; [5]Clin. and Translational Investigation, [6]Neurol., Weill Cornell Med., Ny, NY; [7]Neurol., UC Irvine, Irvine, CA

Abstract: Neuroinflammation is a key target for drug development in Alzheimer's disease (AD). BT-11 [*methanone,1,1'-(1,4-piperazinediyl)bis(1-(6-(1H-benzimidazol-2-yl)-2-pyridinyl)-*] is an orally active lanthionine synthetase C-like 2 (LANCL2) binding compound that has potential to treat AD. Clinical trials in Irritable Bowel Disease (IBD) demonstrated immunomodulatory properties of BT-11, including an increase in T regulatory cells in the gut. This effect is relevant to AD, as this disease is characterized by an imbalance in pro-inflammatory and anti-inflammatory signaling, allowing inflammation to go unchecked. BT-11 is a drug that could be repurposed to treat AD based on its ability to modulate inflammation through its target LANCL2, as well as in silico results. Our studies utilized a unique transgenic rat model (TgF344-AD, Tg-AD) that exhibits progressive age- and hippocampal-dependent spatial learning and memory deficits, as well as AD pathology that mimics that exhibited by AD patients. BT-11 was administered orally (8.5 mg/kg) to a cohort of WT and Tg-AD rats for 6 months, beginning at 5 months of age (pre-pathology) to 11 months of age (full AD-pathology). Age-matched, non-treated WT and Tg-AD rats were included as controls. At 11 months of age, spatial learning and memory were evaluated with an active place avoidance task that is hippocampal-dependent. We did not detect any changes in the BT-11 treated and untreated WT rats. Untreated male Tg-AD rats showed significant spatial learning and memory deficits as well as AD pathology compared to male WT rats. Notably, BT-11 treatment mitigated these deficits and improved learning in Tg-AD rats as compared to untreated Tg-AD rats. Immunohistochemical analyses showed a trend towards a reduction of Aβ plaques in the hippocampus of treated Tg-AD male rats compared to non-treated Tg-AD male controls. Our results support the notion that BT-11 is a potential drug candidate for AD treatment. Our goal is to define the mechanism whereby BT-11 improves cognition and AD pathology, whether by direct effect in the CNS or a potential peripheral effect on immune cells in the gut.

Disclosures: E. Birnbaum: None. **P.A. Serrano:** None. **L. Xie:** None. **C. Henchcliffe:** None. **M. Figueiredo-Pereira:** None. **P. Rockwell:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.13

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

Support: AARF-21-850265
NIH RF1AG072727
NIH RF1AG069378

NIH R01AG048993
NIH P20GM113123
NIH U54GM128729

Title: Asthma induced by house dust mite potentiates brain changes in the App^{NL-G-F} mouse model of Alzheimer's disease

Authors: *B. SAHU, S. NOOKALA, A. M. FLODEN, C. K. COMBS;
Biomed. Sci., Univ. of North Dakota, Grand Forks, ND

Abstract: Alzheimer's disease (AD) is an age-related neurodegenerative disorder affecting around 35 million individuals worldwide. Besides aging, various comorbidities increase the risk of AD, including air pollution and asthma. Epidemiological studies have reported a 2.17-fold higher risk of dementia in asthmatic patients. However, the molecular mechanism(s) underlying this asthma-associated AD exacerbation is unknown. This study was designed to explore house dustmite-induced asthma effects on AD-related brain changes using the *App^{NL-G-F}* transgenic mouse model. Male and female C57BL/6 wild type and *App^{NL-G-F}* mice (8-9 months old) were exposed to either saline or house dust mite (dose: 833μg/kg in saline) every alternate day for 16 weeks. Mice were sacrificed at the end of the experiment, and bronchoalveolar lavage fluid (BALF), blood, and brains were collected. BALF was analyzed for immune cell markers, inflammatory mediators, total protein content, and LDH activity. Serum was analyzed for cytokine and soluble Aβ1-40/42 levels. In addition, brain sections were immunostained for Aβ, GFAP, CD68, and collagen 4. Finally, frozen hippocampi and cortices were used to perform Aβ ELISAs and cytokine arrays, respectively. As expected, dustmite exposure increased inflammatory cells, cytokine levels, total protein content, and LDH activity in the BALF from both sexes and genotypes, suggesting induction of a severe asthma-like condition. This correlated with increased levels of serum cytokines in all dustmite exposed groups. Serum from the *App^{NL-G-F}* dustmite-induced asthma group also had significantly increased soluble Aβ1-42 levels in both sexes. In agreement with this peripheral change, hippocampi from asthma-induced male and female *App^{NL-G-F}* mice demonstrated elevated Aβ plaque load and increased soluble Aβ1-40/42 and insoluble Aβ1-40 levels. Dustmite exposure also increased astrogliosis and microgliosis in both sexes of *App^{NL-G-F}* mice, as indicated by GFAP and CD68 immunoreactivity. Additionally, dust mite exposure-induced asthma elevated cortical levels of several cytokines in both sexes and genotypes. Finally, dustmite exposed groups also showed a disturbed BBB integrity in the hippocampus of *App^{NL-G-F}* mice as indicated by the decreased collagen 4 immunoreactivity. Dust mite exposure was responsible for a severe asthma-like condition in the lungs that exacerbated Aβ pathology, astrogliosis, microgliosis, and cytokine changes in the brains of *App^{NL-G-F}* mice. Also, dust mite exposure potentially disrupts the BBB integrity in the brain. Defining mechanisms of asthma effects on the brain may identify novel therapeutic targets for both asthma and AD.

Disclosures: B. Sahu: None. **S. Nookala:** None. **A.M. Floden:** None. **C.K. Combs:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.14

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

Support: NIA R01AG057555

Title: Atractylenolide III mitigates cognitive decline, inflammation, and amyloid plaque development in Alzheimer's Disease

Authors: *A. STEELE[1,4], L. XIE[2], M. E. FIGUEIREDO-PEREIRA[3], P. ROCKWELL[3], P. SERRANO[1];
[1]Dept. of Psychology, [2]Dept. of Computer Sci., [3]Biol. Sci., Hunter Col., New York, NY; [4]CUNY Neurosci. Collaborative, Dept. of Biol., CUNY Grad. Ctr., New York, NY

Abstract: Alzheimer's Disease (AD) is a debilitating neurodegenerative disease characterized by extracellular amyloid beta (Aβ) accumulation, hyperphosphorylated tau tangles, and neuroinflammation, which ultimately results in cognitive decline. AD's prognosis is projected to triple to 14 million people by 2060. Despite the prevalence of the disease, no effective treatments have been produced due to many treatments opting for a one-drug-one-target approach. In scilico analysis of known drugs suggested atractylenolide III (ATC III) as a therapeutic treatment. ATC III is a compound demonstrated to reduce Aβ accumulation and inflammation, and improve cognition across neurodegenerative and neuroinflammatory models. However, the role of ATC III treatment on learning and molecular pathology in AD remains unknown. We hypothesize that ATC III modulates molecular pathways and the innate immune system to reduce Aβ plaque burden, which in turn results in improved learning. We utilized the TgF344 transgenic rat model of AD to investigate the effects of ATC III on cognition and molecular pathology. Our findings indicate that ATC III treatment improves learning in an active place avoidance task in 11-month transgenic male rats by significantly decreasing the percent of time spent in the shock zone ($p<0.005$). Furthermore, preliminary immunohistochemical analyses suggest that Aβ plaque and microglia counts decrease with ATC III treatment. Our results support our hypothesis that ATC III modulates immune responses to Aβ plaque pathology and improves learning in an AD rat model, further elucidating the molecular pathways that regulate neuroinflammation in AD.

Disclosures: A. Steele: None. **L. Xie:** None. **M.E. Figueiredo-Pereira:** None. **P. Rockwell:** None. **P. Serrano:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.15

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

Title: Studying the protective contribution of PLCG2 P522R variant in microglia-mediated immune pathways in neuroinflammation and Alzheimer's Disease

Authors: *X. LIANG;
USC Neurosci. Grad. Program, Los Angeles, CA

Abstract: As the main innate immune cell in central nervous system (CNS), microglia plays an indispensable roles in neuroinflammation and neurodegenerative diseases, such as Alzheimer's Disease (AD). Large scale population studies have identified an AD-associated rare coding variant in the *Phospholipase C Gamma 2 (PLCG2)* locus (rs72824905, P522R, *P*=5.38x10-10, odds ratio = 0.68), which potentially offers protection against AD. This variant is associated with lower $A\beta_{1\text{-}42}$ and $pTau_{181}$ levels in patients, as well as delays cognitive decline in mild cognitive impairment (MCI) progression. It has been reported that *PLCG2* highly expresses in microglia in CNS and mainly involved in immune response, therefore, *PLCG2* P522R variant may be a central player in the microglial-mediated immune pathways during AD pathogenesis. In order to explore the nature of *PLCG2* P522R variant, we generated a $PLCG2^{522R/R}$ knock-in mouse model carrying this human variant with CRISPR-mediated gene editing, as well as $PLCG2^{522R/R}$;5XFAD mouse model by crossing $PLCG2^{522R/R}$ knock-in model with the 5XFAD mice. Our data showed that $PLCG2^{522R/R}$;5XFAD mice exhibited attenuated AD pathologies compared with $PLCG2^{522P/P}$;5XFAD mice at 6 month age, including less burden of Aβ in cortical and hippocampal regions, and more Aβ-associated microglia clustering around Aβ plaques, which may likely link to *PLCG2* P522R variant's hypermorphic effect in immune pathways and its contribution to neuroinflammation in AD. The better understanding of *PLCG2* P522R variant in neuroinflammation and AD pathogenesis can help us provide new strategies to developing clinical therapy in the future.

Disclosures: X. Liang: None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.16

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

Support: R01AG056478
R01AG055865
The Saban foundation
Tom Gordon Foundation

Title: Immunomodulation restores UCHL1 expression and preserves synaptic integrity in the brains of AD-model mice

Authors: *D.-T. FUCHS[1], A. RENTSENDORJ[1], T. TORBATI[2], A. KASINDI[1], J. DOUSTAR[1], M. DAVIS[1], J. SHEYN[1], K. BLACK[1], M. KORONYO-HAMAOUI[1];
[1]Cedars Sinai Med. Ctr., Los Angeles, CA; [2]Western Univ. of Hlth. Sci., Pomona, CA

Abstract: Background: Ubiquitin carboxyterminal hydrolase L1 (UCHL1) is an abundantly expressed deubiquitinating enzyme in the brain, and a crucial component of the ubiquitin-proteasome system (UPS) with a wide range of alternative functions, importantly required for normal synaptic function. We previously found that immunomodulation with glatiramer acetate (GA) amplified expression of osteopontin (SPP1) in macrophages infiltrating the brains of the $APP_{SWE}/PS1_{\Delta E9}$ double-transgenic mouse models of Alzheimer's Disease (ADtg). Surprisingly, we observed that UCHL1 expression was modulated by SPP1 *in vitro*. The effects of GA on cerebral UCHL1 levels *in vivo* in ADtg mice are unknown. **Methods**: The expression of UCHL1 and synaptic density in the brains of GA-immunized ADtg mice were evaluated by immunohistochemistry, ELISA and proteomics analyses. **Results:** Proteomic analysis revealed that UCHL1 was the most upregulated protein by GA activation and most downregulated protein in SPP1-deficient macrophages. We observed that macrophages indeed express UCHL1 and its expression was dependent on SPP1 expression in vitro. Further, we confirmed *in vivo* that UCHL1 colocalizes within SPP1-expressing $Iba1^{+}CD45^{high}$ infiltrating monocytes surrounding Aβ plaques in cortices of ADtg mice. Quantification of UCHL1 revealed a 30% loss of UCHL1 expression in pyramidal neurons in the hippocampi and cortices of ADtg mice relative to WT mice. Outstandingly, we found that GA immunomodulation restored UCHL1 neuronal expression and axonal length along with reduced Aβ pathology and gliosis in ADtg mice. More importantly, increased UCHL1 expression levels concomitantly rescued amyloid beta-induced decrease in synaptic density. **Conclusions**: UCHL1 is expressed by macrophages in vitro as well as in vivo in infiltrating myelomonocytes and modulated by SPP1 regulation. Furthermore, immunomodulation with GA treatment restored UCHL1 expression in the hippocampus and cingulate cortex of ADtg mice and was associated with axonal and synaptic preservation.

Disclosures: D. Fuchs: None. **A. Rentsendorj:** None. **T. Torbati:** None. **A. Kasindi:** None. **J. Doustar:** None. **M. Davis:** None. **J. Sheyn:** None. **K. Black:** None. **M. Koronyo-Hamaoui:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.17

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

Support: NIA grant RF1 AG074566.

Title: Reduced PLCG2 expression alters microglial responses and exacerbates disease pathology in a murine model of Alzheimer's disease

Authors: *E. MESSENGER[1], A. P. TSAI[1], C. DONG[1], P. B.-C. LIN[4], A. OBLAK[4], M. MOUTINHO[4], G. XU[1], Y. LIU[1], K. NHO[2], S. J. BISSEL[5], B. T. LAMB[4], G. E. LANDRETH[3]; [2]Radiology and Imaging Sci., [3]Stark Neurosci. Res. Institute, NB214C, [1]Indiana Univ. Sch. of Med., Indianapolis, IN; [4]Stark Neurosciences Res. Inst., Indianapolis, IN; [5]Stark Neurosciences Res. Inst., Indiana University-Purdue Univ. Indianapolis, Indianapolis, IN

Abstract: Many of the genes that confer altered risk for Alzheimer's disease (AD) are predominantly expressed in microglia and effect innate immune responses and phagocytosis. Among these genes is phospholipase C gamma 2 (PLCG2). The hyperfunctional P522R variant of PLCG2 has been recently associated with reduced risk of AD. PLCG2 is a critical mediator of transmembrane signaling that acts downstream of many immune receptors on microglia. Yet, the contribution of PLCG2 to AD pathology remains unknown. We analyzed the transcriptional changes of mice with *Plcg2* haploinsufficiency compared to wildtype mice. Analogous to AD hippocampus, the transcriptional profiles perturbed by reduced *Plcg2* expression were associated with learning, metabolism, and synaptic functions. Reduced *Plcg2* expression reduced the ability to induce long-term potentiation similar to the amyloidogenic 5xFAD murine model. In AD brains, gene expression analysis of bulk RNA-Seq data revealed several biological processes altered by low *PLCG2* expression, including pathways associated with the inflammatory response, microglial activation, and phagocytosis. Therefore, we systematically investigated the impact of reduced *Plcg2* expression on microglial response and amyloid pathology in 5xFAD mice. Differential pathway analysis highlights altered inflammation-related pathways in both humans and mice with AD. Reduction in *Plcg2* expression altered the phenotype and response of plaque-associated microglia including suppression of cytokine concentrations. Disease progression was exacerbated with increased compact X34-positive plaque deposition. Our study highlights a role for PLCG2 in eliciting microglial responses to amyloid pathology, with reduced *PLCG2* expression associated with exacerbation of AD pathology.

Disclosures: E. Messenger: None. **A.P. Tsai:** None. **C. Dong:** None. **P.B. Lin:** None. **A. Oblak:** None. **M. Moutinho:** None. **G. Xu:** None. **Y. Liu:** None. **K. Nho:** None. **S.J. Bissel:** None. **B.T. Lamb:** None. **G.E. Landreth:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.18

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

Support: HBI Postdoc Pioneers grant

Title: The role of Ms4a6 genes in neuroimmune regulation of Alzheimer's Disease

Authors: *D. GUNEYKAYA CINAR[1], N. HUNTER[1], V. NIKOLAKI[1], R. ANYOHA[1], B. A. STEVENS[2], S. R. DATTA[1];

[1]Dept. of Neurobio., Harvard Med. Sch., Boston, MA; [2]Harvard Med. Sch. Neurobio., Boston Children's Hosp., Boston, MA

Abstract: The Ms4as have been identified in GWAS as conferring substantial Late onset of Alzheimer's Disease (LOAD) risk, but their ability to influence AD has not yet been assessed. *Ms4a6* genes in particular are implicated among those Ms4a genes known to be expressed in the immune system, but very little is known about their involvement in the immune cell regulation and response. In this study, we investigate the microglia and other immune cell function by taking advantage of Ms4a6d knock-out models crossed with subset of AD mouse model for Tau pathology. Our preliminary data suggested that loss of Ms4a6s have initiate cumulative changes in 3 different immune cells: microglia, NK cells and T cells. Moreover, tauopathy-induced synapse loss and cognitive impairment were rescued in Ms4a6d knock-out mice. Together, these results suggest a novel immune regulatory role for *Ms4a6* genes, and indicate that manipulation of this gene family in different immune cells may help building novel complementary therapeutic strategies for Alzheimer's disease.

Disclosures: D. Guneykaya Cinar: None. **N. Hunter:** None. **V. Nikolaki:** None. **R. Anyoha:** None. **B.A. Stevens:** None. **S.R. Datta:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.19

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

Support: NIA R01AG057555
NIA #2R25GM060665-21

Title: A combined treatment with diazoxide a potassium channel activator and dibenzoylmethane that restores eIF2B activity attenuates AD pathology and improves cognition in a transgenic rat model of Alzheimer's disease

Authors: *G. OLIVEROS[1], C. H. WALLACE[1], L. XIE[2], P. A. SERRANO[3], P. ROCKWELL[1], M. E. FIGUEIREDO-PEREIRA[1];
[1]Biol. Sci., Hunter Col., New York, NY; [2]Computer Sci., CUNY Hunter Col., New York, NY; [3]Dept of Psychology, Hunter College, CUNY, New York, NY

Abstract: Alzheimer's disease (AD) is a multifactorial disease for which therapeutic efficacy should benefit from a multi-target approach. Thus, we utilized a combination drug treatment of diazoxide (DZ) and dibenzoylmethane (DIB). DZ is a potassium channel activator. DIB restores eIF2B activity, thus reversing stress-induced translational depression. Previous studies examined each drug's individual therapeutic benefits on attenuating neurodegeneration and apoptosis in other animal model systems. However, their combined treatment potential was not addressed. To test the efficacy of this combined treatment we used the Fisher transgenic 344-AD rat model of

AD, which expresses human mutant "Swedish" amyloid-precursor protein (APPsw) and Δ exon 9 presenilin 1 (PS1 ΔE9). TgF-344AD (Tg) rats exhibit age-dependent progressive AD pathology that resembles human AD pathology more closely than other model systems. We assessed the cognitive performance and AD pathology in 11-month old DZ/DIB treated (for nine-months) transgenic (Tg) and wildtype (WT) rats compared to untreated age-matched littermates. We tested spatial-working memory performances using the radial 8-arm arm maze task (RAM). We also investigated by immunohistochemistry, the effect of the double drug treatment on AD pathology. Untreated Tg rats exhibit significant working memory deficits and AD pathology compared to untreated WT rats. We did not detect any changes in the DZ/DIB treated and untreated WT rats. Notably, the DZ/DIB treatment mitigated the working memory deficits as well as the buildup of hippocampal Aβ plaques and neurofibrillary tau tangles in Tg-treated rats compared to untreated Tg rats. Our results strongly support that the combination DZ/DIB treatment is an effective strategy to mitigate AD pathology due to its multi-target approach that affects multiple signaling pathways.

Disclosures: G. Oliveros: None. **C.H. Wallace:** None. **L. Xie:** None. **P.A. Serrano:** None. **P. Rockwell:** None. **M.E. Figueiredo-Pereira:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.20

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

Support: Psi Chi Graduate Mamie Phipps Clark Diversity Research Grant

Title: Chronic sleep restriction decreases hippocampal BDNF and disrupts immune response to LPS in C57BL/6J mice

Authors: *P. N. BRADEN-KUHLE[1], K. N. BRICE[1], S. K. MILLER[1], A. REGAN[2], G. W. BOEHM[1], M. J. CHUMLEY[2];
[1]Psychology, [2]Biol., Texas Christian Univ., Fort Worth, TX

Abstract: Sleep loss is known to impair neural plasticity and immune function. Immune impairment increases susceptibility to and reduces clearance of pathogens, and may lead to long-term inflammation. This chronic inflammation can produce adverse effects on cognition and longevity. Indeed, chronic sleep loss likely plays a role in the development of chronic inflammatory diseases, such as Alzheimer's disease (AD). Our laboratory has previously demonstrated that chronic sleep restriction (CSR) induces cognitive deficits in a hippocampus-dependent learning task. The current study explored the effects of CSR on inflammation and brain-derived neurotrophic factor (BDNF) following an immune insult. Male and female mice were subjected to six weeks of CSR or a home cage control (HCC) condition for 10 hours every day. Following the last day of CSR, all mice received one intraperitoneal injection of either LPS

or saline. Four hours post-injection, serum and hippocampal tissue were collected for BDNF and cytokine analysis. Results differed between male and female mice. Males that underwent CSR and received an LPS injection had increased levels of peripheral pro- and anti-inflammatory mediators, whereas central levels of these mediators were decreased compared to HCC mice that received LPS. Conversely, CSR female mice that received LPS had moderately lower levels of both peripheral and central pro- and anti-inflammatory mediators compared to HCC females. Further, CSR males exhibited lower levels of BDNF in the hippocampus compared to HCC males, whereas this difference was not observed in females. Thus, these findings suggest a complicated interaction between chronic sleep loss, immune function, and sex in AD pathogenesis.

Disclosures: P.N. Braden-Kuhle: None. **K.N. Brice:** None. **S.K. Miller:** None. **A. Regan:** None. **G.W. Boehm:** None. **M.J. Chumley:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.21

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

Support: NIH Grant AG062179
NIH Grant AG064811
NIH Grant AG069447

Title: The role of dectin-1 signaling in the pathogenesis of Alzheimer's disease

Authors: *J. YANG[1], M. GRAMMER[1], S. DONOHUE[1], R. LALONDE[2], K.-I. FUKUCHI[1]; [1]Univ. of Illinois Col. of Med. At Peoria, Univ. of Illinois Col. of Med. At Peoria, Peoria, IL; [2]Dept. of Psychology, Univ. of Rouen, Rouen, France

Abstract: Recent genetic studies on late-onset Alzheimer's disease (AD) have identified more than a dozen genetic risk variants that are involved in innate immune responses, highlighting the importance of immune cells in the pathogenesis of late-onset AD. Indeed, activated microglia play pivotal and dual roles in AD progression: either clearing Aβ deposits by phagocytosis and promoting neuron survival and plasticity or releasing cytotoxic chemicals, inflammatory cytokines, exacerbating Aβ load and neurodegeneration. Toll-like receptor 4 (TLR4) has been shown to be an essential component of the receptor complexes for Aβ/LPS-induced microglial activation, which can be modified by β-glucan-dectin-1 signaling. We found that β-1, 3-glucan from Euglena gracilis can reverse LPS tolerance in BV2 cells (murine microglial cell line). In this study, 12-month-old clec7a/dectin-1 KO TgAPP mice were used to study the role of dectin-1 signaling in the pathogenesis of AD. In the open field, clec7a KO mice showed reduced distance travelled, reduced time spent moving fast and increased resting time, compared to clec7a wild-type mice. In the elevated plus maze, clec7a KO mice had fewer enclosed arm entries than clec7a

wild-type mice, suggesting less anxiety and safety awareness in clec7a KO mice. In the probe trial of the Morris water maze, clec7a KO mice spent less time in the target quadrant compare to clec7a wild-type mice, suggesting deficits in long-term memory. In line with the behavioral studies, the levels of brain Aβ load in clec7a KO mice were significantly higher than those in the control mice by immunohistochemistry and ELISA. In conclusion, dectin-1 signaling is involved in the AD progression by altering Aβ load and cognitive function. To investigate the underlying mechanism, we are currently performing bioinformatics based on hippocampal transcriptomic profiles. Funded in part by NIH AG062179, AG064811 and AG069447.Key words: Alzheimer's disease, inflammation, macrophage, microglia, Dectin-1

Disclosures: J. Yang: None. **M. Grammer:** None. **S. Donohue:** None. **R. Lalonde:** None. **K. Fukuchi:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.22

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

Title: Dystrophic neurites in the APP/PS1 rat model of Alzheimer's disease

Authors: *S. SARKAR[1], S. M. LANTZ[2], J. B. RAYMICK[3];
[1]Neurotoxicology, [2]Natl. Ctr. for Toxicological Research/ US FDA, Jefferson, AR;
[3]Neurotoxicology, Natl. Ctr. For Toxicology, Jefferson, AR

Abstract: Among the three Fluoro-Jade dyes, Fluoro-Jade C (FJC) is the most sensitive fluorescent marker of neuronal degeneration, allowing for the localization of degenerating neurons, dendrites, axons, and axon-terminals. FJC also has a high binding affinity for the core of plaques observed in Alzheimer's disease (AD), which mainly consists of the amyloid beta (Aβ) peptide. Never characterized before is FJC's ability to bind dystrophic neurites (DNs; swollen neuritic processes). Here, we propose the presence of DNs may correlate with synaptic dysfunction in AD. Several studies have indicated the mechanism of Aβ deposition in AD; however, little attention has been paid to the location of DNs in AD brains. DNs in the AD brain are morphologically recognized by immunohistochemical staining with antibodies specific to amyloid precursor protein (APP) and reticulon 3 (RTN3). DNs also contain ubiquitin (UBI), β-Site APP cleaving enzyme-1 (BACE1; enriched within a distinct subtype of DNs), endosome/lysosome marker lysosomal associated membrane protein 1 (LAMP-1), and/or CD68. In this study, we localized DNs using FJC in the APP/PS1 rat model of AD. Colocalization immunolabeling on FJC-stained tissues with LAMP-1, CD-68, APP, BACE1, RTN3, and UBI were performed. To determine whether FJC binds to microglia, colocalization with light chain ferritin was used. Moreover, to discriminate what location in the plaques the oligomeric form of Aβ and FJC colocalization occurs as well as the location of axonal degeneration, NU-1 and myelin basic protein (MBP) staining were also performed. Data indicate 84% or more of FJC

colocalization with all markers occurs in the diffuse periphery (DNs) of the Aβ plaques, with less colocalization in the plaque core (i.e.-intense central staining). Excluding MBP (only observed in the periphery), of the total area occupied by the plaque cores, the majority colocalized with markers of DNs, microglia, and the oligomeric form of Aβ. Disregarding the distinction between core and peripheral plaque morphology, the intensity and area of colocalization was highest in ferritin and NU-1, while being lowest in CD68. In conclusion, FJC could be used to detect DNs and axonal swelling in the APP/PS1 rat model of AD.

Disclosures: **S. Sarkar:** None. **S.M. Lantz:** None. **J.B. Raymick:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.23

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

Support: K01AA025713
R01AG072894

Title: Adolescent binge ethanol exposure accelerates Alzheimer's disease neuropathology in the basal forebrain

Authors: *R. FISHER[1], L. MATHENY[2], L. QIN[3], L. G. COLEMAN, Jr.[4], R. P. VETRENO[4]; [1]Univ. of North Carolina Chapel Hill, Univ. of North Carolina Chapel Hill, Chapel Hill, NC; [2]UNC-Chapel Hill, Chapel Hill, NC; [3]Ctr. For Alcohol Studies, Univ. of NC, Chapel Hill, Univ. North Carolina, Sch. Med., Chapel Hill, NC; [4]Univ. of North Carolina at Chapel Hill, Univ. of North Carolina at Chapel Hill, Chapel Hill, NC

Abstract: Cholinergic degeneration and neuroimmune system activation are hallmark features of many disease states, including alcohol use disorder (AUD) and Alzheimer's disease (AD). Heavy alcohol use is an etiological factor associated with AD, but little is known about the interaction between adolescent binge alcohol exposure and AD pathology. Preclinical studies using the adolescent intermittent ethanol (AIE) model, which mimics weekend binge drinking behavior, find basal forebrain cholinergic neuron degeneration and increased neuroimmune activation in brain, similar to observed pathology in AD. Using the 5x Familial Alzheimer's disease (5xFAD) mouse model of AD, we tested the hypothesis that AIE treatment would accelerate onset of AD-associated pathology. We report AIE accelerated the loss of basal forebrain cholinergic neurons and hnRNP expression relative to age-matched 5xFAD CONs in female, but not male subjects. This was accompanied by accelerated accumulation of amyloid beta as well as upregulation of AD-related genes. In addition, AIE upregulated glial genes (e.g., Gfap and Iba1), increased microglial Iba-1+IR, and induced proinflammatory neuroimmune signaling genes in the basal forebrain, relative to 5xFAD CONs. In post-mortem human basal forebrain samples of individuals with AUD and an adolescent age of drinking onset, we found AUD increased

amyloid beta expression in the basal forebrain and in ChAT+ neurons of AUD individuals, and decreased hnRNP expression that was negatively correlated with loss of cholinergic cell markers. These data reveal that adolescent binge ethanol exposure accelerates AD-associated neuropathology in the female adult basal forebrain and suggests that adolescent binge drinking may be an etiological factor contributing to AD neuropathology.

Disclosures: R. Fisher: None. **L. Matheny:** None. **L. Qin:** None. **L.G. Coleman:** None. **R.P. Vetreno:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.24

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

Title: Immunohistochemical analysis of plaque pathology and immune cells in brain of 5xFAD mice crossed with CCR2 KO and CX3CR1 KO mice

Authors: *S. GEISSLER, S. BARENDRECHT, F. LA-DEUR, H. CYNIS;
Fraunhofer Inst. for Cell Therapy and Immunol., Halle, Germany

Abstract: Microglia are brain-resident myeloid cells and make up 8-13% of the total cell population in the CNS. Microglia have many different functions, both in normal brain development and under pathological conditions. Besides their phagocytic function, the cells are known to secrete cytokines, e.g. CC-chemokine ligand 2 in inflammation (CCL2, MCP-1) and they receive input from neuronal cells via the CX3CL1-CX3CR1 axis with an impact on microglial activation. Interestingly, CCL2 receptor CCR2 is not expressed in CNS and could contribute to the migration of peripheral immune cells, which would act supplementary to microglial phagocytosis. Furthermore, disturbed communication between neuronal cells and microglia via the CX3CL1-CX3CR1 axis dysregulates microglial responses and contributes to neuronal loss. The aim of our study was to analyze the influence of CX3CR1 and CCR2 knockout, respectively, under progressing cerebral amyloidosis in the 5xFAD mouse model of Alzheimer's-like pathology. We were interested in AD-like pathology and presence of peripheral immune cells. We compared immunohistochemically stained brain slices of male and female mice at the ages of six and twelve months and quantified the influence of the knock-outs on Abeta deposits and present immune cells with special emphasis on MHCII-positive cells in cortex and hippocampus. Plaque deposits decreased in cortex of 6- and in hippocampus of 12-months-old 5xFAD x CCR2-deficient males compared to 5xFAD and in cortex of 12-month-old 5xFAD x CX3CR1-deficient females. Furthermore, 5xFAD x CCR2-deficient mice showed MHCII-positive cells suggesting that these cells are either recruited from periphery by a different chemokine axis than CCL2-CCR2 or they comprise a subset of resident microglia cell as recently suggested. In summary, CCR2- and CX3CR1-knock-outs lead to aggravated AD-like pathology in older 5xFAD mice and to more immune cells in the brain. The presence of immune cells in the

brain was obviously not triggered by the CCL2-CCR2 axis. Furthermore, we could show, that the presence of MHCII-positive myeloid cells was associated with plaque pathology.

Disclosures: **S. Geissler:** None. **S. Barendrecht:** None. **F. La-Deur:** None. **H. Cynis:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.25

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

Support: Federal Ministry of Education an Research grant: 01ED2012

Title: Investigations on periodontal Porphyromonas gingivalis infection on Alzheimer's-like pathology in 5xFAD mice

Authors: *I. KOSKA[1], A. KLAPPER[1], N. TAUDTE[1,2], S. SCHILLING[1], M. BUCHHOLZ[2], H. CYNIS[1];
[1]Fraunhofer Inst. for Cell Therapy and Immunol., Halle, Germany; [2]PerioTrap Pharmaceuticals GmbH, Halle, Germany

Abstract: Alzheimer's disease is characterized by neuroinflammation including activation of microglia, the inflammasome, the complement system and cytokine profiles. Whereas the cause of Alzheimer's disease remains unclear, several risk factors have been identified including metabolic diseases, head trauma and genetic variations. A new hypothesis considers the involvement of infectious agents as risk factors leading to Alzheimer's disease. Chronic periodontitis became evident to be a potential contributing factor for disease development. Porphyromonas gingivalis (P.g.) is described as the keystone pathogen of chronic periodontitis. In order to study the impact of this risk factor on development of Alzheimer's disease, we infected the 5xFAD mouse model of AD-like cerebral amyloidosis with P.g., which is a widely used transgenic mouse model for studying Alzheimer's disease. These mice are double transgenic and express human amyloid precursor protein (APP) and presenilin 1 (PS1) with five mutations. High APP expression leads to increased formation of Aβ peptide, resulting in the deposition of large amounts of extracellular plaques. For establishing periodontitis in mice, in a first study we infected six-month-old female C57BL/6J mice orally with 10^9 CFU of P.g. (ATCC 33277) three times a week for 8 to 22 weeks in order to determine the best time interval for application of the bacteria. Chronic periodontitis was assessed by alveolar bone loss between the first and second molar of the mandibula using µCT. Afterwards, female 5xFAD mice were infected with P.g. in order to assess the impact of periodontitis on Alzheimer's pathology Aβ plaque load and activation of microglia serve as read out parameters for neuropathological manifestation of Alzheimer's disease. In ongoing studies, the impact of infection with P.g. on behavior (anxiety and cognitive behavior) of 5xFAD will be observed. Therefore, investigations in the elevated-plus maze and the Morris-water maze will be performed. This animal model

could serve as a model for screening of new pharmacological therapies targeting infectious agents in order to modulate Alzheimer's disease onset and progression.

Disclosures: **I. Koska:** None. **A. Klapper:** None. **N. Taudte:** None. **S. Schilling:** F. Consulting Fees (e.g., advisory boards); PerioTrap Pharmaceuticals GmbH. **M. Buchholz:** A. Employment/Salary (full or part-time):; PerioTrap Pharmaceuticals GmbH. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); PerioTrap Pharmaceuticals GmbH. **H. Cynis:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); PerioTrap Pharmaceuticals GmbH.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.26

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

Title: Apoa1bp deficiency exacerbates neuropathology in alzheimer's mouse model

Authors: *Y. KIM, S.-H. CHOI, Y. I. MILLER;
UCSD, La Jolla, CA

Abstract: Neuroinflammatory response and activated microglia play an important role in the development of Alzheimer's disease (AD). We recently reported that the secreted apoA-I binding protein (AIBP, encoded by the *APOA1BP* gene) is a key regulator of cellular cholesterol metabolism in microglia, which can inhibit neuroinflammation in a mouse model of neuropathic pain. The mechanism includes selective depletion of cholesterol from inflammatory but not homeostatic microglia via binding to TLR4 and targeting cholesterol-rich lipid rafts. In addition to the anti-inflammatory effects of secreted AIBP, intracellular AIBP regulates mitophagy and ROS generation in macrophages. We obtained *Apoa1bp*$^{-/-}$ *APP/PS1* mice by crossing *Apoa1bp*$^{-/-}$ and the transgenic *APP/PS1* mice to investigate the role AIBP plays in AD pathophysiology. Amyloid beta (Aβ) deposits were significantly increased in female *Apoa1bp*$^{-/-}$ *APP/PS1* mice compared to *APP/PS1*. Moreover, the AIBP deficiency resulted in an exacerbated dysfunctional microglia phenotype, increased microgliosis and neuronal death, and the increased mortality of *APP/PS1* mice. The AAV-mediated overexpression of a secreted form of AIBP in the brain of *APP/PS1* mice restored the microglial homeostatic phenotype reduced Aβ deposits and increased the animal survival rate. Furthermore, pretreatment with recombinant AIBP protein in organotypic hippocampal slice culture, an ex vivo system, showed a protective effect against dendritic spine loss caused by Aβ overproduction. Taken together, this study is the first report of a protective AIBP function in a mouse model of AD, highlighting its role in controlling Aβ deposition, Aβ-induced microglial activation, and neuronal cell death.

Disclosures: **Y. Kim:** None. **S. Choi:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent. **Y.I. Miller:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); patent. Other; scientific co-founder of Raft Pharmaceuticals LLC.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.27

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

Support: NIGMS P20 GM103429

Title: Iron suppresses NMES1 in cultured murine microglia leading to hyperactivation in response to amyloid-beta stimulation

Authors: *B. BISHOP, S. EWING, E. MORGAN, M. VARGAS, D. DONLEY;
Harding Univ., Searcy, AR

Abstract: Title: Iron suppresses NMES1 in cultured murine microglia leading to hyperactivation in response to amyloid-beta stimulation
Authors: Bre Bishop; Savannah Ewing; Elizabeth Morgan; Mariel Vargas; David Donley
Affiliation: Department of Biology, Harding University
Abstract: Microglial cells are brain-resident immune cells that mediate inflammation during injury and disease. Microglia activate in response to damage or foreign matter. During diseases such as Alzheimer's disease, deposition of amyloid-beta-42 (Aβ42) causes chronic microglial activation. Iron accumulation is coincident with Aβ42 toxicity in microglia, however mechanisms that mediate a potential interaction are not well understood. We identified the Normal Mucosa of Esophagus-Specific Gene 1 (NMES1) as a putative intersection point of iron and Aβ42 using proteomic analysis of mouse immortalized microglial (IMG) cells. We found Aβ42 increases NMES1 expression whereas iron decreases it. Inflammatory activation is driven, in part, by a metabolic shift in microglia. Under conditions of inflammation NMES1 helps regulate metabolic activity by replacing the NDUFA4 subunit of cytochrome c oxidase. This mechanism is associated with decreased oxidative stress. The goal of our current study was to elucidate the role of NMES1 in microglial responses to disease-associated stimuli, including Aβ42. Using flow cytometry, we found that NMES1 and NDUFA4 expression was increased in response to Aβ42 stimulation but cells that were also cultured in elevated iron had decreased NMES1 associated with elevated reactive oxygen species. To address the role of NMES1, we stimulated cultured IMG cells with Aβ42 in the presence and absence of NMES1 silencing RNA. We found that silencing NMES1 increased CD68 expression, a marker of inflammatory activation, in cells stimulated with Aβ42. These results are consistent with NMES1 as an inflammatory braking mechanism. The NMES1 gene, *C15orf48*, also produces a microRNA,

mir-147b, that is implicated in suppressing inflammation. Cells transfected with a mir-147b mimic demonstrate altered patterns of activation in response to Aβ42 suggesting that both mir-147b and NMES1 contribute to the regulation of microglial-mediated inflammation. More work is needed to understand the complete role of mir-147b and NMES1 on Alzheimer's-associated neuroinflammation. However, taken together our results suggest that elevated iron may promote microglial activation in response to disease-associated stimuli by suppressing regulators of inflammation, namely NMES1 and mir-147b.

Disclosures: B. Bishop: None. **S. Ewing:** None. **E. Morgan:** None. **M. Vargas:** None. **D. Donley:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.28

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

Support: CIHR Grant SVB-158618 to MF

Title: Levels of serum anti-nucleosome antibodies are regulated by female gonadal hormones in the 3xTg-AD mouse model of Alzheimer's disease

Authors: *M. FAHNESTOCK[1], W. SONG[1], B. SAKIC[1], D. MA[2];
[1]Dept. of Psychiatry & Behavioural Neurosciences, [2]Dept. of Pathology & Mol. Med., McMaster Univ., Hamilton, ON, Canada

Abstract: Rationale: Sex-dependent discrepancies in disease prevalence and serum autoantibody levels are observed in patients and animal models of Alzheimer's disease (AD). Using the 3xTg-AD mouse model, we previously reported that adult males show early systemic autoimmunity along with lack of plaque/tangle pathology. Conversely, adult females display less severe autoimmunity and exhibit AD-like pathology. **Objectives:** The present study examines whether gonadal hormones play a role in sex differences in serum autoantibody levels in the 3×Tg-AD mouse model of AD. **Methods:** 3×Tg-AD and wild-type (WT) mice were gonadectomised or sham-operated at 3 months of age. At 7 months of age, the animals were assessed for serum autoantibodies by indirect immunofluorescence for antinuclear antibodies (ANA) and by line-immunoblot assay for an additional 16 monospecific autoantibodies including anti-nucleosome antibodies. **Results:** There were significant differences between the genotypes in ANA levels, with the major target antigens confirmed as nucleosomes. The results of ANA and anti-nucleosome assays were combined for further analysis. Further analysis revealed: 1) the level of serum autoantibodies in male 3×Tg-AD mice was higher than in female 3×Tg-AD animals, and this was not altered by orchiectomy; 2) 3×Tg-AD female mice displayed a significantly lower level of serum autoantibodies than WT females; 3) ovariectomy further reduced the level of serum autoantibodies in female 3×Tg-AD mice, whereas gonadectomy in

males had no impact on serum autoantibodies. **Conclusions:** In 3xTg-AD mice, the major target of AD-associated serum autoantibodies is the nucleosome. Our results show that female gonadal hormones play a role in regulation of serum autoantibody levels.

Disclosures: **M. Fahnestock:** None. **W. Song:** None. **B. Sakic:** None. **D. Ma:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.29

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

Support: NIH/NIA R01AG060718-01A1

Title: Sub-chronic FK506 treatment reduces Aβ and Tau pathology in 7-month-old 3xTg-AD mice

Authors: *A. FRACASSI, M. MARCATTI, B. TUMURBAATAR, W.-R. ZHANG, G. TAGLIALATELA;
Neurol., Univ. of Texas Med. Br., Galveston, TX

Abstract: Alzheimer's disease (AD) is the most common form of neurodegenerative dementia. While major histopathological hallmarks of AD are amyloid-β (Aβ) plaques and neurofibrillary tau tangles, soluble oligomeric forms of Aβ and tau (TauO) have been identified as the most neurotoxic species involved in synaptic degeneration. Previous studies associate alterations of the phosphatase calcineurin (CaN) to AβO-driven toxicity. We previously have shown that CaN mediates both the neurotoxic and cognitive effects of AβO and elevated CaN levels have been also shown in AD patients, suggesting a central role of CaN in AD onset and/or clinical progression. Our previous studies in mouse models demonstrated that an acute treatment with the CaN inhibitor FK506/Tacrolimus in Tg2576 acutely injected intracerebroventricularly with AβO restores memory function in these cognitively impaired animals. Moreover, we reported absence of AD in aging human chronically treated with KF506 following solid organ transplant. To deeper investigate the involvement of CaN in AD onset and progression, we used a 3xTg-AD mouse model presenting both Aβ and Tau aggregates. 7-month-old 3xTg-AD mice were chronically treated for two weeks with intraperitoneal administration of either FK506 (1mg/kg) or PBS. Using immunofluorescence and western blotting we analyzed the levels of CaN, total Tau, pTau and Aβ in hippocampal (dentate gyrus-DG, CA1 and CA3) and cortical areas (frontal and parietal-occipital cortex) in our experimental groups. We found significantly decreased levels of CaN in DG, CA1 and CA3 and cortical areas in FK506 treated 3xTg-AD mice as compared to 3xTg-AD mice injected with PBS. Furthermore, performing double staining for Aβ and total Tau, FK506 treated mice displayed significant lower levels of Aβ and Tau in all the considered areas as compared to PBS-treated 3xTg-AD mice. These data suggest that the treatment with FK506 might trigger some mechanisms involved in the elimination of Tau and

Aβ, *i.e.* autophagy, as confirmed by the analyses of autophagic pathway (Beclin1, autophagy-related proteins - ATGs, LC3, Lamp1). Our data confirm the role of CaN as a key player in AD. The reduced key AD pathology following CaN inhibition with FK506 suggests the use of an FDA approved drug as a potential treatment with rapid clinical translation to slow the progression of the disease.

Disclosures: A. Fracassi: None. **M. Marcatti:** None. **B. Tumurbaatar:** None. **W. Zhang:** None. **G. Taglialatela:** None.

Poster

529. Neuroinflammation: Alzheimer's and Amyloid: *In Vivo* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 529.30

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

Support: AHA CDA 856826

Title: The role of long-chain acyl-CoA synthetases 3 (ACSL3) in age-related vascular dementia

Authors: L. XU[1], Y. ZHANG[1], C. LI[1], G. CLEMONS[2], C. T. CITADIN[2], C. H. ACOSTA[2], H. LIN[1], R. H.-C. LEE[1], ***C. Y.-C. WU**[1];
[1]Neurol., [2]Cell. Biol. and Anat., LSU Hlth. Sci. Ctr. Shreveport, Shreveport, LA

Abstract: Alzheimer's disease (AD) is a progressive neurodegenerative diseases which is the most common cause of dementia in the United States. Aged and gender are the major risk factors for AD. Women are more likely to develop a rapid progression of dementia than men with a greater risk of developing vascular dementia as compared to males. Cerebral blood flow derangements found in AD are thought to be the major cause of brain dysfunction and neurological deficits. AD-mediated hypoperfusion plays a vital role in vascular dementia-related neuroinflammation, mitochondrial dysfunction, neuronal cell death, and neurological deficits. Therefore, the major challenge is to alleviate the development and progression of vascular dementia. We previously discovered that expression of long-chain acyl-CoA synthetase 3 (ACSL3) in brain regions were significantly decreased with the aged AD mice. Specific agonist of ACSL3 alleviated AD-mediated neuroinflammation and mitochondria dysfunction, with enhanced neuronal survival and improved functional learning/memory. Our preliminary data suggest that decreased ACSL3 is detrimental in an aged 3xTg-AD mouse model (enhanced β-amyloid and tau aggregation and accumulation). Our central hypothesis is that ACSL3 is critical for age-related brain function to maintain the mitochondria function, prevent neuroinflammation, and learn/memory degradation. We propose to explore the pathophysiological role of ACSL3 in the aged brain. The present study can lead to novel therapies/targets against AD brain progression by investigating the pathophysiological role of ACSL3 in neuroprotection.

Disclosures: L. Xu: None. **Y. Zhang:** None. **C. Li:** None. **G. Clemons:** None. **C.T. Citadin:** None. **C.H. Acosta:** None. **H. Lin:** None. **R.H. Lee:** None. **C.Y. Wu:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.01

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

Support: NIH
UH Foundation

Title: An N-terminal core sequence from beta amyloid activates PI3K/mTOR-dependent downstream signaling to reverse full-length amyloid-beta-induced synaptic dysfunction

Authors: *R. M. TAKETA, R. NICHOLS;
Univ. of Hawaii, Honolulu, HI

Abstract: Alzheimer's disease (AD) is a progressive neurodegenerative disorder manifested by memory impairment and subsequent cognitive decline. Currently, there is no cure and the exact cellular mechanisms underlying the disease are not fully known. Aggregation of beta-amyloid (Aβ) peptide into insoluble Aβ-plaques is one of two prominent histopathological hallmarks of the disease. A series of Aβ peptides is produced through sequential enzymatic processing of the amyloid precursor protein (APP), with the most prominent, plaque-associated Aβ peptide being 42 amino acids long (Aβ42). Accumulation of Aβ42 to pathological levels (μM) contributes to synaptic impairment and eventual synapse loss. However, at physiological concentrations (pM-nM), we and others have shown that soluble Aβ acts as a neuromodulator enhancing synaptic plasticity, indicative of a hermetic action of this neuropeptide.
We have previously reported that an endogenously cleaved N-terminal Aβ fragment (N-Aβ1-15,16) and an essential core sequence, YEVHHQ (N-Aβ Core) within the fragment, are neuroprotective against Aβ42-induced oxidative stress, synaptic and behavioral impairment and apoptosis in in vitro and ex vivo studies. Notably, in the 5xFAD (familial AD) mouse model the N-Aβ Core injected bilaterally into the hippocampus reversed spatial memory deficits. We recently reported that the N-Aβ Core reversal of the impairment in synaptic plasticity in hippocampal slices in long-term potentiation (LTP) is PI3K/mTOR-dependent. However, the downstream molecular mechanisms for the N-Aβ Core at the synaptic level have yet to be uncovered, nor do we understand the relation of synaptic impairment to behavioral deficits.Here, we further investigated the downstream signaling engaged by the N-Aβ Core at the synapse within primary hippocampal cultures, organotypic slice cultures, and acute hippocampal slices. The sequential timing of specific signaling pathways engaged at the synapse by the N-Aβ Core without or with Aβ42 will be assessed before and after induction of LTP via electrical and chemical stimulation in acute slices as well as slice cultures and primary hippocampal neuronal cultures, respectively. Immunohisto/cytochemistry and immunoblots of protein lysates were assessed for differential expression in downstream synaptic translation proteins, focusing on regulators of synaptic protein translation.
Elucidating the specific molecular mechanisms for reversal by the N-Aβ Core of synaptic

impairment and loss may provide insights into Aβ-linked behavioral impairment, while revealing novel targets with therapeutic potential for AD.
Funding: NIH and the UH Foundation

Disclosures: R.M. Taketa: None. **R. Nichols:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.02

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

Support: NIH Grant R01AI121012
NIH Grant R21AI154211
John Sealy Distinguished Chair in Alzheimer's diseases

Title: Alzheimer'S disease circulating-exosomes are proinflammatory in recipient brain microvascular endothelial cell in an RNA-cargo dependent manner

Authors: *J. BEI[1], Y. QIU[1], C. NATARAJAN[1], M. FELICELLA[1], B. KRISHNAN[1], A. GAITAS[2], X. FANG*[1], B. GONG*[1];
[1]Univ. of Texas Med. Br., Galveston, TX; [2]Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Capillaries represent 90% of the endothelium in human brain vasculature and are the major sites of the BBB, which is formed by a tightly sealed monolayer of brain microvascular endothelial cells (BMECs), endothelial tight junctions (TJs), and adherens junctions (AJs), working in concert with pericytes to maintain a homeostatic microenvironment for neuronal function. Breakdown of the BBB in Alzheimer's disease (AD) has been documented by multiple independent postmortem human studies, revealing BMEC degeneration, TJ disruption, microhemorrhages, and hemosiderin deposits. Furthermore, in early-stage AD patients treated with amyloid-modifying therapies, amyloid related imaging abnormalities (ARIA)-vasogenic edema and ARIA-microhemorrhage have been identified and both are related to BBB dysfunction. BBB dysfunction was proposed as an early biomarker of human cognitive dysfunction independent of Tau- and β-amyloid. Aging is the key risk factor for AD, but the correlation between cellular senescence and BBB dysfunction in AD remain unclear. EVs transfer functional mediators to neighboring and distant recipient cells. There has been increased interest in studying the potential of exosomes (Exos) in AD, mainly focusing on their protein contents, i.e. Tau- and β-amyloid (Aβ)-containing Exos. We purified circulating Exos from severe AD patient (AD-Exos) and Ctl (Ctl-Exos) sera using size-exclusion chromatography (SEC), which avoids aggregation and decreased integrity of Exos, compared with ultracentrifugation. EV-specific assays validated Exo particle size (50-150 nm) and qualities, exclusively from synaptosome-sized particle (0.6 μm to 1.6 μm). Exo size and morphology were not significantly altered between groups. Exosomal particle counts were measured using our

nanoparticle tracking analysis system, which showed that Exo counts are not statistically different between groups. Next, we treated human BMECs with AD-Exos vs. Ctl-Exos for 72 hrs prior to measuring the transendothelial electrical resistance, fluidic force microscopy-probed lateral binding force between living BMECs, and immunofluorescence staining of major TJ and AJ proteins. We found that AD-Exos induced loss of paracellular AJ VE-cad and attenuated the recipient BMECs barrier function. To identify the functional cargos in AD-Exos, we employed saponin-assisted active permeabilization to pretreat exosomal cargos with RNase. Such pretreatment mitigated the detrimental effects of AD-Exos on recipient BMECs barrier function. Conclusion: AD-Exos weaken the BBB in an exosomal RNA cargo-dependent manner.

Disclosures: J. Bei: None. **Y. Qiu:** None. **C. Natarajan:** None. **M. Felicella:** None. **B. Krishnan:** None. **A. Gaitas:** None. **X. Fang*:** None. **B. Gong*:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.03

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

Support: NSERC Grant

Title: Soluble Amyloid Beta facilitates excitatory transmission and AMPA receptor phosphorylation in Layer II of the Rat Medial Entorhinal Cortex

Authors: *M. E. SUVANTO[1], O. J. OLAJIDE[2], J. SARAGOSA[1], C. A. CHAPMAN[1]; [1]Concordia Univ., Montreal, QC, Canada; [2]Anat. (Neuroscience Unit), Univ. of Ilorin, Ilorin, Nigeria

Abstract: The earliest symptoms of Alzheimer's Disease (AD) include deficits in episodic memory and spatial navigation, both linked to neurodegeneration in the temporal lobe that begins in layer II of the entorhinal cortex (EC). Toxic accumulation of soluble amyloid beta peptides (Aβ) is thought to contribute to the onset of neurodegeneration. In the hippocampus specifically, elevated Aβ can facilitate postsynaptic Ca^{2+} influx via NMDA glutamate receptors, and increased Ca^{2+} can lead to the phosphorylation of AMPA receptors that can promote synaptic excitability and contribute to excitotoxicity. Aβ-mediated excitotoxicity and the effects of Aβ on excitatory transmission have been studied extensively in the hippocampus, but less is known about the effects of Aβ on excitatory synaptic transmission in layer II of the EC. We studied the effects of incubating 400 μm horizontal slices of Long-Evans rat medial EC in 100 nM $Aβ_{(1-42)}$ (n=24) or a dimethyl sulfoxide control (n=25) for 45 minutes to 3 hours on excitatory synaptic transmission in layer II EC using field excitatory postsynaptic potentials (fEPSPs) in vitro. Further, we examined the effects of including the NMDA receptor blocker D-AP5 50 μM in the medium containing control slices (n-12) or slices exposed to Aβ (n=13). All experiments were blinded. The amplitude of fEPSPs increased in slices incubated in Aβ relative to control slices.

The effect was blocked by constant bath application of D-AP5, showing the activation of NMDA receptors is needed for the facilitation of synaptic excitability. The facilitation of synaptic responses induced by Aβ likely results from activation of postsynaptic NMDA receptors that enhance Ca^{2+} influx. Increased Ca^{2+} within the neuron can promote the phosphorylation of AMPA receptors via activation of CaMKII. We therefore assessed tissue samples of the medial EC using Western blotting to measure the expression of CaMKII, AMPA receptors, and phosphorylated AMPA receptors following a one-hour incubation in either 100 nM $Aβ_{(1-42)}$ (n=6) or a dimethyl sulfoxide control (n=6). We then replicated these experiments with the inclusion of 50 μM of D-AP5 to both incubations (n=6 both groups respectively). Results of protein analysis were consistent with changes in AMPA receptors associated with exposure to Aβ.

Disclosures: M.E. Suvanto: None. **O.J. Olajide:** None. **J. Saragosa:** None. **C.A. Chapman:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.04

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

Title: The role of NOGAR1 on APP/PSEN1 mutant neuronin AD-related early endosomal defects

Authors: *X. RONG[1,2], H. FANG[1,2], J. LEE[1,2], Y. GAO[1,2], T. M. DAWSON[1,2,3,4], V. L. DAWSON[1,2,3,5], J. XU[1,2,6];
[1]Neuroregeneration and Stem Cell Programs, Inst. for Cell Engin., [2]Dept. of Neurol., [3]Solomon H. Snyder Dept. of Neurosci., [4]Pharmacol. and Mol. Sci., [5]Dept. of Physiol., [6]Human iPS Cell Core for Neurodegenerative Dis. Res., Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: Background: Alzheimer's disease (AD) is a major devastating neurological disorder. Lack of effective treatments for AD prompts a broad exploration of disease-related cellular biology and therapeutic targets. Neuronal endosome enlargement has been identified as an early cellular defect in AD, a significant pathology that can be regulated by oxygen & glucose along the endocytic pathway critically involved in brain Aβ clearance. **Innovation:** In our study, the primary innovation is the identification of a novel major regulator through decoding the fundamental pathways associated with oxygen & glucose and energy-demanding N-methyl-D-aspartate receptors (NMDAR)-dependent cognition, which likely have pivotal roles in the early stages of AD pathogenesis. The newly discovered major regulator, NMDAR-, oxygen & glucose-associated regulator 1 (NOGAR1) may function as a regulatory nexus for crosstalk between early endosomes and fundamental pathways in familial and sporadic AD (fAD and sAD). **Significance:** Neurodegenerative processes in AD may be the consequences of NOGAR1-regulated early endocytic stability or vulnerability to oxygen & glucose and NMDAR stress,

either as contributory or collateral pathologies. Secondly, the human neuronal cell model we developed is a system that closely relates to the adult human cortex. Our hiPSC derived neural cells include physiological representation of functionally balanced excitatory & inhibitory cortical neurons and mature neuronal phenotypes, providing human neurons to study AD-related phenotypes using a genuine human model system. **Aim:** Our first aim was to examine the role of NOGAR1 on early endosomal phenotypes in APP and PSEN1 mutant human iPSC-neurons, and the second aim was to examine the impacts of NOGAR1 on the neuropathology of AD. **Results:** Our results show that NOGAR1 *regulates synaptic plasticity and that R*ab5 expression is dramatically increased in the NOGAR-*KO group* compared with its corresponding isogenic WT control. Additionally, NOGAR1 depletion leads to an increase in the soluble/insoluble ratios of Aβ, β-CTFs, and pTau-Thr217. **Conclusion:** In conclusion, NOGAR1 loss of function in APP and PSEN1 mutant human iPSC-neurons exaggerates their early endosome abnormalities. Simultaneously, NOGAR1 loss of function is linked to enhanced Aβ and pTau-induced pathology. Uncovering the role of OGD- and NMDAR- related common major regulator-NOGAR1, which regulates early endosomes, may lead to the identification of alternative therapeutic targets for early-stage AD.

Disclosures: X. Rong: None. **H. fang:** None. **J. lee:** None. **Y. gao:** None. **T.M. Dawson:** None. **V.L. Dawson:** None. **J. Xu:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.05

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

Support: NIH R21AG064630
NIH P20GM109098

Title: Amyloid beta mediated impact on the nanoscale organization of dendritic spines

Authors: *J. SCRIPTER, L. L. MORAIS, G. JONES, M. HRUSKA;
Neurosci., West Virginia Univ., Morgantown, WV

Abstract: Alzheimer’s disease (AD) is associated with abnormal spine morphology and structural plasticity. The morphological aberrations of spines are associated with changes in the size and molecular composition of post-synaptic densities (PSDs). The discovery that synaptic nanomodules that scale in number with spine size and serve as the building blocks for pre- and post-synaptic macrostructures suggests the logic for the nano-organization of the synapse. Yet, it is unknown how synaptic nanoarchitecture is impacted before spines begin to disappear in AD. To determine that synaptic nanoarchitecture is altered in AD, we focused on the effects of A-beta oligomers on nano-organization of the major components of PSDs and active zones, PSD-95 and Bassoon, in dendritic spines after acute and chronic exposure to A-beta (1- 42) in vitro and in the

familial mouse model (5xFAD). Our approach combines multi-color STimulated Emission Depletion (STED) nanoscopy of endogenous synaptic proteins with conventional confocal imaging of spine morphology, enabling correlation of spine size with changes in pre- and post-synaptic nanoarchitecture. Acute A-beta treatment (24 and 48 hours) of DIV21 EGFP-transfected rat cortical neurons significantly increases the proportion of spines without PSD-95 nanomodules, while reducing the number of spines with only one PSD-95 nanomodule. Remarkably, the proportion of spines with multiple PSD-95 nanomodules is not affected. Notably, the number of Bassoon nanomodules in small and large spines is not different between control and A-beta conditions. We have found similar effects on pre- and post-synaptic nano-organization in six-month-old 5xFAD mice which exhibit high loads of A-beta. However, these features of spine nano-organization begin to be disrupted as early as two months of age even when A-beta loads are low, indicating the robust impact of A-beta toxicity on synaptic nanoarchitecture. Finally, we found that A-beta impacts the ability of synapses to alter their nano-organization in response to chronic changes in neuronal activity, suggesting these morphological aberrations have consequences for synaptic function. Overall, these data indicate that synapses in AD exhibit morphological and nanoscale abnormalities likely mediated by the distinct action of A-beta on small and large spines.

Disclosures: **J. Scripter:** None. **L.L. Morais:** None. **G. Jones:** None. **M. Hruska:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.06

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

Support: Swedish Research Council grant 2018-02952 to BB
EXODIAB grants 2019,2020 to BB, ZJ, SVK
Swedish Brain Foundation to BB
Swedish Research Council 2018-02503 to JPL

Title: Age-dependent normalization by insulin of GABA signalling in dorsal hippocampal dentate gyrus granule cells in a tg-APPSwe mouse model of Alzheimer disease.

Authors: O. NETSYK[1], H. HAMMOUD[2], A. TAFRESHIHA[2], S. V. KOROL[2], Z. JIN[2], J.-P. LI[2], ***B. BIRNIR**[2];
[1]Uppsala University; Current address Stockholm Univ., Uppsala, Sweden; [2]Uppsala Univ., Uppsala Univ., Uppsala, Sweden

Abstract: Aim. Insulin is a metabolic hormone regulating peripheral glucose homeostasis and brain function. We examined in tg-APPSwe mice, an Alzheimer disease (AD) model, and their wild-type littermates, if basal, tonic γ-aminobutyric acid (GABA)-activated synaptic and extrasynaptic currents in hippocampal dentate gyrus (DG) granule cells differed and were

modulated by insulin. **Methods.** GABA-activated currents were recorded in DG granule cells in dorsal hippocampal brain slices, from 5-6 or 10-12 (aged) months old wild-type and tg-APPSwe mice, in the absence or presence of insulin, by whole-cell patch-clamp electrophysiology. **Results.** Insulin receptor labelling was detected in the DG granule cell layer of the dorsal hippocampus. Fast and slow spontaneous inhibitory postsynaptic current (sIPSC) densities were similar in wild-type and tg-APPSwe mice at 5-6 months whereas the sIPSC densities were significantly decreased in the aged tg-APPSwe mice. Insulin (1 nM) did not modulate the sIPSCs density in wild-type mice but increased the fast sIPSC density in the aged tg-APPSwe mice. The extrasynaptic tonic current density was increased in the tg-APPSwe mice relative to the wild-type littermates but, only in the aged tg-APPSwe mice did insulin decrease the extrasynaptic current density. **Conclusion**. In hippocampal dorsal DG granule cells of 5-12 months old wild-type mice, the GABA signalling was stable and not altered by insulin. In contrast, in 5-12 months tg-APPSwe mice, extrasynatic GABA signalling was increased. In the aged tg-APPSwe mice, the synaptic GABA signalling was, in addition, decreased. Importantly, insulin normalized the current densities to wild-type values in the aged tg-APPSwe mice.

Disclosures: O. Netsyk: None. **H. Hammoud:** None. **A. Tafreshiha:** None. **S.V. Korol:** None. **Z. Jin:** None. **J. Li:** None. **B. Birnir:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.07

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

Support: CIHR Grant MOP-125901
NSERC Grant RGPIN-05477-2017
Research Manitoba Masters Studentship
Alzheimer Society of Canada Doctoral Fellowship
Alzheimer Society of Manitoba Graduate Studentship
McCrorie-West Family Fellowship for Alzheimer Research
Eirikur and Thorbjorg Stephanson Scholarship

Title: Understanding the role of NMDAR-STIM-Panx1 signaling in amyloid-β oligomer mediated synaptotoxicity

Authors: *C. S. PATIL, N. E. LAVINE, M. F. JACKSON;
Univ. of Manitoba, Univ. of Manitoba, Winnipeg, MB, Canada

Abstract: Introduction: Alzheimer’s disease is associated with increased accumulation of soluble amyloid-β oligomers (AβOs). AβOs are known to initiate toxic cascades that disrupt the function of glutamatergic synapses. In this regard, we now show that pannexin-1 (Panx1) channels, that are known to be activated downstream of NMDA receptors (NMDARs), are

implicated in AβO-mediated toxicity. Further, we found that NMDAR-initiated Panx1 activation is regulated by physical interaction between Panx1 and endoplasmic reticulum resident stromal interacting molecules (STIMs). In AβO treated neurons, we show that disrupting Panx1-STIM interaction using an interfering peptide of our own design had a protective effect on synapse function. Methods: Electrophysiology experiments with minimum 3 independent experimental replicates were conducted to study Panx1 activity in hippocampal neurons cultured from wildtype (WT) or Panx1 knockout (KO) mice. HEK 293T cells were used to map the domain of Panx1-STIM interaction. Results: Panx1 current downstream of NMDAR stimulation was increased in neurons treated with AβOs. This was coupled with reduced frequency of excitatory synaptic events in WT neurons. Interestingly, Panx1 KO neurons were protected from the toxic effects of AβOs. Moreover, upon knockdown of STIMs, NMDAR-initiated Panx1 currents were abrogated. Thus, we reasoned that identifying the Panx1-STIM interaction interface can serve as a direction to develop novel therapeutics able to prevent the toxic effects of AβOs. Using a series of deletions within Panx1 N- or C-terminal domains, we identified that the Panx1-STIM interaction interface resided within the Panx1 N-term. Notably, our identified region regulates Panx1 activation downstream of NMDAR stimulation in neurons. Thus, we generated a cell permeable peptide that interferes with the Panx1-STIM interaction (Tat-Panx1). Confirming the importance of the Panx1 N-term in regulating NMDAR initiated STIM-dependent Panx1 activation, neurons treated with Tat-Panx1 peptide but not with Tat alone (negative control peptide) showed inhibition of Panx1 currents. Importantly, neurons treated with Tat-Panx1 were protected from the toxic effects of AβOs. Thus, preventing Panx1 activation in AβO treated neurons is neuroprotective. Conclusion: We showed that inhibiting Panx1-STIM interaction is a candidate therapeutic strategy to prevent synaptotoxicity mediated by AβOs. Using Tat-Panx1 peptide, we can uncover the physiological and pathological functions mediated by NMDAR-STIM-Panx1 signaling.

Disclosures: **C.S. Patil:** ; Co-inventor for a PCT patent application filed for Tat-Panx1 peptide. **N.E. Lavine:** ; Co-inventor for a PCT patent application filed for Tat-Panx1 peptide. **M.F. Jackson:** ; Co-inventor for a PCT patent application filed for Tat-Panx1 peptide.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.08

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

Support: Alzheimer's Society grant 449: AS-PhD-18-007

Title: Leptin prevents amyloid-beta-induced internalisation of AMPA receptors and aberrant targeting of phosphorylated tau via PI 3 kinase signalling

Authors: *K. HAMILTON, K. MORROW, J. HARVEY;
Univ. of Dundee, Dundee, United Kingdom

Abstract: Accumulation of amyloid-beta (Aβ) and hyperphosphorylated tau are hallmarks of Alzheimer's disease (AD). Recent studies show that Aβ promotes tau phosphorylation causing migration of tau from axons to dendritic spines and synapses, resulting in synaptic impairment. Disruption of leptin production and signalling has been observed in AD patients. Various studies have identified neuroprotective properties of leptin in models of Aβ-related toxicity. However, the effects of leptin on tau-related synaptic dysfunction are unclear. Here, we show that $A\beta_{1-42}$ treatment (1μM, 1hr) to primary hippocampal neurons causes a significant increase in dendritic p-tau compared to control (167 ± 9%, $p<0.001$, n=36 dendrites), and a significant increase in % colocalisation of p-tau and PSD-95 (synaptic marker) from 43 ± 1.4% to 57 ± 1.4% ($p<0.001$, n=36 dendrites). Leptin (10nM) prevents this aberrant targeting of p-tau to dendrites and synapses as dendritic p-tau levels and % colocalisation are not significantly different from control in leptin+$A\beta_{1-42}$ treated neurons ($p>0.05$, n=36 dendrites). PI 3 kinase activation was identified as the likely signalling mechanism underlying the protective effects of leptin. Wortmannin treatment (PI3K inhibitor, 50nM, 1hr) ablated the protective effects of leptin, resulting in a significant increase in dendritic p-tau in wortmannin+$A\beta_{1-42}$+leptin treated neurons (150 ± 6% of control, $p<0.001$, n=36). GluA1-containg AMPA receptors (AMPARs) are key receptors involved in synaptic transmission. Here, we show that $A\beta_{1-42}$ treatment (1μM) induces internalisation of GluA1-containing AMPARs from synapses, as indicated by a significant decrease in % colocalisation of surface GluA1 and PSD-95 following 20 (control = 55 ± 0.9%, $A\beta_{1-42}$ = 42 ± 1.4%, $p<0.001$, n=36;), and 60 minute incubation with $A\beta_{1-42}$ (control = 53 ± 1.0%, $A\beta_{1-42}$ = 46 ± 0.8%, $p<0.001$, n=60). Leptin prevents $A\beta_{1-42}$-induced GluA1 internalisation at both timepoints as % colocalisation of surface GluA1 and PSD-95 are not significantly different from control in leptin+$A\beta_{1-42}$-treated neurons ($p>0.05$). PI3K activation was again identified as the likely signalling mechanism underlying the protective effects of leptin. Wortmannin treatment (50nM) ablated the protective effects of leptin, resulting in a significant decrease in % colocalisation of surface GluA1 and PSD-95 in wortmannin+$A\beta_{1-42}$+leptin treated neurons following 20 (control = 55 ± 0.9%, WM+A+L = 43 ± 1.2%, $p<0.001$, n=36) and 60 minute $A\beta_{1-42}$ treatment (control = 53 ± 1.0%, WM+A+L = 46 ± 1.0%, $p<0.001$, n=60). Together, these data further validate the leptin system as a therapeutic target in AD.

Disclosures: K. Hamilton: None. **K. Morrow:** None. **J. Harvey:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.09

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

Title: Cellular mechanism of amyloid β oligomers suppressing GIRK channel activity

Authors: *H. LUO[1], E. MARRON FERNANDEZ DE VELASCO[2], K. D. WICKMAN[3]; [1]Univ. of Minnesota, [2]Univ. of Minnesota, Minneapolis, MN; [3]Dept Pharmacol, Univ. Minnesota, Minneapolis, MN

Abstract: Amyloid β oligomers (AβOs) are one of the primary pathogenic factors in Alzheimer's Disease and have been shown to induce excitotoxicity in the hippocampus in both acute and chronic models. While many studies have elucidated AβOs' pathological effects on glutamatergic signaling, limited studies focus on the impacts of AβOs on GABAergic inhibitory signaling and its possible role in pathogenesis. Our group and others have established G protein-coupled inwardly-rectifying potassium (GIRK) channels as critical mediators of GABA(B) receptor-dependent signaling, impacting neuronal excitability, synaptic plasticity, and cognitive function. AβOs are shown to downregulate GIRK channel mRNA and protein expression in the rodent hippocampus, but no mechanistic studies have been done to verify the link between GIRK channel downregulation and AβO-induced synaptic and cognitive deficits. To study whether GIRK channels are one of AβOs' early targets whose changes contribute to the downstream pathology, we investigate the effects of synthetic AβOs on cultured mouse hippocampal neurons. Data were analyzed using Welch's t-test or one-way ANOVA with Tukey's multiple comparisons test (n = 16-18), as appropriate. We found that both GABA(B)-evoked somatodendritic whole-cell currents (100 µM baclofen) and directly-activated GIRK channel-mediated currents (10 µM ML297) were significantly suppressed following AβO incubation (0.5 µM) in neurons (baclofen: $p = 0.0391$, ML297: $p = 0.0412$). The effect was seen after only 3 hours of incubation. The suppression observed was successfully blocked by co-incubating with a metabotropic glutamate receptor 5 (mGlu5R) antagonist (10 µM MTEP, baclofen: $p > 0.9999$), suggesting that the suppression of GABA(B)R-GIRK currents is likely through AβO-induced mGlu5R activation. In addition, incorporating phosphatidylinositol 4,5-bisphosphate (PIP2) in the pipette solution rescued the current suppression evoked by AβOs (30 µM diC8PIP2, baclofen: $p > 0.9999$), indicating that a possible reduction in PIP2 (essential for GIRK activation) in the plasma membrane is involved in the early effects of AβOs on GIRK-dependent signaling, and is likely the consequence of mGlu5R activation. It is the first time that a possible mechanism (AβO-mGlu5R-PIP2) for the action of AβOs on GIRK-dependent signaling is investigated, ongoing behavioral tests are estimating the contribution of GIRK channel adaptation to the AβO-induced cognitive deficits.

Disclosures: H. Luo: None. **E. Marron Fernandez De Velasco:** None. **K.D. Wickman:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.10

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

Support: Direktør Emil Hertz og hustru Inger Hertz' Fond
Dagmar Marshalls Fonden
Solar Fonden af 1978
Lundbeck Foundation
Aarhus University, PhD scholarship

Title: Regulating sortilin proteolytic cleavage through a novel molecular switch with relevance to Alzheimer's disease

Authors: *M. OVERBY[1], J. A. LORENTSEN[2], N. A. BERGLUND[2], B. SCHIØTT[2], J. P. WEICK[3], H. K. MÜLLER[1];
[1]Clin. medicine, Aarhu Univ., Aarhus C, Denmark; [2]Chem., Aarhus Univ., Aarhus C, Denmark; [3]New Mexico Sch. of Med., New Mexico city, NM

Abstract: Deposits of extracellular amyloid plaques characterize Alzheimer's disease. Recent studies have shown that these amyloid plaques contain C-terminal fragments of a sorting receptor protein known as sortilin. The levels of sortilin C-terminal fragments are dramatically elevated in cortical lysates from Alzheimer's disease patients, indicating abnormal proteolytic cleavage of sortilin. Here we study how proteolytic cleavage of sortilin can be regulated through specific protein-protein interactions. Using yeast two-hybrid screening and complementary immunoprecipitation approaches, we identified two novel protein binding partners of sortilin, namely neuronal-specific gene 1 (NSG1) and neuronal-specific gene 2 (NSG2) A cell surface biotinylation assay on HEK-293-MSR cells transiently co-expressing sortilin and NSG1 or sortilin and NSG2 showed that NSG1 decreases cell surface expression to 55% ($p<0.01$), while NSG2 increases the cell surface expression up to 200% ($p<0.0001$). Co-immunoprecipitation studies on HEK-293-MSR lysate from the cell surface biotinylation assay showed that this was facilitated through an increase in the interaction between NSG1 ($p<0.05$) or NSG2 ($p<0.01$) to sortilin. Quantitative western blotting on lysate and media from cells co-expressing sortilin and NSG1 or sortilin and NSG2 showed that NSG1 increases sortilin proteolytic cleavage ($p<000.1$), while NSG2 decreases sortilin proteolytic cleavage ($p<0.01$). This was further investigated with an endocytosis-resistant sortilin mutant and a cleavage-resistant (the suspected cleavage area was deleted) sortilin mutant co-expressed with NSG1, which showed that proteolytic cleavage of sortilin was increased with the endocytosis-resistant mutant ($p<0.05$) while the cleavage-resistant sortilin mutant had no significant effect on the cleavage of sortilin. This proteolytic cleavage regulation by NSG1 and NSG2 was tested to see if it influenced neurotrophic signaling. HEK-293-MSR cells transiently co-expressing sortilin and NSG1 or sortilin and NSG2 were added media that included progranulin to see how this affected progranulin uptake through sortilin. NSG1 decreased progranulin uptake by 50% ($p<0.01$), while NSG2 had no significant effect. These data show that NSG1 and NSG2 can act as molecular switches to regulate the proteolytic cleavage of sortilin. NSG1 renders sortilin more susceptible to proteolytic cleavage while NSG2 protects sortilin from proteolytic cleavage.

Disclosures: M. Overby: None. **J.A. Lorentsen:** None. **N.A. Berglund:** None. **B. Schiøtt:** None. **J.P. Weick:** None. **H.K. Müller:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.11

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

Support: R01AG055909
R01NS117446
U01AG061356
R21AG053827
R01GM069808

Title: Identification of a role for synaptic vesicle protein networks in mediating Alzheimer's pathology in trisomy 21 and Alzheimer's disease neurons

Authors: *A. AYLWARD[1], C.-I. WU[2], E. A. VINTON[1], N. T. SEYFRIED[3], K. HEO[4], T. L. SCHWARZ[4], T. L. YOUNG-PEARSE[2];
[1]Brigham and Women's Hosp., Boston, MA; [2]BWH/Harvard Med. Sch., Boston, MA; [3]Emory Sch. Med., Emory Univ., Atlanta, GA; [4]BCH/Harvard Med. Sch., Boston, MA

Abstract: Trisomy 21 (T21) causes Down syndrome and an early-onset form of Alzheimer's disease (EOAD). Other forms of EOAD are caused by dominantly inherited mutations in APP or PSEN1 or 2. However, the majority of Alzheimer's disease (AD) is caused by sporadic forms of late-onset AD (LOAD) which are not associated with specific mutations. Here, we identify common pathways altered in familial Alzheimer's disease (fAD), LOAD, and T21 neurons to investigate potential phenotypes related to convergent disease-relevant pathways. To this end, we performed proteomic profiling using TMT-MS (Tandem Mass Tag Mass Spectrometry) comparing: 1) T21 to T21-reverted iPSC-derived neurons (iNs), 2) fAD to fAD-corrected iNs, and 3) LOAD to not cognitively impaired (NCI) iNs. Proteomic profiles of LOAD iNs shared many protein-level changes with fAD and T21 iNs despite having diverse genetic causes driving dementia in the humans from which they were derived. Analyses of differentially expressed proteins in these models of AD revealed alterations in axonal transport and synaptic vesicle cycling. To investigate potential functional consequences related to these convergent pathways, we performed assays of axonal trafficking and synaptic vesicle release using our iN experimental system. Both fAD and T21 iNs showed defects in axonal trafficking. In contrast, T21 neurons displayed enhanced synaptic vesicle release while fAD did not. However, iNs derived from a subset of individuals with LOAD also displayed enhanced synaptic vesicle release compared to NCI individuals. Taken together, our findings provide insights into the initial molecular alterations within human neurons that ultimately lead to synaptic loss and axonal degeneration in Down syndrome and AD.

Disclosures: **A. Aylward:** None. **C. Wu:** None. **E.A. Vinton:** None. **N.T. Seyfried:** None. **K. Heo:** None. **T.L. Schwarz:** None. **T.L. Young-Pearse:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.12

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

Support: NIH Grant R01 AI132414

Title: Evaluating the influence of HCMV infection on Alzheimer's Disease pathology

Authors: *J. W. ADELMAN[1], M. SCHUMACHER[2], B. S. O'BRIEN[1], S. S. TERHUNE[2], A. D. EBERT[1];
[1]Cell Biology, Neurobio. and Anat., [2]Microbiology and Immunol., Med. Col. of Wisconsin, Milwaukee, WI

Abstract: Alzheimer’s Disease (AD) is a common, incurable neurodegenerative disease characterized by progressive memory deficits, behavioral aberrancies, and death. While its underlying mechanisms remain unclear, previous studies have highlighted roles for protein aggregation (ex. Amyloid beta, phospho-Tau) and synaptic dysfunction in AD pathophysiology. Further, a growing body of research shows viral contribution to disease progression, particularly among human herpesviruses (HHVs). Human Cytomegalovirus (HCMV, HHV5) has been shown to have associations with various AD phenotypes, including altered calcium signaling, increased amyloid beta (Aß) accumulation, and downregulation of synaptic proteins. Therefore, we hypothesized that HCMV infection potentiates AD-like phenotypes in neuronal populations. Here, we used iPSCs derived from healthy and AD-affected individuals to generate a forebrain-specific 2D neuronal culture system and 3D cerebral organoids to directly assess any impacts of HCMV infection on AD pathology in human neurons. We first performed RNA sequencing on control and AD organoids with and without HCMV infection (HCMV clinical variant TB40/e-eGFP) and found an HCMV-dependent reduction in synaptic transcripts (ex. NMDAR and SNARE subunits) in both control and AD organoids; synaptic loss is an early AD-associated phenotype. We then used the 2D culture system to assess HCMV’s effects on neuronal function and aberrant protein accumulation. To evaluate function, calcium imaging and multielectrode arrays (MEAs) were employed to observe changes to calcium signaling and neuronal spiking, respectively. After a minimum of 50 days of neuronal differentiation, cells were either infected with HCMV or mock-treated. Calcium imaging at 7 days post-infection (7DPI) revealed HCMV-dependent reductions in both baseline calcium and potassium response in both control and AD lines. Complimenting this, MEA studies highlighted an HCMV-contingent reduction in evoked and spontaneous action potentials. Interestingly, these viral effects diverge from AD functional phenotypes, wherein both calcium influx and neuronal firing are typically increased. Finally, preliminary data from HCMV-infected control neurons exhibit a slight increase in both intracellular Aß1-42 and p-Tau, whereas ELISAs conducted on neuron-conditioned media revealed HCMV-dependent reductions in extracellular Aß42/40 ratios. Taken together, these results highlight a potentially complex relationship between HCMV infection and AD pathology such that HCMV infection may both induce and suppress AD-relevant phenotypes regardless of genetic background.

Disclosures: J.W. Adelman: None. **M. Schumacher:** None. **B.S. O'Brien:** None. **S.S. Terhune:** None. **A.D. Ebert:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.13

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

Title: Facilitation of alpha 7 nicotinic acetylcholine receptors by amyloid beta (1-42) peptide

Authors: *J. FARLEY[1], J. B. ANDERSON[2];
[1]Neurosci., Indiana Univ., Bloomington, IN; [2]Psychological and Brain Sci., Indiana Univ. Bloomington, Bloomington, IN

Abstract: Soluble oligomeric forms of amyloid beta peptide(s) [Aβ] suppress activation of several nicotinic acetylcholine receptors (nAChRs). This may contribute to Alzheimer's Disease (AD) neuropathology. The alpha 7 (α7) nAChR plays an important role in learning and memory and is situated in neuronal circuits that regulate activity of the hippocampus and cortex. Inhibition of α7 nAChRs (α7 Rs) by high Aβ concentrations [nanomolar (nM) to micromolar (μM)] has been proposed to contribute to the synaptic dysfunction and calcium dyshomeostasis associated with synapse loss and neurodegeneration observed in later stages of AD. Thus, α7 Rs may be important targets for attenuating Aβ-induced cognitive declines associated with AD. In contrast to the neurotoxic effects of high Aβ concentration in AD brains, recent studies suggest that picomolar (pM) Aβ concentrations present in healthy non-AD brains are required for normal learning and memory. Moreover, this is dependent on expression of α7 Rs, suggesting that pM Aβ exerts its beneficial influence on cognition through α7 R, possibly as a direct agonist or positive allosteric modulator (PAM) at α7 Rs. The nature of this interaction, however, has not yet been carefully characterized. The present study demonstrated that low Aβ concentrations likely facilitate α7 Rs through interaction as a PAM rather than as a direct agonist. Specifically, we found that 100 pM Aβ produced a ~ 1.7- to 2.6-fold facilitation of ACh-elicited α7 currents but did not elicit α7 currents directly. Moreover, when applied alongside the type II PAM PNU 120596, 100 pM Aβ occluded the facilitation of the peak current produced by PNU 120596, suggesting that Aβ and PNU 120596 facilitate α7 Rs through the same or overlapping allosteric site(s). This was further supported by analysis of a mutant α7 R construct containing a missense mutation in a critical amino acid for potentiation (M276L in TM 2) by PAMs. Peak current facilitation by 100 pM Aβ was completely abolished for the mutant α7 R construct. Surprisingly, this mutant also abolished the inhibitory effects of high (nM) Aβ concentrations. This suggests that inhibitory (nM to μM) and facilitatory (pM) Aβ concentrations exert their opposing effects on α7 R activity via the same or overlapping sites, rather than at distinct allosteric sites.

Disclosures: J. Farley: None. **J.B. Anderson:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.14

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

Support: 5R21AG064479

Title: Biased agonism of Amylin receptor and its therapeutic implications in Alzheimer's disease

Authors: *L. LABRADOR[1], R. R. CORRIGAN[2], G. CASADESUS SMITH[1];
[1]Pharmacol. and Therapeut., Univ. of Florida, Gainesville, FL; [2]The Steve and Cindy Rasmussen Inst. for Genomic Med., The Steve and Cindy Rasmussen Inst. for Genomic Med., Columbus, OH

Abstract: The pancreatic protein Amylin has biophysical properties similar to amyloid-beta (Aβ), the main pathological hallmark of Alzheimer's disease. Interestingly, while data support the potential pathogenic role of amylin in AD, others have shown that the administration of amylin or an amylin analog (pramlintide) improves function and pathology in murine models of AD. The amylin receptor is a widely expressed G-protein coupled receptor expressed in the cerebral vasculature, glia, and neurons, however, the study of amylin signaling is complicated by the fact that it signals through three different receptor subtypes which are also differentially expressed. Therefore, the goal of this work was to identify how these different receptor subtypes respond to both agonism and antagonism, as well as in the presence of binding at physiologically relevant levels (Aβ). Furthermore, given that amylin has been shown to reduce glucose uptake in muscle cells, we also sought to evaluate how these receptor subtypes regulated glucose uptake in normal and high glucose conditions (glucose resistance). We used SH-SY5Y cells as an in vitro model, with overexpression of AMY1 and AMY3, treated with 4 different concentrations of Amylin (PRAM between 10^{-5}-10^{-11}M), with AB (10^{-5}-10^{-11}M), and AC187 (10^{-5}-10^{-7}M). Cell viability was determined by LDH assay. By western blot, the differential activity of GSK3/AKT or PKA and ERK is detected. To observe a breakdown of the β-arrestin complex to the receptor, we performed co-immunoprecipitation with AKT. We also determined the level of glucose resistance by 2DG fluorescence in animals treated under the above conditions. Preliminary results support the differential effects of these receptor subtypes on the evaluated cascades. This will allow us to clarify amylin receptor signaling mechanisms as well as the current conflicting data on its involvement in AD pathogenesis or neuroprotection.

Disclosures: L. Labrador: None. **R.R. Corrigan:** None. **G. Casadesus Smith:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.15

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

Support: Rhode Island Foundation

Title: The impact of hypoxia on the nuclear pore and its link to neurodegeneration

Authors: *E. POTTS[1], C. FALLINI[2];
[1]Univ. of Rhode Island Interdisciplinary Neurosci. Program, Univ. of Rhode Island Interdisciplinary Neurosci. Program, Kingston, RI; [2]Univ. of Rhode Island, Univ. of Rhode Island, Kingston, RI

Abstract: Hypoxia is a life-threatening condition often associated with medical emergencies like stroke. Lack of oxygen and glucose during stroke leads to acute neuronal death and long-lasting damage to affected brain regions. Having a stroke will increase an individual's chances of developing a neurodegenerative disorder such as Alzheimer's Disease. When cells experience hypoxia, the actin cytoskeleton is impacted first, through aggregation of actin and cofilin, formation of actin stress fibers, and neurofilament destabilization. RNA-binding proteins (RBPs) then begin to accumulate in the cytoplasm, causing insoluble aggregates like those formed in neurodegenerative disorders. However, the molecular link between hypoxia-induced cellular changes and neurodegeneration is not well understood. Our previous research has established a link between actin homeostasis and the functional stability of the nuclear pore complex (NPC). Thus, we hypothesize that rearrangement of the actin cytoskeleton after hypoxia may have long-term impacts on the NPC, leading to the mislocalization and aggregation of RBPs in the cytoplasm and neuronal degeneration. To test this hypothesis, we exposed cortical neurons derived from iPSCs to acute oxygen and glucose deprivation (OGD) followed by a recovery period under normoxic conditions. We then examined the structural changes to the NPC and actin cytoskeleton, as well as subsequent functional changes occurring after hypoxia that may lead to widespread cellular dysfunction. Our results show that OGD induced long-lasting changes in NPC stability and function, leading to protein mislocalization. Collectively, our research will give novel insight into the cellular and molecular mechanisms of degeneration following hypoxia, potentially contributing to our understanding of the impact of environmental stress on neuronal physiology and survival.

Disclosures: **E. Potts:** None. **C. Fallini:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.16

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

Support: NIH AG061800
NIH AG054719
NIH DA039650
NIH AG063755
NIH AG068024

Title: Cross-platform synaptic network analysis of Alzheimer's disease identifies proteomic targets for functional validation

Authors: *J. HERSKOWITZ[1], C. WALKER[1], K. GREATHOUSE[1], J. TUSCHER[1], C. HURST[2], D. DUONG[3], J. DAY[1], N. SEYFRIED[2];
[1]Univ. of Alabama at Birmingham, Birmingham, AL; [2]Emory Univ., Atlanta, GA;
[3]Dduong@emory.Edu, Atlanta, GA

Abstract: Enormous resources have been spent on mass spectrometry (MS)-based studies utilizing postmortem human brain tissue samples to yield robust assessments of the aging and Alzheimer's disease (AD) proteomes. Yet, rarely do these large 'omics studies validate functional consequences of protein abundance changes. Here, we present a network that integrates dendritic spine density and morphology, phospho-tau measurements, and synaptosome proteomics from postmortem human entorhinal cortex samples. Module-trait correlations were used to guide selection of proteins for functional validation using CRISPR-dCas9 activation in experimental model systems. We show that TWF2, a module hub protein that correlates with thin dendritic spine length in humans, regulates thin spine length in rodent hippocampal neurons. Our network-validation framework also uncovers a putative role for PPP1R7 in the regulation of tau phosphorylation state at the synapse. Overall, we demonstrate that incorporating orthogonal cellular and molecular measurements into a proteomic network enables unbiased identification of proteins functionally involved in regulating those traits. Beyond elucidating synaptic processes in AD, this pipeline offers a blueprint to better contextualize omics-based targets with relevant biological mechanisms.

Disclosures: J. Herskowitz: None. **C. Walker:** None. **K. Greathouse:** None. **J. Tuscher:** None. **C. Hurst:** None. **D. Duong:** None. **J. Day:** None. **N. Seyfried:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.17

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

Support: NIA T32NS095775
NIA R01 AG061800
NIA R01 AG054719
NIA R01 AG063755

Title: Synaptic dysfunction caused by Alzheimer's disease patient brain-derived tau seeds

Authors: *A. WEBER, D. PUGH, C. WALKER, K. GREATHOUSE, J. HERSKOWITZ;
Univ. of Alabama at Birmingham, Birmingham, AL

Abstract: Amyloid-β (Aβ) plaques and neurofibrillary tangles (NFTs) of the microtubule-associated protein tau are the pathological hallmarks of Alzheimer's disease (AD). In AD patients, the extent of NFT spread throughout the brain correlates with the severity of cognitive impairment. NFT burden is inversely correlated with dendritic spine loss, suggesting that tau contributes to synapse loss in AD. Additionally, transgenic mouse models of tauopathy demonstrate that tau is associated with dysfunctional neuronal activation. In this study, we investigated dendritic spine density and morphology as well as synaptic function in an *in vitro* model of tau accumulation. Human AD brain-derived tau paired helical filaments (AD-tau) were used to seed aggregation of endogenous rat tau in primary neuronal cultures. AD-tau was purified using differential centrifugation of sarkosyl-insoluble fractions. To begin understanding synaptic alterations, primary cortical rat neurons were seeded with AD-tau on multielectrode arrays (MEA) to measure local field potentials generated by spontaneous firing of neurons. MEA recordings were evaluated over a 23-day time course which assessed longitudinal changes in mean neuronal firing rate. To evaluate dendritic spine alterations, individual dendrites were imaged using wide-field microscopy. Neurolucida 360 was employed for three-dimensional dendritic reconstructions and spine morphometric analysis. Our findings provide evidence that AD-tau induces endogenous tau accumulation, causing robust synaptic dysfunction and alterations in spine density and morphology.

Disclosures: A. Weber: None. **D. Pugh:** None. **C. Walker:** None. **K. Greathouse:** None. **J. Herskowitz:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.18

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

Support: U01AG061357
R01AG061800

Title: Integrated Proteomics Identifies Neuritin (NRN1) as a Mediator of Cognitive Resilience to Alzheimer's Disease

Authors: *C. HURST[1], D. A. PUGH[3], M. H. ABREHA[1], D. M. DUONG[4], E. B. DAMMER[1], D. A. BENNETT[5], J. H. HERSKOWITZ[6], N. T. SEYFRIED[2];
[2]Human Genet., [1]Emory Univ., Atlanta, GA; [3]Univ. of Alabama Birmingham, Birmingham, AL; [4]Dduong@emory.Edu, Atlanta, GA; [5]Rush Alzheimer's Dis. Ctr., Rush Univ. Med. Ctr., Chicago, IL; [6]Neurol., Univ. of Alabama at Birmingham, Birmingham, AL

Abstract: The molecular pathways enabling certain individuals to remain cognitively normal despite high levels of Alzheimer's disease (AD) pathology remain incompletely understood. To address these gaps, we implemented an integrative systems-level analysis of multi-region

postmortem human brain proteomics derived from the Religious Order Study and Rush Memory and Aging Projects (ROSMAP; n=218 total samples) to identify proteins and pathways significantly altered in resilient cases. Multiplex tandem mass tag mass spectrometry (TMT-MS)-based proteomic data (n=7,787 proteins) was implemented for a correlation network analysis and identified protein communities (modules) significantly correlated to AD neuropathology and cognitive capacity. Data from an independent brain proteome wide association study (PWAS; n=>8,000 proteins) of cognitive trajectory was then integrated with the brain network to robustly and unbiasedly nominate modules associated with cognitive resilience. This revealed proteins linked to synaptic biology and cellular energetics. Neuritin (NRN1), a neurotrophic factor previously characterized for roles in synaptic maturation and stability, was prioritized as a hub that co-expressed with a community of proteins highly correlated to cognitive stability in life. To further validate our systems-level analysis and characterize NRN1 neurobiological mechanisms, rat primary neurons were treated with recombinant NRN1 protein followed by TMT-MS (n=8,238 proteins). Differential expression of neurons treated with NRN1 demonstrated a strong bias in increased expression of proteins associated with broad synaptic functions (445 proteins $p<0.05$). In addition, pathways significantly decreased following NRN1 treatment were related to metabolism and cellular energetics (400 proteins $p<0.05$), systems often dysregulated and increased in AD, implicating NRN1 as a dual-action molecular effector able to increase proteins typically vulnerable to or lost in AD and to decrease proteins aberrantly increased in disease. Furthermore, re-integration of NRN1-driven differential expression into the human brain network revealed significant overlap with human modules of resilience. These findings indicate that NRN1 target engagement is highly relevant to human resilience mechanisms identified by unbiased systems-level analyses and further support NRN1 as a potentially valuable effector of resilience in AD.

Disclosures: C. Hurst: None. **D.A. Pugh:** None. **M.H. Abreha:** None. **D.M. Duong:** None. **E.B. Dammer:** None. **D.A. Bennett:** None. **J.H. Herskowitz:** None. **N.T. Seyfried:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.19

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

Support: NIA AG061800
NIA AG054719
NIA AG063755
NIA AG068024

Title: Dendritic spines provide cognitive resilience against Alzheimer’s disease pathology in the entorhinal cortex

Authors: *K. M. GREATHOUSE, C. K. WALKER, B. D. BOROS, K. A. CURTIS, J. H. HERSKOWITZ;
UAB, Birmingham, AL

Abstract: Amyloid-β (Aβ) plaques and neurofibrillary tangles (NFTs) of the microtubule-associated protein tau are the pathological hallmarks of Alzheimer's disease (AD); however, synapse or dendritic spine loss correlates more strongly with cognitive impairment than Aβ plaques or NFTs. Approximately one-third of individuals that come to autopsy in their eighties have Aβ plaques and tau NFTs, yet did not experience dementia in life. These cognitively normal individuals with AD pathology were likely in preclinical stages of AD. Unlike individuals with AD dementia, CAD cases do not exhibit dendritic spine loss in the prefrontal cortex. In this study, we asked whether the entorhinal cortex (EC), one of the earliest regions to exhibit tau pathology, shows alterations in dendritic spine density and morphology in AD. Postmortem human Brodmann Area (BA) 28 EC samples from 20 cognitively normal pathology-free controls (control), 8 cognitively normal individuals with AD pathology (CAD), and 24 symptomatic AD cases were obtained from the Emory University Alzheimer's Disease Research Center Brain Bank. AD cases exhibited a significant reduction in dendritic spine density in the EC, compared to controls and CAD cases. Controls and CAD cases exhibited comparable spine density in the EC, supporting the hypothesis that maintenance and preservation of dendritic spines is critical for retaining cognitive function in the presence of AD pathology. Next, spine density was assessed by determining whether loss of spines within certain morphological subclasses occurred in the EC. We observed a loss of thin, stubby, and mushroom spines in AD cases compared to controls, indicating a global reduction of spines, rather than loss of a specific spine type. Finally, we examined dendritic spine length and head diameter. No differences in dendritic spine length or head diameter were detected among control, CAD, and AD cases. Similar findings on length and head diameter were observed for each spine subclass. Dendritic spine density and morphology measurements were then correlated with case demographic and pathology data. We observed a significant inverse correlation of spine density with age, which was also observed for thin and stubby spine subclasses. Additionally, density of thin spines exhibited a significant positive correlation with MMSE scores. Sex was not associated with any dendritic spine measurements, while NFT burden, reported as Braak stage, displayed a significant inverse correlation with spine density.

Disclosures: K.M. Greathouse: None. **C.K. Walker:** None. **B.D. Boros:** None. **K.A. Curtis:** None. **J.H. Herskowitz:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.20

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

Support: NINDS T32 NS061788
NIA AG061800
NIA AG054719
NIA AG063755

Title: Neuritin (NRN1) Provides Dendritic Spine Resilience against Amyloid-β in Alzheimer's Disease

Authors: *D. PUGH[1], C. HURST[2], D. DUONG[2], D. A. BENNETT[3], N. SEYFRIED[2], J. HERSKOWITZ[1];
[1]Neurol., Univ. of Alabama at Birmingham, Birmingham, AL; [2]Emory Univ., Atlanta, GA; [3]Rush Alzheimer's Dis. Ctr., Rush Univ. Med. Ctr., Chicago, IL

Abstract: Alzheimer's disease (AD) is an irreversible neurodegenerative disease defined by its pathological hallmarks, amyloid-β (Aβ) and tau neurofibrillary tangles (NFTs). There is increasing recognition of a subpopulation of "cognitively resilient" individuals that live to advanced age with intact cognitive function despite the presence of plaque and tangle pathology. Dendritic spine plasticity is hypothesized to be a mechanism of cognitive resilience that protects older individuals with moderate to severe AD neuropathology from developing dementia. It is therefore important to identify genes and proteins that mediate cognitive resilience in order to understand the relationship between dendritic spine loss and cognitive impairment in AD. Towards this goal, Neuritin (NRN1) was recently identified as the most significant protein associated with cognitive resilience in a proteome-wide association study conducted in the Religious Orders Study and the Rush Memory and Aging Project longitudinal cohorts (ROSMAP, n = 391), in which higher levels of cortical NRN1 at autopsy correlated with a slower rate of cognitive decline in life. To validate and explore the potential neuroprotective mechanisms of NRN1 against AD neuropathology, we treated rat primary neuronal cultures with recombinant NRN1 protein and/or toxic Amyloid-beta 1-42 ($A\beta_{42}$) oligomers. Dendrites were imaged using wide-field microscopy. Neurolucida 360 was employed for three-dimensional dendrite reconstruction and dendritic spine morphometric analysis. Dendritic protrusions were classified as dendritic filopodium, thin, stubby, or mushroom spines. Spine density was calculated as the number of spines per 10μm of dendrite length. In parallel, multi-electrode array (MEA) analyses of rat primary neuronal cultures were performed. Single neuron electrophysiological activity was recorded and mean action potential frequency (Hz) was calculated as the ratio of the total number of spikes recorded and the duration of recording in seconds. Morphometric analysis revealed treatment of neuronal cultures with recombinant NRN1 rescued $A\beta_{42}$-induced hippocampal dendritic spine loss and morphologic aberrations. MEA analyses revealed that $A\beta_{42}$ increased mean action potential frequency, however neuronal hyper-excitability was not observed after simultaneous exposure to $A\beta_{42}$ and NRN1. Our findings provide evidence that NRN1 can protect against $A\beta_{42}$-induced dendritic spine degeneration and neuronal hyper-excitability in cultured neurons. We hypothesize that therapeutic targeting of NRN1 may provide dendritic spine resilience to $A\beta_{42}$ and that may promote maintenance of cognition in old age.

Disclosures: D. Pugh: None. **C. Hurst:** None. **D. Duong:** None. **D.A. Bennett:** None. **N. Seyfried:** None. **J. Herskowitz:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.21

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

Support: NIH/NIA 1R01AG072883

Title: Proximity Ligation Assay - Flow Cytometry: a method to study synaptic protein-protein interactions of isolated human synaptosomes

Authors: *M. MARCATTI, D. JAMISON, G. TAGLIALATELA;
Univ. of Texas Med. Br., Galveston, TX

Abstract: Synaptic dysfunction often underscored by altered interaction of key synaptic protein, plays an important role in the pathophysiology of many neurological disorders, including Alzheimer's disease (AD), Parkinson's disease (PD), and schizophrenia. Because of the limitations associated with employing fresh brain tissue samples from human subjects, there is a need to develop methods to study human synapses isolated from frozen tissue. From cryopreserved human brain tissues, it is possible to isolate synaptosomes which are detached-and-resealed synaptic terminals metabolically and enzymatically active. Synaptosomes have become important model systems for studying human synaptic functions, since they are more accessible than ex vivo brain slices or primary neuronal cultures. Many technical approaches have been used to study synaptosomes such as immunostaining, flow cytometry, and immunoprecipitation. Here for the first time, we apply flow cytometry coupled to Proximity Ligation assay (PLA) to synaptosomes to study protein-protein interaction at the synapses. PLA technology detects the interaction between proteins as well as post-translational modifications within the same protein. Two primary antibodies raised in different species are labelled with oligonucleotides which form a circular DNA when the two proteins of interest are present within 40 nm of distance. The amplification of this DNA generates a specific fluorescent signal after the addition of labeled probes, which can be visualized by flow cytometry. This technique has many advantages over immunoprecipitation since it requires less starting material and primary antibody and can be performed in few hours. Most important, PLA reduce general problem of cross-reactivity during IP, by ensuring that only interacting proteins result in a signal. To demonstrate the feasibility of PLA on synaptosomes we investigated the known synaptic interaction between PSD95 and NMDARs (NMDAR2a and NMDAR2B). We first established optimal conditions for synaptosome fixation and permeabilization, as well as concentration for each primary antibody by performing flow cytometry staining on human synaptosomes from frontal cortex (FC), hippocampus (HP), and cerebellum (CB). Then, we used commercially available PLA kits (Millipore-Sigma and Navinci) to perform PLA on HP and FC synaptosomes to evaluate PSD95-NMDAR2A PSD95-NMDAR2B interactions, respectively. Our data encourage the possibility to use PLA as an important tool to identify synaptic interactors that can be useful for the study of AD and other neurodegenerative diseases characterized by synaptic dysfunction

Disclosures: **M. Marcatti:** None. **D. Jamison:** None. **G. Taglialatela:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.22

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

Support: NIH/NIA R01AG072883
UTMB Presidential Scholars Program

Title: Elucidating the Protein Substrates for Amyloid Beta and Tau Oligomer Interactions in Alzheimer's Disease using Proximity Ligation Assays

Authors: *D. JAMISON[1], M. MARCATTI[2], G. TAGLIALATELA[2];
[1]Pharmacol. and Toxicology, [2]Neurol., Univ. of Texas Med. Br., Galveston, TX

Abstract: Elucidating the Protein Substrates for Amyloid Beta and Tau Oligomer Interactions in Alzheimer's Disease using Proximity Ligation Assays**Authors**D. JAMISON[1], M. MARCATTI[2], G. TAGLIALATELA[2]; [1]Pharmacology and Toxicology, University of Texas Medical Branch, Galveston, TX, [2]Neurology, University of Texas Medical Branch, Galveston, TX

AbstractAlzheimer's Disease (AD) is the most prevalent form of neurodegeneration and represents a health crisis for the United States. In 2020, nearly six million Americans were afflicted with AD, and the number will continue to increase as the aging population grows. AD's pathological hallmarks include amyloid beta plaques and neurofibrillary tangles. Recently, the field has recognized the role amyloid beta (AβO) and tau (tauO) oligomers play in the progression of the disease, concluding that these oligomers are the true toxic species. Previously, we have shown that tauO can outcompete AβO from human synaptosomes in a concentration-dependent manner, but this phenomenon was lost in synaptosomes devoid of superficial protein. Therefore, we hypothesized that the ability of tauO to outcompete AβO from human synapses occurs on a protein substrate. Determining what proteins are involved in this interplay could provide pharmacological targets and the potential to develop new therapeutic approaches for AD. Here, we used proximity ligation assays (PLAs) combined with flow cytometry to investigate this phenomenon on synaptosomes isolated from control human hippocampus challenged with exogenous AβO and tauO. Results from the flow cytometric PLAs demonstrated that exogenous AβO at 2.5 μM and 5 μM concentrations interact with both NMDAR 2A and 2B receptors, with more interaction observed at the 2A subunit. Here, we observed, consistent with our previous data, a decrease in AβO synaptic binding in presence of tauO. Altogether, this would suggest that NMDAR 2A and 2B are shared protein substrates for both tauO and AβO with more affinity for tauO when their levels are high, such as during the late stage of AD. Pharmacological blocking of these substrates could represent a novel treatment to lessen or prevent the deleterious effects oligomers have on the synapse, thus slowing the progression of AD.

Disclosures: **D. Jamison:** None. **M. Marcatti:** None. **G. Taglialatela:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.23

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

Support: NIH Grant NS041783
NIH Grant AG063520

Title: Presenilins regulate synaptic plasticity in the lateral and medial perforant pathways in the hippocampus

Authors: *S. LEE[1], V. Y. BOLSHAKOV[2], J. SHEN[1];
[1]Neurol., Brigham and Women's Hosp. / Harvard Med. Sch., Boston, MA; [2]Psychiatry, McLean Hosp. / Harvard Med. Sch., Belmont, MA

Abstract: Mutations in the *Presenilin* (*PSEN*) genes are the major cause of familial Alzheimer's disease (AD), highlighting the importance of Presenilin (PS) in AD pathogenesis. Previous studies of PS function in the hippocampus demonstrated that loss of PS results in the impairment of neurotransmitter release, short- and long-term synaptic plasticity at hippocampal mossy fiber (MF) synapses as well as Schaffer collateral (SC) synapses. Cortical input to the hippocampus through the lateral perforant pathway (LPP) and the medial perforant pathway (MPP) is critical for normal cognitive functions and is particularly vulnerable during aging and early stages of AD. Whether PS regulates synaptic function in the perforant pathways remained unknown. In the current study, we investigate PS function in LPP and MPP by performing whole-cell and field-potential electrophysiological recordings using acute hippocampal slices from postnatal forebrain-restricted excitatory neuron-specific *Presenilin* conditional double knockout (*PS* cDKO) mice at 2 months of age. We found that paired-pulse ratio (PPR) and synaptic responses induced by tetanic stimulations are impaired in both LPP and MPP of *PS* cDKO mice. Moreover, the efficacy of excitatory neurotransmission as assessed with synaptic input/output relations for evoked excitatory postsynaptic currents (EPSCs) is markedly lowered in LPP and MPP of *PS* cDKO mice, while both the frequency and the amplitude of spontaneous EPSCs (sEPSCs) are similar between *PS* cDKO and control neurons. Furthermore, we found that long-term potentiation (LTP) is impaired in LPP and MPP of *PS* cDKO mice. Consistent with the LTP deficit, *PS* cDKO mice display reduced evoked NMDA receptor-mediated responses. Interestingly, depletion of intracellular Ca^{2+} stores by inhibition of sarcoendoplasmic reticulum Ca^{2+} ATPase results in a reduction of EPSC amplitude in control hippocampal neurons but not in *PS* cDKO neurons, suggesting that impaired intracellular calcium homeostasis in the absence of PS may contribute to the deficits in synaptic transmission. These findings show the importance of PS in the regulation of synaptic plasticity and intracellular calcium homeostasis in the hippocampal perforant pathways.

Disclosures: **S. Lee:** None. **V.Y. Bolshakov:** None. **J. Shen:** None.

Poster

530. Synaptic Dysfunction in Alzheimer's Disease: *In Vitro* Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 530.24

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

Support: Alzheimer's Association (MI)
NIH Grant AG051179 (MI)
NIH Grant AG070868 (MI)
NIH Grant NS37853 (CI)

Title: Amyloid beta 42 causes intracellular calcium dysregulation and arcuate neuropeptide Y (NPY) neuronal dysfunction by modulating $Ca_V1.3$ channels

Authors: *G. WANG, W. XIE, D. LEE, C. IADECOLA, M. ISHII;
Weill Cornell Med., New York, NY

Abstract: Accumulating evidence suggests that systemic metabolic deficits attributable to hypothalamic dysfunction can occur before the cognitive decline in Alzheimer's disease (AD) (*Cell Metab,* 2015, 22, 761). We previously found in young 3 months old (3M) Tg2576 mice overexpressing the Swedish mutation of the amyloid precursor protein that Aβ pathology can cause dysfunction in hypothalamic arcuate (ARC) neuropeptide Y/agouti-related peptide (NPY/AgRP)-expressing neurons, which was associated with early systemic metabolic deficits prior to memory deficits and plaque formation (*J Neurosci,* 2014, 34, 9096). Furthermore, the ARC NPY dysfunction was caused by dysregulation of voltage-gated L-type Ca^{2+} currents (LTCC) by Aβ (*J Neurosci,* 2019, 39, 8816). However, the mechanisms by which $Aβ_{42}$ leads to dysregulation of LTCC or disruption of intracellular Ca^{2+} homeostasis remain elusive. Here we examined which of the two major isoforms of LTCC in the brain, $Ca_V1.2$ or $Ca_V1.3$, is targeted by $Aβ_{42}$ in ARC NPY neurons. We used the Cre-lox system to selectively knock-out $Ca_V1.2$ or $Ca_V1.3$ in ARC NPY/AgRP neurons ($Ca_V1.2$ cKO, $Ca_V1.3$ cKO) in wild-type (WT) mice. All mice were crossed to NPY-GFP reporter mice. LTCC in ARC NPY neurons from 3M mice were recorded by whole-cell voltage-clamp. Cytoplasmic Ca^{2+} levels $[Ca^{2+}]_i$ were measured ratiometrically by loading Fura-2 AM in dissociated ARC NPY neurons from 3M mice. First, we tested the effects of $Aβ_{42}$ in ARC NPY neurons from the Cre-negative ($Cre^{-/-}$) WT mice as controls. Similar to our previous data, we found that in the presence of 100 nM $Aβ_{42}$ the peak current of LTCC I-V curves in $Ca_V1.3$ $Cre^{-/-}$ ARC NPY neurons were shifted from a high voltage-threshold in the control to a low voltage-threshold, an effect reversed by the LTCC blocker nimodipine (NMD, 2 μM) (SP=-30 mV: Veh -172.4±114.9 pA, $Aβ_{42}$ -615.7±138.8 pA, NMD -153± 196.5 pA, n=4-5, $p<0.05$). $Aβ_{42}$ (100 nM) also increased $[Ca^{2+}]_i$ in $Ca_V1.3$ $Cre^{-/-}$ ARC NPY neurons (Veh 0.145± 0.018, $Aβ_{42}$ 0.162±0.019, n=16, $p<0.01$). In ARC NPY neurons from $Ca_V1.2$ cKO mice, $Aβ_{42}$ had similar effects on the I-V curves (SP=-30 mV: Veh -165±51.7

pA, $A\beta_{42}$ -300.8± 56.4 pA, NMD -72.3±34.9 pA, n=6, $p<0.05$) and $[Ca^{2+}]_i$ (Veh 0.109±0.01, $A\beta_{42}$ 0.134± 0.01, n=26, $p<0.01$). Interestingly, in ARC NPY neurons from $Ca_v1.3$ cKO mice, $A\beta_{42}$ (100 nM) had no effect on the I-V curves (SP=-30 mV: Veh -221.9±73.7 pA, $A\beta_{42}$ -236.9 ±33.8 pA, n=4, $p>0.05$) or $[Ca^{2+}]_i$ (Veh 0.18±0.017, $A\beta_{42}$ 0.178±0.016, n=21, $p>0.05$). These data support that $Ca_v1.3$ but not $Ca_v1.2$ is required for the pathological effects of $A\beta_{42}$ on ARC NPY neurons. Future studies will be important in further elucidating the pathological role and therapeutic potential of modulating $Ca_v1.3$ in AD.

Disclosures: G. Wang: None. **W. Xie:** None. **D. Lee:** None. **C. Iadecola:** None. **M. Ishii:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.01

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

Support: NIH (NINDS): 2R01NS045860-16A1 (RET)
Cure Alzheimer's Fund: 2021A010138 (RB)

Title: Mitochondria-associated ER Membranes (MAMs) promote axonal amyloid b (Ab) generation in a cellular model of Alzheimer's disease (AD)

Authors: *R. BHATTACHARYYA[1], M. B. TARANTINO[2], M. S. LOTLIKAR[2], M. JORFI[2], J. C. ZELLMER[2], D. M. KOVACS[3], R. E. TANZI[4];
[1]Genet. and Aging Res. Unit, Dept. of Neurol., Massachusetts Gen. Hosp. and Harvard Med. Sch., Charlestown (Boston), MA; [2]Genet. and Aging Res. Unit, Dept. of Neurol., Massachusetts Gen. Hosp., Charlestown, MA; [3]Neurobio. of Dis. Lab. / Genet. and Aging Res. Unit / MIND / Neurol., Massachusetts Gen. Hosp. / Harvard Med. Sch., Charlestown, MA; [4]Massachusetts Gen Hosp, Harvard Med. Sch., Charlestown, MA

Abstract: Aggregation of amyloid β (Aβ) begins with the processing of APP by β- and γ-secretases in Alzheimer's disease (AD) (*amyloid hypothesis*). Several reports have shown that the crosstalk between mitochondria and the ER at the mitochondria-ER contact sites or MAMs (Mitochondria-associated ER Membranes) are altered in Alzheimer's disease and AD-related models supporting the hypothesis that MAMs play a role in AD pathogenesis (*MAM hypothesis*). MAMs are cholesterol-rich lipid raft-like microdomains in the ER where APP, BACE1, and components of γ-secretase are found [Schon, E.A. and E. Area-Gomez (2013)]. BACE1 (β-secretase)-cleaved C-terminal fragments of APP (β-CTF) is also found in the MAMs suggesting MAMs as a critical microdomains for Aβ-generation. Earlier, we showed that APP undergoes palmitoylation, and palmitoylated APP (*pal*APP) is recruited to lipid rafts resulting in increased BACE1-mediated β-cleavage and Aβ generation *in vitro* [Bhattacharyya, R. (2013, 2016)]. We have now used a newly developed cellular model of AD (human neural progenitor stem cells

overexpressing APP containing FAD mutations; FAD-hNPCs) to demonstrated that the majority (>70%) of *pal*APP resides in MAMs, primarily in axons and in neuronal processes [Bhattacharyya, R. et al. (2021)]. Here, we report that activation of the MAM-resident sigma 1 receptor (S1R) by its agonist (PRE-084) or inactivation by its antagonist (NE-100) exclusively altered MAMs in the axons, without affecting somal MAMs. Most importantly, employing a novel microfluidic device that can separate somal and axonal environments [Lotlikar, M.S. et al. (2021)], we have found that modulation of axonal MAMs via S1R altered axonal Aβ generation [Bhattacharyya, R. et al. (2021)]. It is not clear how axonal Aβ contributes to Aβ deposition surrounding dystrophic neurons in later stages of the disease, but nearly 40% of neuronal Aβ is generated in neuronal processes and axons. We found that S1R mediated axonal MAM-modulation regulated the stability of newly synthesized *pal*APP and promoted its cell-surface association [Bhattacharyya, R. et al. (2021)], thus supporting a novel hypothesis that axonal MAMs promote cell surface trafficking of MAM-stabilized newly synthesized palmitoylated APP for Aβ generation in AD.

Disclosures: R. Bhattacharyya: None. **M.B. Tarantino:** None. **M.S. Lotlikar:** None. **M. Jorfi:** None. **J.C. Zellmer:** None. **D.M. Kovacs:** None. **R.E. Tanzi:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.02

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

Support: UK Dementia Research Institute
Alzheimer's Association Grant
BrightFocus Foundation Grant A2019112S
UKRI Future Leaders Fellowship MR/S017003/1

Title: A novel role for APP in neural circuit function

Authors: *S. HARRIS[1], R. RAJANI[1], J. ZUENKLER[1], R. A. ELLINGFORD[2], M. YANG[2], M. KEHRING[2], B. LEE[2], B. HYMAN[3], U. MULLER[4], M. BUSCHE[2];
[2]UK Dementia Res. Inst., [1]Univ. Col. London, London, United Kingdom; [3]Massachusetts Gen. Hosp., Charlestown, MA; [4]Heidelberg Univ., Heidelberg, Germany

Abstract: Recent evidence has implicated amyloid precursor protein (APP)-derived peptides, including amyloid-beta, in abnormal brain activation that precedes cognitive impairment in Alzheimer's Disease (AD). However, despite the importance of the APP protein family to AD pathophysiology and therapy, how these contribute to normal brain circuit function remains unclear. Here, we sought to interrogate how the APP family regulates neuronal cell function and long-range neural circuit dynamics.$APP^{flox/flox}/APLP2^{flox/flox}/APLP1^{-/-}$ mice were crossed with NexCre mice expressing Cre prenatally from ~E12.5 in postmitotic neuronal precursor cells of

the cortex and hippocampus, producing a conditional triple knockout model (cTKO) of the APP family in excitatory forebrain neurons that is associated with pronounced behavioral deficits (Steubler et al., 2021, *EMBO Journal*). Neuronal function at the single cell/unit and circuit-level was characterised in all mouse lines using in-vivo two-photon and mesoscopic calcium imaging, and bilateral Neuropixel recordings in awake and anesthetised animals. Cortical neuronal activity in cTKO mice was profoundly suppressed relative to controls, with a marked loss of cross-hemispheric coherence of oscillatory activity (including sleep-dependent slow waves). cTKO animals also displayed behavioural abnormalities in open field tests relative to controls. These cellular and circuit-level impairments and behavioral deficits were tightly correlated and were partially rescued by NMDA receptor agonism. These findings indicate that the APP family plays a critical, but previously unknown, role in modulating neuronal excitability and long-range circuit connectivity. Our research provides novel mechanistic insights into the physiological function of the APP family and their potential role in AD pathogenesis, with important implications for current and future AD therapeutic approaches.

Disclosures: **S. Harris:** None. **R. Rajani:** None. **J. Zuenkler:** None. **R.A. Ellingford:** None. **M. Yang:** None. **M. Kehring:** None. **B. Lee:** None. **B. Hyman:** None. **U. Muller:** None. **M. Busche:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.03

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

Support: NIH, NS84324
NMRC, NMRC/StaR/009/2012
OF-YIRG, MOH-000237-00

Title: Evidence for a clathrin-independent endocytic pathway for APP internalization in the neuronal somatodendritic compartment

Authors: *J. AOW[1,2], T.-R. HUANG[3], G. THINAKARAN[4], E. H. KOO[5,2];
[1]Genome Inst. of Singapore, Singapore, Singapore; [2]Dept. of Med., [3]Grad. Sch. for Integrative Sci. and Engin., Natl. Univ. of Singapore, Singapore, Singapore; [4]Byrd Alzheimer's Inst., Univ. of South Florida, Tampa, FL; [5]Dept. of Neurosciences, UCSD, La Jolla, CA

Abstract: APP internalization via clathrin-/dynamin-mediated endocytosis (CME) mediated by its YENPTY motif into β-secretase-containing endosomes is proposed to be critical for amyloid-beta (Aβ) production. Aβ production and secretion is activity-dependent, but the underlying mechanisms remain poorly understood. Here we show that somatodendritic APP internalization in primary rodent neurons is not blocked by inhibiting dynamin or mutating the YENPTY motif, in contrast to human and rodent non-neuronal cell lines. These phenomena in neurons were

observed under basal conditions and during chemical long-term depression stimulus. However, both dynamin inhibition and the YENPTY mutant significantly decreased secreted Aβ in primary neuronal culture. Thus, somatodendritic APP internalization and downstream Aβ secretion can be decoupled, and internalized somatodendritic APP may not be a major source of secreted Aβ. Indeed, internalized somatodendritic APP puncta overlapped poorly with APP/β-secretase proximity-ligation-assay puncta. Interestingly, somatodendritic low-density-lipoprotein receptor (LDLR) internalization does not require its CME motif, suggesting that this novel sorting behavior is not unique to APP. These results point to intriguing differences in endocytic mechanisms that exist in different neuronal compartments and refine our understanding of Aβ production and secretion in neurons.

Disclosures: J. Aow: None. **T. Huang:** None. **G. Thinakaran:** None. **E.H. Koo:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.04

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

Title: The tripeptide arginine-glutamate-arginine (RER), an active region of secreted amyloid precursor protein-alpha, upregulates LTP in a CamKII- and CRMP-dependent manner

Authors: *K. D. PARFITT[1], E. P. ROSE[4], K. C. LANKER[1], C. I. CASPER[2], J. M. WARD[2], A. H. SHERIDAN[2], D. J. O'LEARY[3];
[1]Neurosci., Pomona Col., Claremont, CA; [2]Neurosci., [3]Chem., Pomona Col., CLAREMONT, CA; [4]Neurosci., Oregon Hlth. and Sci. Univ., Portland, OR

Abstract: Secreted APPα (sAPPα), a 612-residue protein derived from amyloid precursor protein, has neurotrophic and neuroprotective properties and enhances hippocampal long term potentiation (LTP) in vivo and in vitro. In addition, overexpression of sAPPα can prevent deficits in spatial memory seen in the APP/PS1 mouse model of Alzheimer's Disease. A tripeptide sequence —arginine-glutamate-arginine (RER)—within the central APP domain (E2) of sAPPα has been proposed as an active region of the protein that enhances memory in a passive-avoidance task in chicks. In previous work (Morrissey et al., 2019), we have observed that congeners of sAPPα's RER motif, in an acylated and diastereomeric form (Ac-rER), can mimic the LTP-enhancing effects of full-length sAPPα in rat or mouse hippocampus. In particular, Ac-rER can produce LTP induced by an otherwise sub-threshold theta burst stimulation (TBS). Hippocampal slices were prepared from adult male C57Bl/6N mice and allowed to recover for at least 2 h. Baseline EPSPs were evoked in area CA1 by electrical stimulation of the Schaffer collateral pathway, and recorded in stratum radiatum. Application of a sub-threshold theta-burst stimulation (TBS, 5 trains (5 Hz) of 5 pulses (100 Hz)) induced post-tetanic potentiation (PTP), but not LTP, in untreated slices. In contrast, when applied 25 min before, during and following the mild TBS, Ac-rER (10 nM) facilitated both PTP and LTP compared to controls. The ability

of sAPPα to enhance LTP by this mild TBS protocol depends on CaMKII activation. Similarly, the Ac-rER-mediated LTP was blocked by the CaMKII inhibitor KN62 (10 uM). Neither KN62 nor Ac-rER affect paired-pulse facilitation, suggesting that Ac-rER is facilitating LTP through CaMKII in a post-synaptic manner. A potential substrate of CaMKII in hippocampal neurons, in addition to glutamate receptors, is the collapsin response mediator protein-2 (CRMP2), which has been shown to be required for the memory-enhancing actions of Ac-rER in chicks. An inhibitor of CRMP2, lacosamide, does not inhibit conventional LTP induced by a strong TBS; however, the co-application of lacosamide and Ac-rER results in significant reduction of Ac-rER's LTP-inducing effects. Taken together, and considering previous work, these data suggest that Ac-rER enhances synaptic plasticity by activating CaMKII, CRMP2, and protein synthesis, and by promoting intracellular trafficking of critical proteins to the hippocampal pyramidal cell surface. Given the similarity of these mechanisms to those of sAPPα, and the ability of Ac-rER to enhance memory in other vertebrates, this tripeptide may have notable therapeutic benefits.

Disclosures: K.D. Parfitt: None. **E.P. Rose:** None. **K.C. Lanker:** None. **C.I. Casper:** None. **J.M. Ward:** None. **A.H. Sheridan:** None. **D.J. O'Leary:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.05

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

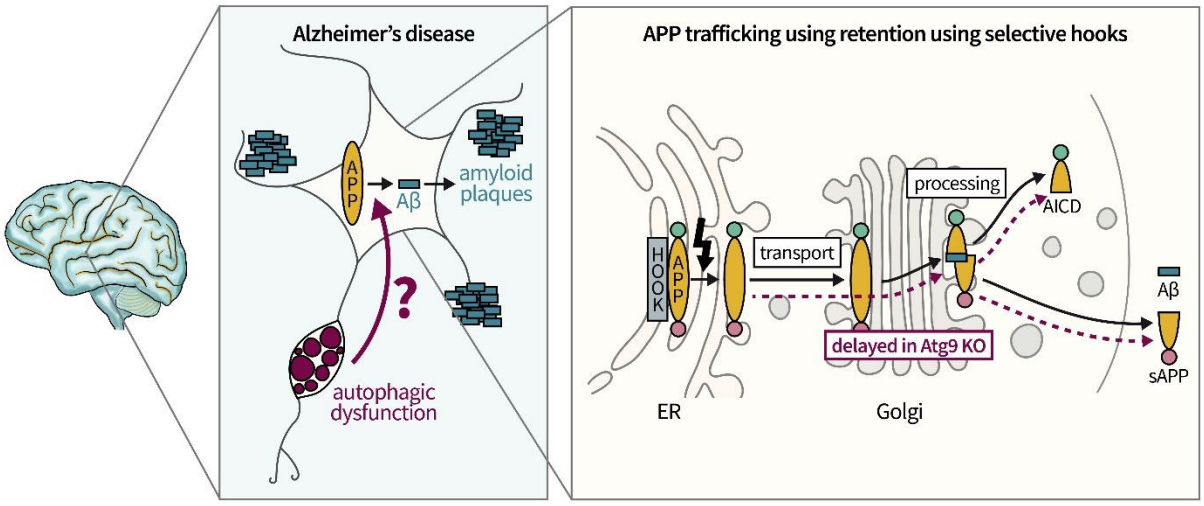
Support: H2020-MSCA-ITN-2019 SAND 860035
Gun och Bertil Stohnes Stiftelse

Title: ATG9-deficiency delays amyloid precursor protein transport and processing associated with Alzheimer's disease

Authors: *J. MAYER[1], D. BOECK[1], M. WERNER[1], M. SHIMOZAWA[1], H. FARHAN[2], P. NILSSON[1];
[1]Karolinska Institutet, Solna, Sweden; [2]Innsbruck Med. Univ., Innsbruck, Austria

Abstract: Alzheimer's disease (AD) pathology is characterized by amyloid plaques and dysfunctional autophagy. In particular, the core autophagy protein ATG9 was increased in the brains of *amyloid precursor protein* (*App*) knock-in AD mice. The amyloid plaques consist of amyloid beta-peptides, which are generated by the sequential proteolytic cleavage of APP. The site of intracellular APP processing is highly debated and might include autophagosomes. We aim to investigate the autophagy involvement in intracellular APP transport and cleavage, by applying the "retention using selective hooks" (RUSH) system. RUSH allows studying the transport of fluorescently labeled APP in a systematic way. HeLa cells expressing the RUSH-APP system were investigated by live-cell imaging and Western blot. Under baseline conditions, full-length APP is retained in the endoplasmic reticulum (ER). After releasing APP from the

hook, APP starts accumulating in the Golgi within 20 min. Autophagy-deficient ATG9 KO cells showed a significant delay in ER-to-Golgi transport of APP and more APP remained in the ER and Golgi over time. Our study highlights that dysfunctional autophagy is interfering with physiological APP transport potentially leading to the intracellular accumulation of cytotoxic amyloid-beta and contributing to Alzheimer's disease.



Disclosures: **J. Mayer:** None. **D. Boeck:** None. **M. Werner:** None. **M. Shimozawa:** None. **H. Farhan:** None. **P. Nilsson:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.06

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

Title: Intertwining between neuroblastoma cells differentiation and APP over-expression and processing

Authors: A. NIGRO[1], Z. LOMBARDI[1], C. CARBONE[1], ***M. CALAMAI**[2,1];
[1]European Lab. for non-linear spectroscopy, Sesto Fiorentino (Firenze), Italy; [2]CNR-INO, CNR-INO, Sesto Fiorentino, Italy

Abstract: While the expression of APP is up-regulated during neuronal differentiation and growth, little is still known on the effect of neuronal differentiation on APP processing. A common strategy to induce neuronal differentiation is to treat precursor cells with retinoic acid.

Experimental evidence has shown that many genes associated with Alzheimer's disease are affected by the action of retinoic acid. Some studies have demonstrated that retinoic acid increases the expression and activity of ADAM 10 in cell models. On the other hand, in mouse models that express APP, it has been found that retinoic acid can inhibit the proteolytic processing of this protein. To complicate further the picture, APP itself has also a role in neurogenesis and neuronal differentiation. Here, we tested the effects of human neuroblastoma SH-SY5Y cells differentiation on APP processing in cells stably transfected with a previously developed fluorescent construct. The chimeric construct (mBFP-APP-mGFP) is composed by APP695 and two different fluorescent proteins fused at the N-terminal and C-terminal ends. The value of the ratio between the intensities of the two fluorescent proteins, determined by means of imaging or flow cytometry, is correlated with alterations in the proteolytic processing of APP. Live cell imaging and flow cytometry results demonstrate a decrease of APP cleavage and a higher presence of its full-length form in SH-SY5Y cells differentiated with retinoic acid. Co-labeling with LysoTracker showed a higher accumulation of C-term APP-mGFP fragments in lysosomes in differentiated with respect to undifferentiated cells. An increased number of lysosomes was found in differentiated cells. Finally, we used Fura Red AM coupled to flow cytometer analysis to investigate any variations of intracellular Ca^{2+} concentration in SH-SY5Y cells treated with retinoic acid, or untreated, and in the presence, or absence, of APP overexpression. In agreement with previous literature, we noticed a dramatic decrease (~7 fold reduction) in Ca^{2+} levels in cells treated with retinoic acid that were not transfected. However, in cells overexpressing APP the effect of retinoic acid on calcium levels was only mild (~1 fold reduction). It is important to note that APP overexpression in undifferentiated cells did not lead to any significant increase in calcium levels. Our data underline that retinoic acid could contribute to the prevention of the development of Alzheimer's disease through the general inhibition of APP processing, and also highlight a higher sensitivity, in terms of changes in calcium levels, of differentiated cells to APP overexpression, with respect to undifferentiated cells.

Disclosures: **A. Nigro:** None. **Z. Lombardi:** None. **C. Carbone:** None. **M. Calamai:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.07

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

Support: NIH Grant R35 GM142726
NIH Training Grant NIA T32 AG052363
NIH Grant P20 GM125528
Presbyterian Health Foundation Pilot Grant

Title: Investigating the Role of the Amyloid Precursor Protein and $GABA_B$ Receptor in Neuroinflammation

Authors: *S. HOUMAM[1], S. BHANDARI[1], C. LACY[1], K. SHUKLA[1], D. BARBER[2], S. SIMONS[1], H. C. RICE[1];
[1]Oklahoma Ctr. for Geroscience & Healthy Brain Aging, Dept. of Biochem. & Mol. Biol., [2]Oklahoma Ctr. for Geroscience & Healthy Brain Aging, Dept. of Neurosci., Univ. of Oklahoma Hlth. Sci. Ctr., Oklahoma City, OK

Abstract: Neuroinflammation is a major component of Alzheimer's Disease (AD), and understanding the mechanisms involved in this response is critical to the development of new therapeutic approaches. Chronic neuroinflammation in AD is attributed in part to the responses of glial cells to accumulation of the amyloid-beta (Aβ) peptide. Aβ is derived from the Amyloid Precursor Protein (APP), which undergoes proteolytic processing to generate multiple products including secreted APP (sAPP). sAPP has protective effects on neurons that oppose the toxic effects of Aβ, but whether a similar pattern is present in glia is yet to be uncovered. We recently identified sAPP as a ligand for the $GABA_B$ Receptor ($GABA_BR$) to regulate synaptic activity in neurons. To explore the role this interaction plays in astrocytes and microglia, we used mouse primary microglia and astrocyte cultures, as well as organotypic brain slice cultures. We confirmed the expression of $GABA_BR$ subunits and isoforms in astrocytes and microglia using RT-qPCR and Western Blotting. We found that treatment with Baclofen, a $GABA_BR$ agonist, reduced pro-inflammatory cytokine release. These results offer insight into the involvement of $GABA_BR$ signaling in the inflammatory response of glial cells and represent the first step in uncovering the role of the $GABA_BR$-sAPP interaction in neuroinflammation and its potential as a target for therapeutics.

Disclosures: S. Houmam: None. **S. Bhandari:** None. **C. Lacy:** None. **K. Shukla:** None. **D. Barber:** None. **S. Simons:** None. **H.C. Rice:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.08

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

Support: NIH Grant NS115898

Title: Novel Candidate Effectors Of Amyloid Precursor Protein (APP) Signaling May Regulate Go(alpha)-Dependent Aspects Of Neuronal Migration And Guidance In A Model System.

Authors: *P. COPENHAVER;
Oregon Hlth. & Sci. Univ., Portland, OR

Abstract: Amyloid Precursor Protein (APP) is the source of beta-amyloid (Aβ) peptides that accumulate in Alzheimer's disease (AD). In addition, APP is upregulated in the developing and injured nervous system, where it can function as a guidance receptor that regulates multiple aspects of neuronal motility. However, the mechanisms by which APP signaling affects these

responses remain poorly understood. Early studies showed that APP interacted with the heterotrimeric G protein Goα to control neuronal responses, while misregulation of this pathway could provoke neurodegenerative responses. However, subsequent studies produced contradictory results, and proof that APP acts as an authentic Go-coupled receptor *in vivo* remained lacking. Defining the normal functions of APP has been complicated by two orthologs in mammals (APLP1 and APLP2) with overlapping activities. As an alternative, we adapted a well-characterized assay of neuronal migration in the moth Manduca, which expresses a single APP ortholog (APP-like; APPL) that also interacts with Goα in neurons. Inhibiting APPL-Go signaling in cultured embryos resulted in ectopic neuronal migration and outgrowth, analogous to the aberrant migration caused by deleting all APP family members in mice. Using genetic manipulations and split-GFP assays in Drosophila, we showed that APPL directly binds Goα via a conserved Go-binding motif, and that this interaction is regulated by Go activation. Using affinity screening methods, we then found that Manduca Contactin functions as an authentic ligand for APPL, paralleling evidence that mammalian Contactins also bind APP family proteins. These results demonstrate that Manduca provides a useful discovery system for defining the mechanisms that underlie this evolutionarily conserved pathway. Using our protocols for manipulating APPL in cultured embryos, we have now investigated candidate Goα effectors that may regulate different aspects of APPL-Goα signaling. All experiments were repeated at least 3 times ($N \geq 10$) using males and females, and results were analyzed blind to treatment conditions. Our recent studies indicate that APPL-Go signaling in the developing nervous system restricts ectopic neuronal migration via RhoGEF2, an ortholog of mammalian PDZ-RhoGEFs that activate the small GTPase RhoA (a regulator of actin remodeling). In addition, APPL-Go signaling can also induce APPL cleavage by the α-secretase ADAM10/Kuzbanian, providing a novel mechanism for terminating APPL activation. Understanding the normal mechanisms of APP-Go signaling may help identify new targets for treating diseases in which APP activity is misregulated.

Disclosures: P. Copenhaver: None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.09

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

Support: Pennsylvania Department of Health (Project Number: 4100087331)
NIH (NS120922 and AG069912)

Title: Amyloid precursor protein (APP) in myelin and nodes of Ranvier: implications for Alzheimer's disease

Authors: F. MA, ***K. HERRUP**;
Neurobio., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: APP (the amyloid precursor protein) is the parent protein of the Aβ peptide - the main constituent of the amyloid plaques found in the Alzheimer's disease brain. The generation of Aβ from APP is well studied, but the normal function of the full-length APP protein remains largely unexplored. Its structure is that of a Type I transmembrane protein that resembles a cell surface receptor or cell adhesion molecule. It is an evolutionarily ancient protein that is synthesized by most cells in the body beginning very early in development. Diagrams of its location in the brain tend to emphasize its presence in neurons, yet several studies, including single nuclear RNA-seq analyses, have shown its levels are just as high in oligodendrocytes. We have verified this location and begun to assess its significance. In vitro, the velate processes of mature oligodendrocytes are positive for APP immunostaining. In vivo, APP colocalizes with myelin markers such as myelin basic protein (MBP), confirming its presence in mature myelin. For axons sectioned longitudinally APP immunostaining appears as “railroad tracks” surrounding a central core of neurofilament light chain immunoreactivity. Axons cut transversely show “donuts” of staining that co-localize with MBP. Not unexpectedly, co-staining with MAP2 reveals the presence of APP in neuronal dendrites. In this location, however, it is interspersed with MAP2 and does not appear in the “railroad track” pattern seen surrounding neuronal axons. In Alzheimer's disease, APP levels are increased, independently of the presence of amyloid plaques. This increase is seen in oligodendrocytes as well. In R1.40 Alzheimer's disease model mice, the levels of APP staining that colocalize with MBP increase markedly indicating increased enhanced expression in myelin. Using MAP2 as a marker of neuronal dendrites, we find that APP levels are comparably increased in neurons. The presence of APP in both neurons and oligodendrocytes led us to ask about its presence in the node of Ranvier (NOR). As reported previously in optic nerve, APP is found in <10% of NOR of the neocortex in wild type mice. In Alzheimer's disease, the number of immunopositive NOR nearly triples. The NOR structure also changes. The node itself doubles in length, but the surrounding paranodes shrink leaving the length of the total NOR largely unchanged. These changes will reduce the energy efficiency of the axon and affect action potential velocity. All of these changes are found in both human and mouse and suggest that a full understanding of the biology of Alzheimer's disease is not possible without including oligodendrocytes and myelin in the analysis.

Disclosures: **F. Ma:** None. **K. Herrup:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.10

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

Support: NIH R01 AG067258

Title: Ad pathological changes in mouse model of amyloid and apoe are unaffected by chemotherapy

Authors: *C. S. NG[1], L. P. BIRAN[2], E. GALVANO[2], G. REBECK[2];
[1]Georgetown Univ. Med. Ctr., Georgetown Univ. Med. Ctr., Washington, DC; [2]Dept. of Neurosci., Georgetown Univ., Washington, DC

Abstract: Cancer survivors often experience cancer-related cognitive impairment (CRCI), which is characterized by problems of attention, working memory, and executive function following chemotherapy. APOE4, the strongest genetic risk factor for Alzheimer's Disease (AD), is also a risk factor for CRCI. We examined here whether the effects of APOE in CRCI were associated with an increase in AD pathological processes. We used a mouse model of amyloid, which expresses five familial AD mutations (5XFAD) along with the E3 or E4 alleles of human APOE (E3FAD and E4FAD). We treated these mice with doxorubicin, a chemotherapeutic drug commonly used for breast cancer treatment. Six-month-old female E3FAD mice (control n=5, treated n=5) and E4FAD (control n=6, treated n=6) were treated with either doxorubicin (10 mg/kg) or DMSO vehicle. After six weeks, mice were euthanized, and brains were perfused and collected; one hemisphere was fixed for immunohistochemistry and the other was dissected into cortex, hippocampus, and cerebellum for biochemical assays. Immunostaining with 6E10, an antibody for APP and amyloid, showed no differences in plaque accumulation across cortical and hippocampal brain regions after doxorubicin treatment in either APOE genotype. Consistent with other studies, we observed greater levels of plaques in regions of the untreated E4FAD mouse brains compared to the untreated E3FAD mouse brains. Sequential protein extraction with Tris-buffered saline (TBS), TBS with 1% Triton-X (TBSX), and formic acid (FA) was performed for measurements of Aβ42 and Aβ40 in the hippocampus. No effects of doxorubicin treatment were seen for Aβ42 nor Aβ40 levels in any fractions. AT8 levels, measured with western blots, indicated there were no effects of chemotherapy on phosphotau levels. We assessed glial reactivity to the presence of Aβ accumulation in the isocortex by co-staining with Moab2 for the detection of Aβ plaques and with either GFAP, an astrocytic marker, or Iba1, a microglia marker. While E4FAD controls showed increased microglia reactivity to diffuse plaques when compared to E3FAD controls, no effects of chemotherapy were found in microglial activation. Chemotherapy was also not generally associated with astrocytic responses: only reactivity to dense core plaques was increased in treated E3FADs when compared to control E3FADs. These data show no effects of doxorubicin on important aspects of AD pathogenesis, suggesting the effects of APOE on risk of CRCI are related to other aspects of cognitive dysfunction.

Disclosures: C.S. Ng: None. **L.P. Biran:** None. **E. Galvano:** None. **G. Rebeck:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.11

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

Support: NIH 1R01AG054598 - 01A1

Title: In vivo multiphoton microscopy reveals an increase in cytosolic and mitochondrial calcium in astrocytes caused by soluble Abeta

Authors: *M. SANCHEZ-MICO, M. CALVO-RODRIGUEZ, P. KELLY, S. WHITEMAN, E. KHARITONOVA, S. HOU, B. BACSKAI;
114 16th Street, Massachusetts Gen. Hosp., Boston, MA

Abstract: Amyloid β (Aβ) plaques are one of the main hallmarks of Alzheimer's disease (AD). We have previously reported that astrocytic cytosolic resting calcium (Ca^{2+}) is globally elevated in a transgenic mouse model of cerebral β-amyloidosis (APPswe/PS1dE9) using *in vivo* multiphoton microscopy. However, this phenomenon is independent of the proximity of astrocytes to Aβ plaques. In addition, soluble Aβ oligomers (Aβo) are thought to mediate neuronal toxicity by increasing neuronal calcium and causing synapse loss. For those reasons, we aimed to investigate how Aβo, rather than Aβ plaques, contribute to the Ca^{2+} insult in astroglial cytosol and mitochondria *in vivo*. To test this hypothesis, we topically applied naturally secreted soluble Aβo (conditioned medium from cultured transgenic primary neurons) onto the brain surface of 4-6-mo-old wild-type mice. We investigated changes on astrocyte and mitochondrial Ca^{2+} levels relative to baseline via intravital multiphoton imaging, by using a ratiometric genetically-encoded FRET-based Ca^{2+} indicator (Yellow Cameleon 3.6) targeted to astroglial cytosol or mitochondria, respectively. We observed that Ca^{2+} levels dramatically increased upon topical soluble Aβo application, in all cytosolic astrocyte compartments (soma, branches and end feet), but also in astrocyte mitochondria, which in excess could be harmful to the cells. Aβ-immunodepleted transgenic conditioned media and wild-type conditioned media did not alter astrocyte cytosolic or mitochondrial Ca^{2+}, supporting the specificity of the observed effects. Furthermore, we previously reported that upon blocking the mitochondrial calcium uniporter (MCU) with Ru360, calcium levels in neuronal mitochondria were restored. However, there was still an increase of mitochondrial calcium in astrocytes in presence of soluble Aβo and Ru360 in vivo. Thus, soluble Aβo is disrupting calcium levels in astrocytes through a different mechanism. Ultimately, these results together support a detrimental role of Aβo which leads to astrocyte Ca^{2+} dyshomeostasis *in vivo*, implying that Aβo are involved in the astrocytic dysfunction observed in AD. Future studies will continue addressing the source of soluble Aβo-induced Ca^{2+} increase, including intracellular Ca^{2+} stores and channels involved, which could have therapeutic value.

Disclosures: **M. Sanchez-Mico:** None. **M. Calvo-Rodriguez:** None. **P. Kelly:** None. **S. Whiteman:** None. **E. Kharitonova:** None. **S. Hou:** None. **B. Bacskai:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.12

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

Support: NIH Grant R01AG061151
NIH Grant R01AG055523
DH Chen Foundation (R-86U55A)

Title: Increased APP gene dose in Down syndrome induces deficits in the retromer complex

Authors: *X.-Q. CHEN[1], K. D. RYNEARSON[1], A. BECKER[1], D. KARACHENTSEV[1], E. DORAN[2], E. HEAD[3], R. E. TANZI[4], W. C. MOBLEY[1];
[1]Neurosciences, UCSD, La Jolla, CA; [2]Pediatrics, [3]Pathology & Lab. Med., UCI, Irvine, CA; [4]Neurol., Massachusetts Gen. Hosp., Charlestown, MA

Abstract: Motivation: The *retromer complex* plays an essential role in intracellular endosomal sorting. Deficits retromer complex function are linked to enhanced Aβ production. The levels of the components of the retromer complex are reported to be downregulated in Alzheimer's disease (AD). Down syndrome shares many neuropathological features with AD. Recent evidence also points to dysregulation of the retromer complex in DS, but in an AD-independent manner. Importantly, the mechanisms underlying retromer deficits in DS and AD are poorly understood. Methods: We measured the levels of retromer components in the frontal cortex of cases of AD-DS (AD in DS) as well as DS; the frontal cortex of a person partially trisomic for HSA21 (PT-DS), whose genome had only the normal two copies of the *APP* gene, was also examined. To explore the biological basis for changes in retromers we also analyzed these proteins in the Dp16 mouse model of DS. To further explore the molecular mechanism for changes in the retromer complex, we treated Dp16 mice with a γ-secretase modulator (GSM) (UCSD 776890), a treatment that reduces the levels of Aβ42 and Aβ40. Results: We found VPS26A and VPS29, but not VPS35, were significantly reduced in both DS and AD-DS, but not in PT-DS. Downregulation of VPS26A and VPS29 was recapitulated in the brains of only old Dp16 mice (at 20 months of age). Significantly, GSM treatment for 4 months beginning at age 16 months completely prevented reductions of the retromer complex. Conclusions: Together, our studies inform the mechanism underlying the deficits in the retromer complex in DS by pointing to a necessary role of increased *APP* gene dose. Additionally, studies in the mouse model give evidence for a causal role for Aβ species in reducing protein components of the retromer complex. They raise the possibility that Aβ species are not only increased by retromer complex deficiency but also induce them.

Disclosures: X. Chen: None. **K.D. Rynearson:** None. **A. Becker:** None. **D. Karachentsev:** None. **E. Doran:** None. **E. Head:** None. **R.E. Tanzi:** None. **W.C. Mobley:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.13

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

Support: MOE Grant MOE2018-T1-002-033
MOE Grant MOE2017-T3-1-002
Nanyang Technological University NAP Start-up fund

Title: Amyloid-beta-mediated nuclear pore complex dysfunction in a mouse model of Alzheimer's disease

Authors: *V. A. BANSAL[1], J. M. TAN[1], H. R. SOON[1], T. SAITO[2], T. C. SAIDO[3], T. CH'NG[1]; [1]Home, Lee Kong Chian Sch. of Medicine, NTU, Singapore, Singapore; [2]Dept. of Neurocognitive Science, Inst. of Brain Sci., Nagoya City Univ. Grad. Sch. of Med. Sci., Nagoya City, Japan; [3]Lab. for Proteolytic Neurosci., RIKEN Ctr. for Brain Sci., Wako, Japan

Abstract: The nuclear pore complex (NPC) is a vital component of the nuclear membrane that regulates multiple processes such as protein movement across the nuclear membrane, chromosome organisation, and transcriptional regulation. While there is a gradual loss of NPCs in neurons during normal ageing, further NPC dysfunction has been reported in neurodegenerative disorders such as frontotemporal dementia, Huntington's disease, and Amyotrophic lateral sclerosis. There are few reports on the state of NPCs in Alzheimer's disease (AD), and fewer still that link amyloid-beta (Aβ) with nuclear pore dysfunction. Here, we show evidence that AD-associated Aβ expression accelerates the loss of select nucleoporins, NUP98 and NUP107, and a reduction in overall number and distribution of NPCs on the nuclear envelope of hippocampal neurons. The increase in intracellular Aβ concentration correlates with the severity of the NPC loss in a time- and age-dependent manner. This loss of NPCs degrades the nuclear permeability barrier which leads to defective subcellular compartmentalization of nucleocytoplasmic proteins, and an impairment of the active import of proteins via the classical nuclear import pathway. As a result of this nuclear pore dysfunction, AD neurons become more vulnerable to inflammation-induced necroptosis - a programmed cell death pathway where the core components are activated via phosphorylation through nucleocytoplasmic shutting. Collectively, our results suggests that an Aβ-driven mechanism is responsible for enhancing age-dependent nuclear pore damage, with potential consequences for neuronal loss in AD.

Disclosures: V.A. Bansal: None. **J.M. Tan:** None. **H.R. Soon:** None. **T. Saito:** None. **T.C. Saido:** None. **T. Ch'ng:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.14

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

Support: ANID-FONDECYT 1200908 (J.F.)
MillenMiNuSPain, Millennium Scientific Initiative NCN19_038
ANID-FONDECYT 1211082 (G.E.Y.)

ANID-FONDECYT 1211095 (G.M.)
grant fellow 21200612

Title: Increases calpain-1 levels by P2X2R activation promote the Abeta peptide generation & potentiate their cellular toxicity

Authors: J. GAVILAN[1], J. PANES-FERNANDEZ[1], P. A. GODOY[1], O. RAMIREZ[1], R. DURAN[1], P. A. CASTRO[1], C. MUNOZ-MONTESINOS[1], G. YEVENES[1], G. MORAGA[1], E. L. JARA[2], *J. FUENTEALBA[3];
[1]PHYSIOLOGY, [2]PHARMACOLOGY, [3]Univ. de Concepción, Univ. de Concepción, Concepcion, Chile

Abstract: Beta-amyloid peptide (Aβ) forms pore-like structures in cell membranes; our group we have shown that ATP leaks through this pore, activating purinergic P2X receptors (P2XR). The P2XRs represent a family of 7 distinct subunits with high Ca^{2+} conductance. In our hands, exposure to Aβ was found to induce a specific increase in P2X2 receptor (P2X2R) levels. Increases in $[Ca^{2+}]i$ is able to activate cytosolic proteases called calpains. Two types of calpains are present in the brain: calpain-1 and calpain-2, and it has been described that the overactivation of these proteases produces an increase in the APP amyloidogenic pathway, which leads to an increase in the Aβ peptide formation. Briefly, when calpains are activated by an increase in $[Ca^{2+}]i$, they produce a p35 protein cleavage, generating the p25 fragment, which acts as a neuronal activator of cyclin-dependent kinase 5 (Cdk5). Once activated, Cdk5 phosphorylates to the STAT3 transcription factor, and p-STAT3 goes to the nucleus to upregulate BACE1 transcription. The increase in the expression of the enzyme BACE1 is associated with a greater proteolysis of APP by the amyloidogenic pathway and an increase in the Aβ peptide generation. In this work, we study the activity of calpains after P2X2 purinergic receptors activation to determine its impact on the amyloidogenic pathway and Aβ formation, in cell models that overexpress P2X2R. In PC-12 cells overexpressing P2X2R, 100 µM ATP produced an increase in calpain-1 protein levels at 30 and 60 min (36±4% and 46±6%, respectively), while calpain-2 levels were not altered. The endogenous calpain inhibitor: calpastatin, did not present variations in its expression at the different times of ATP exposure. Furthermore, we observed that spectrin; calpain-specific substrate, its 240/150 kDa ratio decreased about 30 and 60 min of incubation with 100 µM ATP. (38±5% and 36±5%, respectively) On the other hand, PC-12 cells that overexpress P2X2R showed an increase in the p25/35 ratio at 60 min of incubation with 100 µM ATP (23±4%), while co-incubation with the calpain inhibitor MDL28170 maintained the p25/35 ratio with values close to the control. In addition, in PC-12 cells that overexpress P2X2R, an increase in the levels of the enzyme BACE1 and a decrease in the expression of the β-carboxyterminal fragment of APP (C99) were observed, which correlated with an increase in the levels of the Aβ peptide and increased cell toxicity. Our results suggest that the P2X2R-calpain1 axis is involved in the reinforced beta-amyloid generation, and this could represent a novel, unexplored pathway in Alzheimer's disease, opening a new space to develop pharmacological strategies to treat AD.

Disclosures: J. Gavilan: None. **J. Panes-fernandez:** None. **P.A. Godoy:** None. **O. Ramirez:** None. **R. Duran:** None. **P.A. Castro:** None. **C. Munoz-montesinos:** None. **G. Yevenes:** None. **G. Moraga:** None. **E.L. Jara:** None. **J. Fuentealba:** None.

Poster

531. APP and Metabolites: Function and Processing

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 531.15

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

Support: NRF-2022R1A2C1012031
NRF-2019R1F1A1063005

Title: Low dose of corticosterone attenuates beta-amyloid-induced neurotoxicity in SH-SY5Y cells

Authors: B. DO[1], **S. SHIN**[1], C. LEE[2], J.-H. JANG[3], *G. PARK[1];
[1]Kyungpook Natl. Univ., Daegu, Korea, Republic of; [3]Dept. of Pharmacol., [2]Sch. of Medicine, Keimyung Univ., Daegu, Korea, Republic of

Abstract: Alzheimer's disease (AD) is a neurodegenerative disease characterized by neuronal cell death and memory impairment. Corticosterone (CORT) is a glucocorticoid hormone produced by the hypothalamic-pituitary-adrenal axis in response to a stressful condition. In this study, the effects of low dose CORT on Aβ-induced neurotoxicity in SH-SY5Y cells and underlying molecular mechanisms have been investigated. Cytotoxicity caused by Aβ was significantly inhibited by the low dose of CORT treatment in the cells. Furthermore, CORT pretreatment significantly reduced Aβ-mediated pro-apoptotic signals, such as increased Bim/Bcl-2 ratio and caspase-3 cleavage. Moreover, low dose of CORT treatment inhibited the Aβ-induced cyclooxygenase-2 and pro-inflammatory cytokine expressions, including tumor necrosis factor-α and interleukin-1β. Aβ resulted in intracellular accumulation of reactive oxygen species and lipid peroxidation, which were effectively reduced by the low dose of CORT. As a molecular mechanism, low dose of CORT activated the NF-E2-related factor 2, a redox-sensitive transcription factor mediating cellular defense and upregulating the expression of antioxidant enzymes, such as NAD(P)H:quinone oxidoreductase, glutamylcysteine synthetase, and manganese superoxide dismutase. These findings suggest that low dose of CORT exerts protective actions against Aβ-induced neurotoxicity and might be used to treat and/or prevent AD.

Disclosures: B. Do: None. **S. Shin:** None. **C. Lee:** None. **J. Jang:** None. **G. Park:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.01

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

Title: Ang - a critical factor in pathological aging and alzheimers disease?

Authors: *M. JOERG[1], J. E. PLEHN[1], T. NGUYEN[2], L. BESSLER[1], K. ENDRES[2], M. HELM[1], K. FRIEDLAND[1];
[1]Inst. of Pharmaceut. and Biomed. Sci., Johannes Gutenberg Univ., Mainz, Germany;
[2]Psychiatry and Psychotherapy, Univ. Med. Ctr. of the Johannes Gutenberg Univ., Mainz, Germany

Abstract: Human angiogenin (ANG) belongs to the major gene superfamily, which exclusively expresses secreted ribonuclease (RNase) in vertebrates, mainly located in the nucleus. Besides its function in neovascularization, ANG also plays a crucial role in stress response and protecting cells. As a result of stress, ANG translocates from the nucleus to the cytoplasm resulting in the cleavage of cytoplasmic tRNAs into stress-induced ANG-mediated tRNA halves (tiRNAs), resulting in the inhibition of the pro-apoptotic signal pathway and caspase 3 activity which leads to a protective effect on cells. First publications reveal different ANG variants in amyotrophic lateral sclerosis and Parkinson's disease, thus linking ANG and neurodegenerative diseases for the first time. Using Western Blot, we examined the expression of ANG in different aging and Alzheimer's Disease (AD) cell, animal, and human post-mortem models. Our group determined an age, gender, and AD-dependent dysregulation of ANG in animal and human post-mortem cortical brain samples. We also analyzed RNAseq data from the human Aging, Dementia and TBI study and confirmed the data obtained. These age-specific results suggest that ANG is a critical factor in the further development of AD, as aging per se is the greatest risk factor for developing neurodegenerative disease. In addition to that, tRNA modifications play a crucial role in RNA function and stability. So far, 150 different RNA modifications have been discovered, but the role of tRNA modifications in pathological aging and AD is still unknown. Therefore, we used Liquid Chromatography with tandem coupled mass spectrometry (LC-MS/MS) to determine whether specific tRNA modifications in cell and animal models contribute to mitochondrial defects following the dynamic changes in tRNA modifications pathological process of AD. The tRNA modification 5-methylcytosine (m^5C) located in the anticodon and variable loop region at positions 34/38/48/49/50 plays a crucial role in the stress-induced ANG-mediated tRNA cleavage. First results of LC-MS/MS revealed various changes in tRNA modifications in pathological aging and AD, especially for m^5C. This line of research could be a new road to defining early biomarkers for AD and represent an important step toward developing new therapeutic strategies to improve the symptoms of AD patients.

Disclosures: M. Joerg: None. **J.E. Plehn:** None. **T. Nguyen:** None. **L. Bessler:** None. **K. Endres:** None. **M. Helm:** None. **K. Friedland:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.02

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

Support: NIH Grant NS051715
NIH Grant AG030539
NIH Grant AG051266
NIH Grant AG059695
NIH Grant AG065651
Carlos III Institutes of Health
Queen Sofia Foundation

Title: Translational potential of Clusterin in the pathogenic mechanisms of Aβ and non-Aβ cerebral amyloidosis

Authors: *A. A. ROSTAGNO[1], M. CALERO[2], J. HOLTON[3], T. REVESZ[3], T. LASHLEY[3], J. GHISO[4];
[1]New York Univ. Sch. of Med., New York Univ. Sch. of Med., New York, NY; [2]Inst. de Salud Carlos III, Inst. de Salud Carlos III, Majadahonda - MADRID, Spain; [3]Univ. Col. of London, London, United Kingdom; [4]New York Univ. Grossman Sch. of Med., New York, NY

Abstract: Clusterin, also known as apolipoprotein J, is increasingly recognized as an important contributor to many pathological conditions including systemic and localized amyloidosis. In Alzheimer's disease (AD), the most common form of dementia, clusterin genetic variants and plasma levels were identified as risk factors for the disorder while investigations into the potential pathways modulated by the apolipoprotein demonstrated its ability to bind soluble Aβ in biological fluids, facilitate its brain clearance, and prevent its aggregation. To investigate whether clusterin exerted a more general role in the pathophysiology of cerebral amyloidoses, we turned to two unrelated hereditary conditions, familial British and Danish dementias (FBD and FDD), which share striking neuropathological similarities with AD, including neurofibrillary degeneration in limbic areas as well as parenchymal and vascular amyloid deposition. Two different 4 kDa molecules - ABri in FBD and ADan in FDD - totally unrelated to the 4 kDa Alzheimer's Aβ, are the main constituents of the amyloid deposits in these forms of cerebral amyloidoses. Using brightfield and a combination of single and double immunofluorescence microscopy the current work demonstrated clusterin co-localization with brain parenchymal and cerebrovascular amyloid deposits in both disorders, mirroring findings in AD. Clusterin was identified as the major ABri- and ADan-binding plasma protein using ligand affinity chromatography with downstream Western blot and amino acid sequence analyses. Further analysis using solid-phase enzyme-linked immunosorbent assays highlighted a specific saturable binding of clusterin to ABri and ADan and demonstrated Kd values within the same low nanomolar range as those identified for the clusterin-Aβ interaction. Consistent with its reported chaperone activity, clusterin showed a modulatory effect on ABri, ADan, and Aβ aggregation/fibrillization properties evaluated by thioflavin T binding assays, suggesting the potential participation of the apolipoprotein in common protective molecular pathways. Our findings, together with the known multifunctional activity of clusterin and its modulatory activity on the complex cellular pathways leading to oxidative stress, mitochondrial dysfunction, and the induction of cell death mechanisms - all known pathogenic features of protein folding disorders, including AD, FBD, and FDD - suggests the likelihood of a more complex role and a translational potential of clusterin in the amelioration/prevention of these pathogenic mechanisms.

Disclosures: **A.A. Rostagno:** None. **M. Calero:** None. **J. Holton:** None. **T. Revesz:** None. **T. Lashley:** None. **J. Ghiso:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.03

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

Support: TUBITAK-Grant#1919B012004851

Title: Layer specific morphological changes in the rat motor cortex following viral vector mediated TDP-43 overexpression

Authors: *E. ULUPINAR[1,2], E. POLAT CORUMLU[1], B. CAKıR[3], I. ARMAGAN[4], H. KAPKAC[5], M. ARSLANYOLU[5];
[1]Interdisciplinary Neurosci., Eskisehir Osmangazi Univ., Eskisehir, Turkey; [2]Anat., [3]Eskisehir Osmangazi University, Fac. of Med., Eskisehir, Turkey; [4]Hacettepe University, Fac. of Med., Ankara, Turkey; [5]Biol., Eskisehir Tech. University, Fac. of Sci., Eskisehir, Turkey

Abstract: In amyotrophic lateral sclerosis (ALS) and fronto-temporal dementia (FTD), selective population of neurons appears to be more vulnerable to neurodegeneration and mutation in TarDNA-binding protein-43 (TDP-43)has been shown as one potential candidate playing role in underlying pathology. In the current study, we aimed to investigate the number of neurons in different layers of the motor cortex following overexpression of TDP-43 in the central nervous system with the use of a stable, constitutive and ubiquitous promoter for gene expression. Sprague-Dawley male rats (n=10) was injected with Adeno-Associated Virus serotype 9 at the dose of $6.4X10^{12}$ gc/ml, containing the native TDP-43 with GFP under CMV promoter (AAV9-pCMV-TDP43-GFP). Control groups were received IV injections of either saline (SF) or vectors encoding only GFP (AAV9-pCMV-GFP) at postnatal day 30. Phenotypic characterisation of animals was determined by using motor behavioural tests including the rota-rod, horizontal ladder and novel object recognition tests. Following transduction, TDP-43 delivery to the central nervous system produced a rapid and highly consistent motor deficit especially in the hindlimbs of animals. Four weeks later they were sacrificed by intracardiac perfusion under ketamine and xylazine anaesthesia. Immunohistochemically stained frozen sections with NeuN antibody selected from the motor cortex region were analysed by using systematic randomised sampling method. Statistical analyses showed that the number of NeuN (+) neurons per unit area in the layer 5 of the motor cortex was decreased significantly ($p<0.05$) after transduction with TDP-43 compared to the control groups. On the other hand, density of neurons in the layer 2/3 displayed no significant differences. Our data show that AAV9-based TDP-43 overexpression caused alterations in the motor cortex specifically in deep layers containing the corticospinal motor neurons while sparing the callosal projection neurons. This relatively low-cost and rapidly

induced animal model might be used to investigate TDP-43 mediated disease mechanisms and to test novel therapeutic approaches.

Disclosures: E. Ulupinar: None. **E. Polat Corumlu:** None. **B. Cakır:** None. **I. Armagan:** None. **H. Kapkac:** None. **M. Arslanyolu:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.04

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

Title: Meta-analysis of NIH Alzheimer's Disease-Related Dementias (ADRD) programs responsive to the National Plan to Address Alzheimer's Disease (AD)

Authors: A. J. MCCARTNEY, K. WHITAKER, S. DODSON, E. BRYANT, A. K. SHUKLA, X. YIN, K. WHITEHEAD, R. A. CORRIVEAU;
NIH/National Inst. of Neurolog. Disorders and Stroke, Bethesda, MD

Abstract: AD/ADRD diseases currently impact more than 6 million people in the United States. Rare forms of AD/ADRD are caused directly and unambiguously by genetic mutations. However, most AD/ADRD burden is complex in etiology and thought to result from an interplay among multiple incompletely understood genetic, biological, lifestyle, environmental and psychosocial risk factors. Moreover, these risk factors interact with other health factors to impact AD/ADRD outcomes. For example, while the most common dementia diagnosis is AD, research over the past decade revealed that most people with a diagnosis of AD have multiple brain pathologies, other comorbidities, and many people diagnosed with clinical AD do not have AD pathology at all. This new knowledge highlights the importance of better understanding dementia syndromes, and the relationships among them, to increase chances of developing effective interventions. The National Institute of Neurological Disorders and Stroke (NINDS) partners with the National Institute on Aging, the National Institutes of Health (NIH) lead for AD/ADRD, in the NIH response to the National Plan. NINDS leads ADRD including frontotemporal dementia (FTD), Lewy body dementias (LBD), vascular contributions to cognitive impairment and dementia (VCID) and multiple etiology dementias (MED). This project aims to evaluate NIH and the ADRD field's responsiveness to ADRD research milestones from triennial NINDS ADRD Summits by mapping NIH-funded research activities to ADRD milestones. The approach used text-based analysis/mining of grant titles, abstracts, and specific aims to map awards and activities to specific ADRD milestones. Mapping was followed by expert vetting and concurrence, including for funding opportunity announcements. The study includes all new or competitively renewed NIH ADRD awards out of total $1.15 Billion toward NIH funded ADRD research from fiscal years 2017-2019. Findings will help NIH to better understand the state of the field, identify research gaps and opportunities for future efforts, and enable public communication of research activities with an accurate, accessible, compact, and updatable data

visualizations. Results indicate the approach is feasible, and importantly the field and NIH have largely been responsive to the ADRD milestones in the National Plan to Address AD. The analysis points to several cross-cutting ADRD research areas that may benefit from increased attention going forward, including health equity, the multiple etiology hypothesis of common dementias, emerging risk factors for common dementias (e.g. COVID-19, traumatic brain injury, and TDP-43 proteinopathy), and clinical trials.

Disclosures: A.J. McCartney: None. **K. Whitaker:** None. **S. Dodson:** None. **E. Bryant:** None. **A.K. Shukla:** None. **X. Yin:** None. **K. Whitehead:** None. **R.A. Corriveau:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.05

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

Support: NIH grant F31NS116947
Hydrocephalus Association 2020 Innovator Award
Fraternal Order of Eagles (University of Iowa)
Kwak-Ferguson Award (University of Iowa)

Title: Kaolin-induced hydrocephalus as model of normal pressure hydrocephalus

Authors: *M. TISH, J. GEERLING;
Neurol., Univ. of Iowa, Iowa City, IA

Abstract: Normal pressure hydrocephalus (NPH) is a chronic form of communicating hydrocephalus in older adults, in which the cerebral ventricles expand, yet the intracranial pressure is not raised above normal levels. NPH produces a triad of progressive symptoms: gait impairment, urinary incontinence, and cognitive dysfunction. Shunt insertion can treat NPH symptoms, but often does not completely eliminate them, with gait improving more often than cognition and micturition. Additionally, the brain pathophysiology underlying the symptoms remains unknown, preventing more targeted treatments. Kaolin (aluminum silicate) has been used to produce mammalian models of hydrocephalus for decades, but its use in rodents has been limited to a small number of studies with typically severe, fatal hydrocephalus. We developed a chronic, survivable model of communicating hydrocephalus with symptoms similar to human NPH. We inject 5 µL of a 10-15% kaolin suspension into the cisterna magna of adult mice, which produces gradually progressive enlargement of the cerebral ventricles. We use brain MR imaging to measure ventricular volume at different points before and after kaolin injection. We also perform weekly tests of bladder function (Micturition Video Thermography), gait and balance, and a variety of behavioral testing 10 weeks after kaolin injection. Hydrocephalic mice, but not saline-injected controls, develop impairments in gait and balance as well as urinary frequency with incontinent features. Intracranial pressure is not elevated at any time points we

have measured (1-3 & 10 weeks). Inserting a shunt into the right lateral ventricle at different time points following kaolin injection affects ventricular volume and behavior. Ventricular enlargement is correlated with weight loss in the two weeks following kaolin injection. Brain tissue from hydrocephalic mice shows several periventricular abnormalities that may help account for differential symptoms. This model allows us to investigate the pathophysiology of chronic hydrocephalus, and can be combined with other neuroscience techniques to test additional, non-shunt treatments for NPH.

Disclosures: M. Tish: None. **J. Geerling:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.06

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

Support: The Weinbaum Lakritz Research Fund

Title: The Sorting Factor VPS13A is Pivotal for Quality Control of Mitochondria and Endoplasmic Reticulum

Authors: G. RODRIGUEZ[1], E. TANK[1], R. FULLER[2], A. M. TIDBALL[1], T. QIAO[1], J. M. PARENT[1], S. BARMADA[1];
[1]Neurol., [2]Biol. Chem., Univ. of Michigan, Ann Arbor, MI

Abstract: Chorea acanthocytosis (ChAc) is an autosomal-recessive neurodegenerative disorder caused by mutations in the gene encoding vacuolar protein sorting factor 13A (VPS13A). The yeast orthologue, VPS13, aids in the transport of lipids between organelles, and the N-terminal region shows strong sequence homology to the evolutionarily conserved autophagy-related genes (*Atg*) protein family. Although these studies suggest a function for VPS13 proteins in lipid transfer and autophagy, the physiological function and regulation of VPS13A in mammalians cells remains unknown.
Here I utilized HEK293T cells in which the native VPS13A has been labeled with mNeonGreen, to identify VPS13A interactors by mass spectroscopy and unbiased proteomics. I also used VPS13A-KO HEK293T cells to determine how functional loss of VPS13A affects critical pathways relevant to ChAc. These studies, as well as published work on yeast VPS13, led me to investigate specialized branches of autophagy that may be regulated by VPS13A. Loss of VPS13A affects the level of receptors involved in mitochondria-specific autophagy (mitophagy) and endoplasmic reticulum-limited autophagy (ER-phagy). These findings highlight a central function for VPS13A in ER-phagy and mitophagy, a hypothesis I am now exploring through dynamic studies of ER-phagy and mitophagy in HEK293T cells.
Future studies will examine the contribution of VPS13A to protein clearance and cellular survival, in human neurons differentiated from induced pluripotent stem cells donated by

individuals with ChAc. Collectively, these investigations will help define a function for VPS13A in neurons, outline disease mechanisms, and highlight pathways that may be targeted to prevent neuron loss in this disorder.

Disclosures: **G. Rodriguez:** None. **E. Tank:** None. **R. Fuller:** None. **A.M. Tidball:** None. **T. Qiao:** None. **J.M. Parent:** None. **S. Barmada:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.07

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

Support: NRF Grant 2022R1 C1C1 006166

Title: Progression of Alzheimer's disease with NTN4/Tau signals via Inflammatory Cytokines activations in the Gut

Authors: *H. JEON[1], E. KANG[1], S. JANG[1], Y. LEE[1], Y. KIM[1], D. HONG[1], S. LEE[1], X. LIU[2], S. KANG[2], E. AHN[1];
[1]Hallym Univ., Hallym Univ., Chuncheon, Korea, Republic of; [2]Dept. of Pathology and Lab. Med., Emory Univ. Sch. of Med., Atlanta, GA

Abstract: Alzheimer's disease (AD) neuropathological hallmarks include senile plaques with aggregated amyloid beta as a major component, neurofibrillary tangles (NFT) containing truncated and hyperphosphorylated Tau, a large amount of neuronal cell loss, and chronic neuro-inflammation. The recent research studies supported that the gut/brain axis is bidirectional, and the gut microbiome trigger neurodegenerative disord ers including Alzheimer's disease (AD). However, the gut/brain axis related molecular mechanism that dominates the pathogenesis of AD is not yet clear. Here we show that the NTN4/Tau molecular signaling spatiotemporally mediates chronic inflammation with hyperphosphorylated or aggregated Tau in the gut enteric neurons. We orally administered the 0.5% DSS during for 3 months to generate the gut chronic inflammation in young MAPT mice (3 months) and age-matched non-transgenic mice. We observed the 0.5% DSS treated MAPT mice involved Tau aggregation, NTN4 reduction and cytokines activations in the enteric neurons but not in the control group. Moreover, we investigated the NTN4 mRNA levels on Gene Expression Omnibus (GEO) database in AD VS age-matched CTL groups brain samples. Strikingly, we observed the NTN4 deprivation in AD patient brain samples not in healthy control brain samples. Our findings suggest the hypothesis that NTN4/Tau molecular mechanism is activated by gut inflammation, implicated in AD pathogenesis in the gut.

Disclosures: **H. Jeon:** None. **E. Kang:** None. **S. Jang:** None. **Y. Lee:** None. **Y. Kim:** None. **D. Hong:** None. **S. Lee:** None. **X. Liu:** None. **S. Kang:** None. **E. Ahn:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.08

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

Support: NIH Grant R01 NS115716
a Chan Zuckerberg Initiative Ben Barres Early Career Acceleration Award

Title: The Gag-like PNMA2 protein forms virus-like capsids that cause autoantibody production in a paraneoplastic neurological syndrome.

Authors: *J. XU[1], S. ERLENDSSON[3], M. REGIER[1], J. A. G. BRIGGS[3], S. L. CLARDY[2], J. D. SHEPHERD[1];
[1]Dept. of Neurobio., [2]Dept. of Neurol., Univ. of Utah, Salt lake city, UT; [3]MRC Lab. of Mol. Biol., Cambridge, United Kingdom

Abstract: The *paraneoplastic Ma antigen* (*PNMA*) genes are associated with paraneoplastic syndromes that present with neurological symptoms. Why these particular genes, which encode intracellular proteins, cause a severe autoimmune response is unclear. *PNMA2* is conserved in mammals and highly expressed in the brain, but not normally expressed outside the CNS. Intriguingly, PNMA2 and other family members are homologous to the retroviral Gag protein. We recently discovered that another brain gene, Arc, has similar homology and retains retroviral properties such as capsid formation. We purified mouse and human PNMA2 protein from *E.coli*. Negative-staining electron microscopy (EM) showed that purified PNMA2 protein forms virus-like capsids. Cryo-EM further showed mouse PNMA2 forms T1 capsids. To test whether PNMA2 is released by neurons, we collected media from mouse primary cortical neuronal culture and used size-exclusion chromatography to purify particles. PNMA2 was found in particle fractions of neuronal media, similar to extracellular vesicles (EVs). However, when we performed a Proteinase K protection assay, PNMA2 protein was degraded by Proteinase K without detergent present to disrupt membranes. This suggests that PNMA2 is not released in EVs. To further isolate PNMA2 particles, we conducted iodixanol gradient ultracentrifugation, and found that PNMA2 protein was in higher density fractions than EVs. We isolated the PNMA2 fraction on an EM grid and observed PNMA2 capsids. Based on these findings, we hypothesize that tumor cells release naked PNMA2 capsids that activate the immune system to generate autoantibodies. To test the immunogenicity of PNMA2 capsids, we injected mice (P42-56, 4 mice/group) with mouse PNMA2 capsids or vehicle. Compared with vehicle-injected mice, ELISA showed PNMA2 injected mice produce a significant high titer of PNMA2 antibodies three weeks after injection. As the control, purified endophilin did not induce endophilin antibodies. These PNMA2 antibodies bind mouse PNMA2 capsids, as shown by immunogold labeling. Similarly, PNMA2 autoantibodies in paraneoplastic patient CSF also bind human PNMA2 capsids. These data support the hypothesis that naked PNMA2 capsids trigger the production of autoantibodies in paraneoplastic patients. Taken together, we found that another

brain gene can form virus-like capsids, but unlike Arc, PNMA2 capsids are not released in EVs. This suggests a novel secretory pathway in neurons. The ability to form capsids also provides a mechanism for immunogenicity, when abnormally expressed in peripheral tumors. Further work will determine the normal function of PNMA2 intercellular signaling in the brain.

Disclosures: **J. Xu:** None. **S. Erlendsson:** None. **M. Regier:** None. **J.A.G. Briggs:** None. **S.L. Clardy:** None. **J.D. Shepherd:** Other; co-founder and consultant for VNV, LLC.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.09

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

Support: AFTD Holloway Fellowship

Title: Machine learning reveals behavioral differences in a tauopathy model

Authors: *D. E. OKOBI, Jr[1,2], P. GOLSHANI[1,2];
[1]UCLA Dept. of Neurol., Los Angeles, CA; [2]West Los Angeles VA Med. Ctr., Los Angeles, CA

Abstract: In recent decades, research in dementia has proceeded outward from basic pathology to the clinical manifestations of disease. This has led to revolutionary advancements in our understanding of the molecular and cellular mechanisms of neurodegeneration. However, actual patients are often diagnosed much later, when symptoms affect their daily function. Early detection and treatment may hold the key to more effective therapies for this chronic, terminal illness. In frontotemporal dementia (FTD), the disease is often marked by earlier onset and behavioral changes such as compulsions and apathy that precede more typical symptoms of dementia, such as memory loss. There are stronger genetic ties in the pathophysiology of FTD, with about half of cases being characterized by tau aggregation. Traditionally, behavior has been difficult to study because close description by trained humans is time-intensive and prone to subjectivity. In the past few years, applications of machine learning to animal behavior have led neuroscientists to new insights and higher throughput studies. I hypothesized that automated behavioral observation is an effective way to identify the earliest changes. Using MoSeq (Motion Sequencing), an unsupervised machine learning program pioneered by Robert Datta's lab, I examined hTau.P301S (Goedert) mice between 4 and 6 months of age. A depth camera was used to track free exploration behavior of single animals in an open cylindrical chamber over at least 5 20-minute sessions. Mouse kinematic data was automatically extracted, allowing principal components undergirding movements to be established. From these, several dozen discrete behavioral syllables could be defined. Both the distribution and sequence relationship of the syllables differed significantly between heterozygote animals and littermate controls, allowing for phenotypic discrimination at an earlier age than previously reported in the literature. (These differences were abolished when the behavioral data was re-analyzed after genetic identity was

randomly reassigned.) Earlier detection and characterization of subtle behavioral differences in animal models could allow for more precise understanding of the neural basis of such behaviors, as well as earlier interventions to slow neurodegenerative decline.

Disclosures: D.E. Okobi: None. **P. Golshani:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.10

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

Support: NIH Grant 5R01AG057290-03
NIH Grant 5R01AG053999-03
NIH Grant 5R01NS116058

Title: Investigating the Neuroimmune Axis in Amylin-Induced Type-2 Diabetic Brain Injury

Authors: *E. WINFORD[1], N. VERMA[2], A. STOWE[3], F. DESPA[2];
[1]Neurosci., [2]Pharmacol. and Nutritional Sci., [3]Neurol., Univ. of Kentucky, Lexington, KY

Abstract: Background: Type-2 diabetes is a metabolic disorder that increases the risk for cerebrovascular diseases and dementia. We and others reported previously that this risk is associated with elevated blood levels of amylin (an amyloidogenic hormone synthesized and co-secreted with insulin), which promotes amylin amyloid deposition in the brain microvasculature (cerebral amylin vasculopathy). We have shown that inducing amylin dyshomeostasis in rats by pancreatic-specific overexpression of human amylin (HIP rats) leads to cerebral amylin vasculopathy and neurological deficits. Here we tested the hypothesis that neuroinflammation caused by cerebral amylin vasculopathy leads to a dysregulated immune response in the brain. Methods: We conducted brain RNAseq analysis in 16-month-old male HIP rats and normal rats expressing wild-type (WT) rat amylin (n =10/group). Library and construction, and sequencing were performed using commercial services (Omega Bioservices). Isolated brain homogenates from WT and HIP (n =7/group) rats were used for western blot analysis for proteins in the brain that are important for immune cell migration into the brain. Immunohistochemistry (IHC) was conducted on WT and HIP (n=10/group) rats to determine the activation of resident immune cells. We finally conducted flow cytometry in the blood, spleen, and brains of WT and HIP rats (n = 5-7/group) to determine immune cell migration during cerebral amylin vasculopathy. Results: In the brains of HIP rats, RNAseq analysis shows significantly altered pathways important for neuroinflammation signaling, T cell signaling, and B cell signaling compared to WT littermates. We also saw significant changes in proteins such as VCAM-1 and ICAM-1 by western blot. In IHC, we see an increased activation of microglia shown by increases in IBA-1 and CD68. Finally, we see an overall decreased number of immune cells in the blood, spleen, and brain compared to WT rats. Conclusion: Increased amylin levels in the circulation cause

significant alterations in genes in the brain involved in neuroinflammation and immune cell signaling, proteins involved in immune cell migration, activation of resident immune cells in the brain, and decreased immune cell migration. These data suggest that cerebral amylin vasculopathy contributes to diabetes-related neuroimmune signaling through altering genes, altering immune cell migration, and altering proteins important for immune cell migration and activation.

Disclosures: E. Winford: None. **N. Verma:** None. **A. Stowe:** None. **F. Despa:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.11

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

Support: Weston Brain Institute #TR150199

Title: Hipsc-derived grn-deficient astrocytes delay spiking activity of developing neurons

Authors: *C. LEE[1], J. FREW[1], N. L. WEILINGER[3], S. WENDT[4], W. CAI[1], S. SORRENTINO[1], X. WU[1], B. A. MACVICAR[2], S. WILLERTH[5], H. B. NYGAARD[1]; [2]Ctr. for Brain Health/Psychiatry, [1]Univ. of British Columbia, Vancouver, BC, Canada; [3]Psychiatry, Ctr. For Brain Hlth. / UBC, Vancouver, BC, Canada; [4]Brian MacVicar, Djavad Mowafaghian Ctr. For Brain Hlth., Vancouver, BC, Canada; [5]Univ. of Victoria, Victoria, BC, Canada

Abstract: Background: Frontotemporal dementia (FTD) refers to a group of neurodegenerative disorders characterized by pathology predominantly localized to the frontal and temporal lobes. Approximately 40% of FTD cases are familial, and 25% of these are caused by heterozygous loss of function mutations in the gene encoding for progranulin, *GRN*. Research has attempted to explain the mechanisms for how loss of progranulin leads to FTD, but an entire picture remains unclear. Recent findings suggest mutations in *MAPT* - another leading cause of familial FTD - greatly alters astrocyte gene expression which leads to subsequent non-cell autonomous effects on neurons. In this study, we aimed to determine how *GRN* mutant astrocytes affect neurons in human-derived neural tissue models.**Methods:** To model *GRN* FTD we utilized human induced pluripotent stem cell derived neural tissue carrying homozygous *GRN* R493X-/- knock-in mutation and corresponding wildtype control tissue. Differentiated neuronal cells were co-cultured with wildtype or R493X$^{-/-}$ astrocytes and excitatory electrical development was investigated using microelectrode array analysis. Immunocytochemistry was used to examine synaptic expression of GABAergic and glutaminergic synaptic markers. **Results:** We demonstrated that *GRN* R493X-/- astrocytes impact neuron maturation by significantly delaying excitatory electrical development. Histologically, neurons during the delay showed a decrease in GABAergic synaptic markers while also showing an increase in glutaminergic synaptic markers.

We also demonstrate that this effect is due in-part due to soluble factors.**Conclusions:** Overall, this work represents the first study investigating astrocyte-induced neuronal pathology in *GRN* mutant hiPSCs and supports the hypothesis of astrocyte involvement in the progression of FTD.

Disclosures: C. Lee: None. **J. Frew:** None. **N.L. Weilinger:** None. **S. Wendt:** None. **W. Cai:** None. **S. Sorrentino:** None. **X. Wu:** None. **B.A. MacVicar:** None. **S. Willerth:** None. **H.B. Nygaard:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.12

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

Support: NIH R00 AG056597

Title: Interactions between progranulin insufficiency and TDP-43 in a mouse model of frontotemporal dementia

Authors: *A. K. COOK, S. E. DAVIS, D. A. SAVOSTIKOVA, G. A. VOLLMER, A. R. HAKIM, A. E. ARRANT;
Univ. of Alabama, Birmingham, Univ. of Alabama, Birmingham, Birmingham, AL

Abstract: Loss of function mutations in progranulin (*GRN*) are a major cause of dominantly-inherited frontotemporal dementia (FTD), most commonly causing behavior changes such as apathy, disinhibition, compulsivity, and social withdrawal. Most pathogenic mutations in *GRN* result in haploinsufficiency, which likely drives the development of FTD. *GRN* mutations cause FTD with TDP-43 pathology, characterized by TDP-43 inclusions in the nucleus and cytoplasm, as well as TDP-43 mislocalization to the cytoplasm. $Grn^{+/-}$ mice provide a model of progranulin insufficiency and demonstrate age-dependent behavioral abnormalities. However, $Grn^{+/-}$ mice fail to develop TDP-43 pathology. Homozygous $Grn^{-/-}$ mice develop TDP-43 aggregates in the thalamus and pons at advanced ages, but model the complete progranulin deficiency that causes Neuronal Ceroid Lipofuscinosis in humans. To investigate TDP-43 in the context of progranulin insufficiency, we crossed $Grn^{+/-}$ mice with a mild TDP-43 transgenic line (Jackson Lab #012836). This TDP-43 transgenic line expresses moderate levels of human wild type TDP-43 via the Thy-1 promoter. Hemizygous TDP-43 transgenic mice develop mild motor deficits and gliosis, but do not develop TDP-43 pathology. Tests for motor function revealed mild TDP-related motor defects in wire hang and pole tests that were not worsened by *Grn* genotype. However, no motor abnormalities were observed in rotarod or open field. $Grn^{+/-}$ mice develop deficits in sociability and social dominance, however these deficits were more severe and had an earlier onset in the $Grn^{+/-}$:hTDP+ mice. Interestingly, $Grn^{+/+}$:hTDP+ mice exhibited no social abnormalities in these tests. $Grn^{+/-}$:hTDP+ mice show elevated levels of RIPA-insoluble TDP-43 in the frontal cortex, a region important for sociability and social dominance behaviors.

However, TDP overexpression did not worsen lysosomal abnormalities or induce lipofuscinosis or gliosis in the *Grn*$^{+/-}$ mice. Our data shows that progranulin insufficiency interacts with TDP-43 overexpression to worsen behavioral phenotypes in mice. Ongoing studies are investigating if progranulin insufficiency induces TDP-43 mislocalization or aggregation.

Disclosures: A.K. Cook: None. **S.E. Davis:** None. **D.A. Savostikova:** None. **G.A. Vollmer:** None. **A.R. Hakim:** None. **A.E. Arrant:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.13

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

Support: NIH Grant NS108115
Brightfocus Foundation
Consortium for Frontotemporal Dementia Research (CFR)/Bluefield Project to Cure FTD

Title: Small molecule correction of the progranulin haploinsufficient lysosomal proteome

Authors: *G. WERTHMANN[1], J. HERZ[2];
[1]Univ. of Texas, Southwestern Med. Ctr., Dallas, TX; [2]UT Southwestern, Dallas, TX

Abstract: Progranulin is a key regulator of lysosomal function. Loss of one copy of *GRN* (the gene which encodes progranulin) leads to frontotemporal dementia while loss of both copies leads to the lysosomal storage disorder neuronal ceroid lipofuscinosis 11. How progranulin alters lysosomal function and composition is still unknown. In this study, we analyzed *Grn*$^{+/+}$, *Grn*$^{+/-}$, and *Grn*$^{-/-}$ endo-lysosomal proteomes using a novel technique called Lyso-IP followed by liquid chromatography/mass spectroscopy. We found several downregulated proteins in both *Grn*$^{+/-}$ and *Grn*$^{-/-}$ endo-lysosomes that are associated with key lysosomal functions such as autophagy, lysosomal trafficking, and catabolism. Proteins mutated in various lysosomal storage disorders are also downregulated in both *Grn*$^{+/-}$ and *Grn*$^{-/-}$ endo-lysosomes. These changes to the endo-lysosomal proteome were not reflected in either the *Grn* mutant transcriptome or whole-cell proteome, suggesting that progranulin functions on a post-transcriptional level to regulate lysosomal protein content. Finally, we used this robust proteomic phenotype to test the efficacy of the progranulin boosting histone deacetylase (HDAC) vorinostat (a.k.a. SAHA). We found SAHA administration improved several of the dysregulated lysosomal proteins in *Grn*$^{+/-}$ endo-lysosomes, demonstrating both the potential of this assay to test FTD-GRN therapies as well as the potential of progranulin boosting small molecules to treat FTD-GRN.

Disclosures: G. Werthmann: None. **J. Herz:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.14

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

Support: VA Grant

Title: Nad biosynthetic enzymes are recruited as chaperones to combat polyglutamine-induced proteotoxic stress

Authors: *M. PINKERTON[1], A. BARRIENTOS[2];
[1]Univ. of Miami, Univ. of Miami, Miami, FL; [2]Miller Sch. of Med., Miller Sch. of Med., Miami, FL

Abstract: NAD Biosynthetic Enzymes are Recruited as Chaperones to Combat Polyglutamine-induced proteotoxic Stress
Age-associated neurodegenerative proteinopathies, such as Huntington's disease, involve the misfolding and aggregation of disease-specific proteins in the brain. The exploration for a treatment identified nicotinamide/nicotinic acid salvage NAD^+ biosynthetic pathway enzyme NMNAT (nicotinamide mononucleotide (NMN) adenylyltransferase) as an effective suppressor of proteotoxicity. Although there has been some controversy regarding whether the neuroprotective effect of NMNAT is mediated by increased NAD^+ levels or a non-catalytic chaperone role of NMNAT, it is now accepted that both may contribute. Screens in yeast models of HD and PD in our lab have allowed us to identify not only the NMNAT homologs Nma1/2 but three additional enzymes of the NAD^+ salvage pathway that achieve similar protection against extended polyglutamine-induced proteotoxic stress: Npt1, Pnc1, and Qns1. Under proteotoxic stress, the four proteins are recruited to promote the clearance of misfolded and oligomerized proteins. We have shown that the suppression mechanism by NAD^+ proteins is independent of their catalytic activity with in vitro and in vivo studies indicating that these proteins have the ability to act as molecular chaperones. Our data suggest that they perform holdase and foldase chaperone activities to contribute to the reduction of misfolded toxic proteins, while promoting their refolding. In the case of Nma1, structure-function relationship studies have shown that the C-terminus of the protein is essential for its chaperone activity. We have now corroborated these data in neuronal models of HD. Preliminary data have shown that the human homologs NMNAT1-3, NADSynthetase, NAMPT, NAPRT, and PNP1 have chaperone activity. Fluorescence microscopy and cell viability assays have shown that these proteins prevent mutant huntingtin protein misfolding and protect against mutant huntingtin-induced cell death. Our data illustrates the existence of an evolutionarily conserved strategy of repurposing housekeeping enzymes under proteotoxic stress conditions in the yeast and human models.

Disclosures: M. Pinkerton: None. **A. Barrientos:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.15

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

Support: Swiss National Science Foundation. Grant Number: 310030_192650
Association of Frontotemporal Dementia
Stiftung Synapsis - Alzheimer Forschung Schweiz AFS (Stiftung Synapsis - AFS). Grant Numbers: 2020-CDA02
Stiftung Synapsis - Alzheimer Forschung Schweiz AFS (Stiftung Synapsis - AFS). Grant Numbers: 2019-CDA01

Title: Template dependent amplification of pathological TDP-43 and roles of phosphorylation

Authors: *P. DE ROSSI[1], A. J. LEWIS[2], J. FURRER[1], L. DE VOS[1], T. DEMETER[1], W. ZHONG[1], A. ZBINDEN[1], V. I. WIERSMA[1], C. SCIALO[1], T. LASHLEY[3], H. STAHLBERG[2], M. POLYMENIDOU[1];
[1]Dept. of Quantitative Biomedicine, Univ. of Zurich, Zurich, Switzerland; [2]Lab. of Biol. Electron Microscopy, EPFL, Lausanne, Switzerland; [3]Dept. of Neurodegenerative Dis., UCL Inst. of Neurol., London, United Kingdom

Abstract: TDP-43 is an RNA binding protein found aggregated in several neurodegenerative diseases such as frontotemporal lobar degeneration (FTLD), amyotrophic lateral sclerosis (ALS) and Limbic-predominant age-related TDP-43 encephalopathy (LATE). The pathological hallmarks of these diseases are characterized by the presence of hyperphosphorylated TDP-43 within these pathological aggregates. It is assumed that TDP-43 proteinopathies follow the prion-like cascade, but the molecular mechanisms remained unknown. In our study, we demonstrated that isolated pathological seeds from post mortem tissue of patient with FTLD-TDP could trigger *de novo* aggregation in cells in a template-dependent manner. Our results also suggested that phosphorylation of these neoaggregates was sequential, N- to C-terminal, with subtype-specific timelines and aggregation profiles. We are currently investigating the role of TDP-43 phosphorylation in neuronal function, as well as disease pathogenesis and progression.

Disclosures: P. De Rossi: None. **A.J. Lewis:** None. **J. Furrer:** None. **L. De Vos:** None. **T. Demeter:** None. **W. Zhong:** None. **A. Zbinden:** None. **V.I. Wiersma:** None. **C. Scialo:** None. **T. Lashley:** None. **H. Stahlberg:** None. **M. Polymenidou:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.16

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

Title: Ionomycin and hydrogen peroxide induce neurite degeneration via ER and mitochondrial dysfunction

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

Abstract: Low concentrations of hydrogen peroxide induce axonal degeneration prior to neuronal cell death, depending on concentration- and time-dependent manner. The phenomenon of neurite degeneration has also been found in the early stage of human Alzheimer's disease (AD) brain and other neurodegenerative disorders. However, the detailed mechanisms of the onset of neurite degeneration have not been fully elucidated. One possible mechanism is a decrease in intracellular calcium homeostasis. Here we attempted to clarify the relationship between mitochondrial and endoplasmic reticulum (ER) dysfunction in reactive oxygen species (ROS) and calcium ionophore, ionomycin-induced neurite degeneration. N1E-115 cells with elongated neurites were used to elucidate the mechanism of neurite degeneration in hydrogen peroxide or ionomycin. Treatment with both reagents induced neurite degeneration, including beading formation. The fluorescent emissions of Fluoo-4AM and Mito-SOX were dramatically increased in both materials-treated samples compared to the controls. Especially, membrane oxidation was induced after treatment with hydrogen peroxide. On electron microscopic analysis, mitochondria and ER were in the area of neurite degeneration, and the mitochondrial appeared to be swollen. Mitochondria and ER are famous calcium stores in the cells, co-treatment with calcium channel inhibitor significantly was decrease Fluo-4 AM emission. These results indicate that ROS- and calcium ionophore-induced neurite degeneration may relate to the collapse of mitochondria- and ER-related calcium homeostasis.

Disclosures: **K. Fukui:** None. **K. Houga:** None. **M. Wakuzawa:** None. **Y. Kato:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.17

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

Support: AMED to SM (19dm0107103)
AMED to NM (JP21ek0109486, JP21ek0109549, JP21cm0106503 and JP21ek0109493)
MEXT of Japan to NN (17H01564)
MEXT of Japan to TY (17KT0131)
MEXT of Japan to AF (JP20K17936)
MEXT of Japan to SM (16H06277)

Title: Hornerin deposits in neuronal intranuclear inclusion disease: direct identification of proteins with compositionally biased regions in inclusions

Authors: *H. PARK[1,2], T. YAMANAKA[4,2], Y. TOYAMA[3], A. FUJITA[5], H. DOI[6], T. NIRASAWA[7], S. MURAYAMA[8], N. MATSUMOTO[5], T. SHIMOGORI[9], M. IKEGAWA[3], M. J. HALTIA[10], N. NUKINA[2,11];
[1]Inst. of Mol. Biol. and Genet., Seoul Natl. Univ., Seoul, Korea, Republic of; [2]Grad. Sch. of Brain Sci., [3]Dept. of Life and Med. Systems, Doshisha Univ., Kyoto, Japan; [4]Dept. of Neurosci. of Disease, Brain Res. Inst., Niigata Univ., Niigata, Japan; [5]Dept. of Human Genet., [6]Dept. of Neurol. and Stroke Med., Yokohama City Univ. Grad. Sch. of Med., Yokohama, Japan; [7]Bruker Japan K.K., Yokohama, Japan; [8]The Brain Bank for Aging Res., Tokyo Metropolitan Geriatric Hosp. & Inst. of Gerontol, Tokyo, Japan; [9]Mol. Mechanisms of Brain Develop., RIKEN Ctr. for Brain Sci., Saitama, Japan; [10]Dept. of Pathology, Fac. of Med., Univ. of Helsinki, Helsinki, Finland; [11]Juntendo Univ., Tokyo, Japan

Abstract: Neuronal intranuclear inclusion disease (NIID) is a neurodegenerative disorder, characterized by the presence of eosinophilic inclusions (NIIs) within nuclei of central and peripheral nervous system cells. This study aims to identify the components of NIIs, which have been difficult to analyze directly due to their insolubility. In order to establish a method to directly identify the components of NIIs, we first analyzed the huntingtin inclusion-rich fraction obtained from the brains of Huntington disease model mice. Although the sequence with expanded polyglutamine could not be identified by liquid-chromatography mass spectrometry, amino acid analysis revealed that glutamine of the huntingtin inclusion-rich fraction increased significantly. This is compatible with the calculated amino acid content of the transgene product. Therefore, we applied this method to analyze the NIIs of diseased human brains, which may have proteins with compositionally biased regions, and identified a serine-rich protein called hornerin. Since the analyzed NII-rich fraction was also serine-rich, we suggested hornerin as a major component of the NIIs (Fig. 1 and Fig. 2). A specific distribution of hornerin in NIID was also investigated by Matrix-assisted laser desorption/ionization imaging mass spectrometry (Fig. 3) and immunofluorescence. Finally, we confirmed a variant of hornerin by whole-exome sequencing and DNA sequencing. This study suggests that hornerin may be related to the pathological process of this NIID, and the direct analysis of NIIs, especially by amino acid analysis using the NII-rich fractions, would contribute to a deeper understanding of the disease pathogenesis.

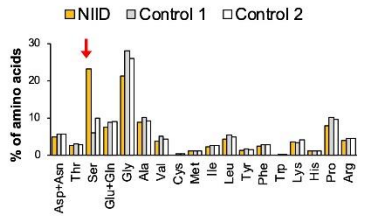


Fig 1. Serine increased in the NIIs of NIID

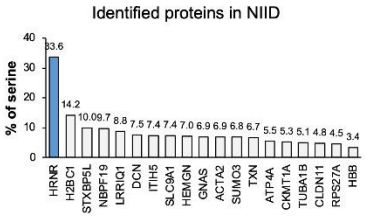


Fig 2. Hornerin was rich in Serine

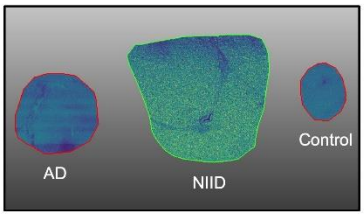


Fig 3. The specific distribution of hornerin in NIID observed by MALDI-IMS

Disclosures: **H. Park:** None. **T. Yamanaka:** None. **Y. Toyama:** None. **A. Fujita:** None. **H. Doi:** None. **T. Nirasawa:** None. **S. Murayama:** None. **N. Matsumoto:** None. **T. Shimogori:** None. **M. Ikegawa:** None. **M.J. Haltia:** None. **N. Nukina:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.18

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

Support: T32 NS082168
P50AG047266
R01NS089022
R01NS100876

Title: Effect of Amyloid β Deposition on the Proteolytic Processing of α-synuclein

Authors: *G. LLOYD, B. GIASSON, D. BORCHELT;
Univ. of Florida, Gainesville, FL

Abstract: Neurodegenerative diseases (NDDs) are a spectrum of disorders characterized by the the progressive dysfunction and eventual loss of neurons, and are histologically hallmarked by the accretion of misfolded proteins into stacked β-sheets to form insoluble structures called amyloid fibrils. Alzheimer's disease (AD) is the most common form of age-related NDD and the most common cause of dementia. Parkinson's disease (PD) is the second most common NDD, and the most prevalent movement disorder. Despite initially being considered separate and distinct clinicopathologies, it is now understood that features of AD and PD overlap. The overlap of clinical presentation can make an accurate diagnosis difficult to discern, with some symptoms having a wide range of potential underlying pathologies. This complicates treatment strategies and jeopardizes disease-specific therapeutic research as currently, confirmed diagnosis of the disease can only be established upon autopsy. Therefore, it is critical that the underlying distinctions of pathophysiology between these disorders are further elucidated and correlated with clinical presentation. This will allow for increased accuracy in diagnostics and improved precision in future biomarkers. We have previously generated a novel mouse model by crossing APPswe/PS1dE9 (L85) mice, which develop amyloid β (Aβ) deposition by 4-6 months of age, with mice expressing WT human αSyn (M20), which develop extensive α-synuclein (αSyn) pathology following intracerebral injection of αSyn preformed fibrils (PFFs) and found that Aβ deposition dramatically potentiates αSyn pathological spread and exponentially accelerates glial interactivity throughout the neuroaxis. In this work, we further probe the consequences of co-pathology by examining the conformational variety of αSyn in order to elucidate potential mechanisms for the observed potentiated spread. Using antibodies targeted for C-terminally truncated αSyn, we have found a stark variation between mice with co-deposition of Aβ and αSyn compared to mice with αSyn inclusion pathology alone. Overall, our work provides mechanistic insight into αSyn processing in the presence of Aβ pathology and provides clues for how one protein may lead to the further dysfunction of another.

Disclosures: G. Lloyd: None. **B. Giasson:** None. **D. Borchelt:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.19

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

Support: EP/N509577/1

Title: Progranulin deficiency causes impairments in PINK1/Parkin mediated mitophagy

Authors: *J. M. CASEY[1], D. MELANDRI[1], C. ARBER[1], B. COSTA[1], M. SOUTAR[1], B. O' CALLAGHAN[1], C. HOLLER[5], T. KUKAR[5], J. POCOCK[2], A. ISAACS[3], J. ROHRER[4], H. PLUN-FAVREAU[1], S. WRAY[1];
[1]Dept. of Neurodegenerative Dis., [2]Dept. of Neuroinflam., [3]UK Dementia Res. Inst., [4]Dementia Res. Ctr., UCL Queen Square Inst. of Neurol., London, United Kingdom; [5]Dept. of Pharmacol. and Chem. Biol., Emory University, Sch. of Med., Atlanta, GA

Abstract: Introduction: We examined mitophagy in frontotemporal dementia (FTD) caused by heterozygous *GRN* mutations, which result in haploinsufficiency of the progranulin protein. Impairments in mitophagy, the selective autophagy of damaged mitochondria, have been identified in a number of neurodegenerative diseases, with a number of FTD genes known to play a role in mitophagy (e.g, *TBK1* and *OPTN*). Reduced xenophagy, the selective clearance of non-host pathogens, has been identified in $GRN^{-/-}$ mice and is reliant on some proteins involved in mitophagy (TBK1 and Parkin). We therefore hypothesised that progranulin might affect mitophagy.
Methodology: We examined PINK1/Parkin mitophagy in neuroblastoma Parkin overexpressing SHSY5Y cells (POE-5Ys) and neuroglioma H4 cells +/- *GRN* siRNA. We also investigated induced pluripotent stem cells (iPSCs) differentiated to cortical neurons, astrocytes and microglia from 4 controls, 3 *GRN* FTD patient and an isogenic R493X *GRN* mutation CRISPR series (control, heterozygous and homozygous) from the human iPSC Neurodegenerative Disease Initiative (iNDI). Mitophagy was induced using oligomycin and antimycin A (O/A). PINK1 accumulation, levels of S65 phosphorylated ubiquitin (pUb) and other proteins involved in PINK1/Parkin mitophagy were examined using western blotting and immunofluorescence (ICC).
Results: Lower levels of pUb were detected in POE-5Ys following *GRN* knockdown and O/A treatment in a high throughput imaging screen. We also detected a significant reduction in mitophagy in *GRN* siRNA treated H4 cells by ICC and western blotting of mitophagy markers. There was no significant difference in mitophagy between control and patient iPSC-derived neurons, but there was variability in mitophagy and progranulin levels between inductions. We found a significant reduction in mitophagy in the homozygous R493X mutation neurons. There was no significant difference in mitophagy in the heterozygous line, potentially due to compensatory upregulation of progranulin protein levels from the wild type allele. CDC37, which traffics PINK1 to the mitochondria and affects its stability at the mitochondria, was significantly reduced in both H4 cells treated with *GRN* siRNA and homozygous R493X

mutation neurons.
Conclusions: We have shown that loss of progranulin leads to impairments in PINK1/Parkin mitophagy. Current work aims to further understand the mechanisms of this process. Work is also ongoing in iPSC derived astrocytes and microglia to dissect cell-type specific contributions of progranulin to mitophagy.

Disclosures: **J.M. Casey:** None. **D. Melandri:** None. **C. Arber:** None. **B. Costa:** None. **M. Soutar:** None. **B. O' Callaghan:** None. **C. Holler:** None. **T. Kukar:** None. **J. Pocock:** None. **A. Isaacs:** None. **J. Rohrer:** None. **H. Plun-Favreau:** None. **S. Wray:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.20

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

Support: NINDS RO1 NS097542
NINDS RO1 NS113943
DoD W81XWH2110182

Title: Regulation and toxicity of truncated TDP43 isoforms in ALS and FTD

Authors: *M. DYKSTRA[1], K. WESKAMP[2], N. GOMEZ[2], J. MORAN[2], N. GROTEWOLD[2], J. MCBRIDE[2], S. BARMADA[2];
[1]Neurol., Univ. of Michigan Neurosci. Grad. Program, Ann Arbor, MI; [2]Univ. of Michigan, Ann Arbor, MI

Abstract: TDP43 (TAR DNA/RNA binding protein, 43 kDa) is a critical nuclear RNA binding protein involved in several aspects of RNA metabolism. In nearly all individuals with amyotrophic lateral sclerosis (ALS), and most of those with frontotemporal dementia (FTD), TDP43 is mislocalized to the cytoplasm, abnormally phosphorylated and ubiquitinated. Even so, we understand little about the underlying reasons for TDP43 mislocalization, or the impact of TDP43 cytoplasmic deposition and/or loss of nuclear TDP43 on neuronal survival. Our previous studies suggested that TDP43 mislocalization may be due to the production of alternatively spliced and truncated TDP43 isoforms that are prone to aggregation and actively exported from the nucleus. These 'short' (s)TDP43 isoforms are evolutionarily conserved, but their regulation and function remain fundamentally unclear. Here, we show that sTDP43 is produced by the same negative feedback loop that regulates TDP43 levels in all cells, wherein TDP43 binds to its own RNA, resulting in alternatively spliced transcripts that are destabilized by nonsense mediated RNA decay (NMD). We found that the alternatively spliced transcripts generated in the process of TDP43 autoregulation are those that encode sTDP43. Consistent with this, TDP43 overexpression and NMD inhibition both increase sTDP43 production. To validate these results and also identify additional splicing factors involved in the generation of sTDP43, we created an

sTDP43-specific splicing reporter which we are adapting for use in high-throughput and unbiased screens in human neurons. In primary rodent neurons, sTDP43 overexpression recapitulates nuclear TDP43 exclusion and cytoplasmic TDP43 aggregation, together with cellular toxicity. sTDP43-dependent toxicity requires functional RNA binding domains as well as residues that interact with endogenous TDP43, implying both gain-of-function and loss-of-function mechanisms contributing to neurodegeneration in ALS and FTD. In ongoing studies, we are testing approaches capable of selectively reducing or eliminating sTDP43, while leaving full-length TDP43 unaffected. Together, these investigations may prove essential for elucidating the function of sTDP43, as well as the consequences of sTDP43 accumulation in ALS and FTD.

Disclosures: M. Dykstra: None. **K. Weskamp:** None. **N. Gomez:** None. **J. Moran:** None. **N. Grotewold:** None. **J. McBride:** None. **S. Barmada:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.21

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

Title: Study of the protective effect of the extract of Hedysarum alpinum L in vitro

Authors: *A. JALSRAI[1], A. BECKER[2], H. HSEIH-LI[3], L. SANZHIEVA[4], Y. CONG[5], U. MYADAGBADAM[6];
[1]Inst. of Traditional Med. and Technol., Ctr. of Traditional Med., Ulaanbaatar, Mongolia; [2]5Institute of Pharmacol. and Toxicology, Fac. of Medicine,, Otto von Guericke Univ., Magdeburg, Germany; [3]Inst. of Pharmacol. and Toxicology, Fac. of Med., Natl. Taiwan Normal Univ., Taiwan, Taiwan; [4]Dept. of Physiol., Sanzhieva L.Ts2, Buryat State University, Russian Federation; [5]Pharmaceut. Col., Henan Univ., Kaifeng city, China; [6]Inst. of Traditional Med. and Technol., Ulaanbaatar, Mongolia

Abstract: *Hedysarum* species have a variety of uses in traditional medicine, including the treatment of individuals suffering from brain injury or dementia. *Hedysarum alpinum L* is believed to support the immune system and the peripheral nervous system. However, the biological activities of *Hedysarum alpinum L* are not well studied. In current work, we conducted phytochemical screenings and investigated the protective effects of an extract of Hedysarum alpinum L.**Material and methods:** The Aerial part of *Hedysarum alpinum L* was collected from Bulgan Aimagk in western Mongolia. An extract of the specimens preserved in 70% ethanol was filtered and dried under vacuum for use in the study. Established conventional methods were used for quantitative determination of total phenols and flavonoid content. Hydroxyl radicals and lipid radicals were detected using spin trapping agents with ESR spectroscopy. The protective effects of the extract were assessed using an in vitro insult assay, i.e. mitochondrial dysfunction induced by malondialdehyde (MDA 100µM).**Results:** Phytochemical screening for the ethanolic extract of *Hedysarum alpinum L* detected polyphenols including quercetin (0.61%), rutin

(0.93%), gallic acid (0,89%). Hedysarum aplinum L extract exhibited dose dependent scavenging effects on hydroxyl radicals and lipid radicals. The half maximal inhibitory concentration (IC_{50}) of the extract was 6.72 mg/ml and 8.37 mg/ml for hydroxyl and lipid radicals respectively. These results demonstrate that the extract of *Hedysarum alpinum L* effectively inhibited free radicals. We found that the ethanolic extract of *Hedysarum alpinum L* provided significant protection against malondialdehyde-induced oxidative damage by improving the activity of mitochondrial respiratory transport chains (complex I and complex II) at a concentration 32 mg/ml in isolated brain mitochondria exhibiting MDA oxidative damage. **Conclusion:** These results provide an insight into a possible mechanism by which the extract of *Hedysarum alpinum L* could protect against oxidative damage in cells.

Disclosures: A. Jalsrai: None. **A. Becker:** None. **H. Hseih-Li:** None. **L. Sanzhieva:** None. **Y. Cong:** None. **U. Myadagbadam:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.22

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

Support: Ministry of Science and ICT (21-BR-02-08)

Title: Phosphoproteomics of Neuro-2a cells under impaired insulin sensitivity provides new targets for Alzheimer’s disease.

Authors: *Y. JO[1,2], D. KIM[3], H.-S. JO[1], S. BAE[1], Y. KWON[1], Y.-S. OH[2], J. YOON[1];
[1]Neurodegenerative Dis. Res. Group, Korea Brain Res. Institute(KBRI), Deagu, Korea, Republic of; [2]Brain-Cognitive Sci., Daegu-Gyeongbuk Inst. of Sci. and Technol. (DGIST), Deagu, Korea, Republic of; [3]New Drug Develop. Ctr., Daegu-Gyeongbuk Med. Innovation Foundatio0n(K-MEDI hub), Deagu, Korea, Republic of

Abstract: Insulin is a well-known critical factor in brain development and the control of neurogenesis, in-cluding in the hippocampus. The alteration of insulin signaling in the brain can induce brain aging and regulate brain plasticity and could promote neurodegeneration in the late stage of Alzheimer’s disease (AD). The precise molecular mechanism of the relationship between insulin resistance and AD remains unclear. The development of phosphoproteomics has advanced our knowledge of phosphorylation-mediated signaling networks and could elucidate the molecular mechanisms of certain conditions. Here, we applied a reliable phosphoproteomic approach to Neuro-2a (N2a) cells to identify their molecular features under two different clinically reliable insulin-resistant conditions: inflammation and dyslipidemia. We found different informatic characteristics between the two insulin-resistant phosphoproteomes by comparative informatics analysis. We also found commonly changed molecular signatures, including phosphoproteins, in the

integrin and adenosine monophosphate-activated protein kinase pathways under insulin resistance and verified these targets by subsequent biochemical experiments. Among the commonly changed molecular signatures, the phosphorylation of acetyl-CoA carboxylase and Src was also found to be altered in the brains of 5xFAD mice. This study provides new molecular signatures for insulin resistance in N2a cells and possible links between the molecular features of insulin resistance and AD.

Disclosures: Y. Jo: None. **D. Kim:** None. **H. Jo:** None. **S. Bae:** None. **Y. Kwon:** None. **Y. Oh:** None. **J. Yoon:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.23

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

Support: NIH Grant GM127513
Ara Parseghian medical research fund

Title: Npc1-dependent alterations in ca_v1-$k_v2.1$ nanodomains drives neurodegeneration

Authors: *M. CASAS[1], K. HINO[2], N. VIERRA[1], S. SIMÓ[2], J. TRIMMER[1], R. DIXON[1], E. DICKSON[1];
[1]Physiol. and Membrane Biol., [2]Cell Biol. and Human Anat., Univ. of California, Davis, CA

Abstract: Lysosome dysfunction is associated with the pathogenesis of a variety of neurodegenerative disorders. Thus, mechanisms that link lysosome dysfunction to disruption of neuronal homeostasis offer opportunities to understand the molecular underpinnings of neurodegeneration and to potentially identify novel therapeutic targets. A key mechanism through which lysosomes communicate and receive instruction is via transfer of cholesterol at ER-lysosome MCS. At these contacts, the Niemann Pick C1 cholesterol transporter (NPC1) facilitates the efflux of cholesterol out of the lysosome before it is transferred to the ER for distribution to other cellular membranes. Underscoring its importance, loss of function mutations in NPC1 lead to the progressive neurodegenerative disorder, NPC disease. This fatal condition has no cure and is characterized by the progressive neurodegeneration of several brain regions and a host of devastating symptoms including seizures, psychiatric problems, and dementia. Despite a general calcium handling defect being reported, the molecular mechanism(s) linking loss of NPC1 function to NPC disease associated neuropathology are unknown. Given the pathophysiological importance of regulated lysosomal cholesterol transport at ER-lysosome MCS, we wanted to understand if the molecular contents of other ER MCS are altered and contribute to neuron toxicity in NPC disease. In neurons, a prominent ER-PM contact site forming protein is the $K_V2.1$ ion channel, whose interactions with the ER-localized VAP proteins generate contact sites to tune lipid and calcium nanodomains. Using a combination of super-

resolution TIRF nanoscopy, high-speed fluorescence imaging, biochemistry, and animal models of NPC disease, we determined that loss of NPC1 function increases the area, density, and number of PM $K_V2.1$ channels. Furthermore, interactions between $K_V2.1$ and Ca_V1 voltage-gated Ca^{2+} channels are elevated and result in significantly increased Ca_V1 clustering and activity, leading to elevated Ca^{2+} entry into neurons. A major consequence of increased $K_V2.1$-supported Ca_V1 Ca^{2+} entry is elevated mitochondrial calcium concentrations which promotes neurotoxicity. Importantly, disrupting $K_V2.1$-Ca_V1 interactions using a synthetic peptide or reducing $K_V2.1$ phosphorylation with a Cyclin-dependent kinase 5 inhibitor rescues the enhanced Ca_V1 clustering, mitochondrial Ca^{2+}, and neurotoxicity seen in NPC neurons. Collectively, these data demonstrate that NPC is a nanostructural ion channel clustering disease with altered ion channel distribution/activity at ER-PM contacts contributing to neurodegeneration.

Disclosures: M. Casas: None. **K. Hino:** None. **N. Vierra:** None. **S. Simó:** None. **J. Trimmer:** None. **R. Dixon:** None. **E. Dickson:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.24

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

Support: NIH Grant NS108115
Consortium for Frontotemporal Dementia (CFR)
Blue Field Project to Cure FTD

Title: The interplay between solubility, absorbance, brain penetrance and activity in small molecule analogs that increase progranulin protein production in vivo.

Authors: *R. TESLA[1], C. GUHL[3], D. DIXON[1], J. FERRELL-PENNIMAN[1], N. WILLIAMS[2], J. READY[2], J. HERZ[1];
[1]Mol. Genet., [2]Biochem., Univ. of Texas, Southwestern, Dallas, TX; [3]Organic Chem. and Macromolecular Chem., Friedrich-Schiller-University, Jena, Germany

Abstract: Frontotemporal dementia (FTD) is the most common cause of pre-senile dementia, and the granulin (GRN) gene is one of several genes whose dysfunction can lead to a hereditary form of FTD. There is no approved treatment to delay onset, slow progression, or cure FTD, and the mechanisms involved in GRN induced FTD are not well understood. We chose to use high-throughput screening (HTS) to discover novel compounds which can induce progranulin (PGRN) protein production *in vitro* and *in vivo*. The goal of our small molecule study is to use these small molecules to elucidate the mechanisms underlying GRN triggered FTD and create a safe, long-term treatment for FTD related to GRN haploinsufficiency. A GRN luciferase reporter cell line was used to assess the effects of 200,000 small molecules on GRN activation. Following HTS and *in vitro* evaluation of top HTS hits, several compounds were dosed via continuous

intracerebroventricular infusion in C57BL/6J mice. Three compounds (C40, C41 and C127) rescued brain PGRN protein levels in *Grn*$^{+/-}$ mice back to *Grn*$^{+/+}$ levels. Due to *in vitro* pharmacokinetic analysis, C41 was eliminated before *in vivo* evaluation. After IP dosing of CD1 mice, we used mass spectroscopy to determine the PKs of our compounds of interest. Parameters assessed for C40 vs. C127 were maximum concentration (C_{max}), terminal elimination half-life (Terminal T1/2), time to reach maximum concentration (T_{max}) and brain to plasma absorption (6246 vs 1112ng/g, 41 vs 58.75 minutes (min), 30 vs. 10 min, and 5.34:1 vs. 2.3:1 respectively). Unfortunately, solubility issues forced us to use an unusually dilute concentration of compound (10mg/kg) for the PK studies. We hypothesized that more soluble analogs of C40 would increase compound delivery and cause a rise in compound absorption. This increased absorption would ultimately increase compound penetrance into the brain. Hoping to solve compound solubility issues, medicinal chemistry was used to create 30 analogs of C40. Seven of the created analogs increased PGRN protein levels in several mouse and human cells lines, and were 3-5 times more soluble than C40. Two analogs, A21 and A41, were also able to increase PGRN levels in C57BL/6J mouse brains after IP delivery at 50mg/kg. Both analogs had PK evaluations conducted *in vivo* in CD1 mice with compound delivery 2.5 times that of C40. Surprisingly, A21 and A41's *in vivo* pharmacokinetic evaluation exhibited 30% and 38% reduction in overall compound absorption and 37% and 60% decrease in uptake into the brain respectively when compared to C40. We are currently evaluating how these differences in compound absorption translate into variations in PGRN protein production in the brain *in vivo*.

Disclosures: R. Tesla: None. **C. Guhl:** None. **D. Dixon:** None. **J. Ferrell-Penniman:** None. **N. Williams:** None. **J. Ready:** None. **J. Herz:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.25

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

Support: NIH Grant F30AG071114
NIH Grant T32NS095775
Bluefield Project to Cure Frontotemporal Dementia

Title: Abnormalities in sphingolipid degradation caused by progranulin deficiency

Authors: N. BOYLE, A. E. ARRANT, T. DUNN, E. D. ROBERSON;
Neurol., Univ. of Alabama at Birmingham, Birmingham, AL

Abstract: Frontotemporal dementia (FTD) is a leading cause of early-onset dementia with no disease-modifying therapies. Heterozygous loss of function progranulin (*GRN*) mutations, causing haploinsufficiency of the progranulin protein, are causative of FTD and are inherited in an autosomal-dominant fashion. Progranulin is a secreted and lysosome-resident protein with

several identified functions, acting as a growth factor, an immunomodulator, and a regulator of lysosomal function. Complete loss of progranulin results in lipofuscinosis and aberrant activities of several lysosomal enzymes indicating a critical role of progranulin in lysosomal function. Despite the clear link between progranulin deficiency and lysosomal dysfunction, the role of progranulin in lysosomal function remains unclear. Recent reports have implicated abnormalities in glycosphingolipid (GSL) degradation caused by progranulin deficiency. Our lab and others have found a deficiency of β-glucocerebrosidase (GCase), which cleaves glucosylceramide into ceramide in the GSL degradation pathway, in both progranulin-deficient models and patients with FTD caused by progranulin mutations (FTD-*GRN*). We have found that enzymes upstream in the GSL degradation pathway have elevated activities accompanied by transcriptional upregulation of the enzymes. GCase, downstream of these enzymes, appeared to be a unique case, as it had deficient activity with no transcriptional up- or downregulation, suggesting that the elevations upstream may reflect a compensatory mechanism due to overall lysosomal dysfunction. The association between GSL abnormalities and neurological abnormalities is increasingly clear; mutations in several of the enzymes in the GSL pathway are causative for multiple neurodegenerative diseases, and GCase-associated SNPs are a leading risk factor for Parkinson Disease. Progranulin interacts with several of these enzymes but does not directly modify their activities, unlike the related protein prosaposin. We predicted that GCase was not uniquely deficient as a result of progranulin deficiency, and characterized other enzymes involved in ceramide production. Here, we report deficiency of other enzymes in this pathway, also without transcriptional changes. This discovery suggests that progranulin may play a much broader role in regulating sphingolipid degradation, and that further investigation is needed to understand the effects of progranulin-regulated enzymes to determine the mechanism by which progranulin deficiency causes FTD.

Disclosures: **N. Boyle:** None. **A.E. Arrant:** None. **T. Dunn:** None. **E.D. Roberson:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Owner of IP related to tau. F. Consulting Fees (e.g., advisory boards); Consulting for Lilly, AGTC.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.26

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

Support: Amsterdam Neuroscience Alliantieproject CTG-2019-05
NOMIS Foundation
JPND - Risk and Modifying factors for Fronto-Temporal Dementia (RiMod-FTD)

Title: Development of a human proteomics frontotemporal dementia disease framework. Using protein profiles of hereditary frontotemporal dementia to explore sporadic disease cases

Authors: *S. S. M. MIEDEMA[1], M. O. MOL[2], S. C. M. DE BOER[3], A. A. DIJKSTRA[3], F. T. W. KOOPMANS[1], D. C. HONDIUS[1], P. VAN NIEROP[1], K. MENDEN[5], C. F. DE VEIJ MESTDAGH[1], J. VAN ROOIJ[2], A. B. GANZ[1], I. PALIUKHOVICH[1], C. BAKALI[1], S. MELHEM[2], K. W. LI[1], H. HOLSTEGE[3], P. RIZZU[5], R. E. VAN KESTEREN[1], W. D. J. VAN DE BERG[4], J. C. VAN SWIETEN[2], P. HEUTINK[5], Y. A. L. PIJNENBURG[3], A. B. SMIT[1]; [1]Dept. of Mol. and Cell. Neurobio., Vrije Univ. Amsterdam, Amsterdam, Netherlands; [2]Dept. of Neurol., Erasmus Med. Ctr., Rotterdam, Netherlands; [3]Alzheimer Ctr. Amsterdam, [4]Dept. of Neurosci., Amsterdam UMC location VUmc, Amsterdam, Netherlands; [5]German Ctr. for Neurodegenerative Dis. (DZNE)-Tübingen, Tübingen, Germany

Abstract: Frontotemporal dementia (FTD) is a major cause of a lethal early-onset dementia that affects personality, behaviour, and language. Finding a solution for FTD is hampered by the considerable heterogeneity of the disorder. FTD is hereditary in up to 30% of cases, mainly induced by mutations in the C9orf72 (FTD-C9), progranulin (FTD-GRN), and microtubule associated protein tau (FTD-MAPT) genes. The remaining 70% (sporadic FTD) can be conceived as a complex trait disorder. Our research aims to determine key cell types and disease mechanisms for hereditary FTD and to construct a disease framework to enable profiling of sporadic FTD cases. We investigated the proteome of post-mortem frontal and temporal cortex from FTD-C9 (n = 16), FTD-GRN (n = 9) and FTD-MAPT (n = 13) cases and compared them with non-demented controls (NDCs; n = 11) using a data-independent mass spectrometry-based quantitative approach. Differential abundance analysis (FDR-corrected at $q < 0.05$) revealed brain-area specific protein signatures, with minor regulation in the frontal cortex for FTD-C9 (n = 41), and major regulation in the frontal cortex for FTD-GRN (n = 579) and in the temporal cortex for FTD-MAPT (n = 488). Subsequent gene ontology analysis of these partially overlapping profiles indicated the presence of distinct genetic subtype-specific disease mechanisms. Using scRNAseq data resources we deduced the involvement of major brain cell types in these distinct biological processes. For the FTD-GRN subtype, we observed a role for immune processes related to endothelial cells and for mitochondrial dysregulation in neurons. For the FTD-MAPT subtype, we observed involvement of dysregulated RNA processing, oligodendrocyte dysfunction, and axonal impairments. Comparison of FTD-MAPT with Alzheimer's disease proteomic data indicated that alterations in RNA processing and oligodendrocyte function are specific to FTD-MAPT. Our results indicate the distinct involvement of different brain cell types and biological mechanisms in genetic FTD subtypes. Proteomic profiling of sporadic FTD cases is currently ongoing. We are investigating multiple affected brain regions (i.e. frontal and temporal cortex, frontal insula, and anterior cingulate) from both hemispheres to generate an extensive overview. The inclusion of a healthy region (i.e. occipital cortex) per case will generate within-patient controls. Proteomic data will be supplemented with scRNAseq to generate in-depth insight into affected cell types. With our efforts we aim to reveal disease mechanisms for sporadic FTD within the wider FTD disease spectrum to pave the way for the development of disease subtype-specific treatment.

Disclosures: S.S.M. Miedema: None. **M.O. Mol:** None. **S.C.M. de Boer:** None. **A.A. Dijkstra:** None. **F.T.W. Koopmans:** None. **D.C. Hondius:** None. **P. van Nierop:** None. **K. Menden:** None. **C.F. de Veij Mestdagh:** None. **J. van Rooij:** None. **A.B. Ganz:** None. **I. Paliukhovich:** None. **C. Bakali:** None. **S. Melhem:** None. **K.W. Li:** None. **H. Holstege:** None. **P. Rizzu:** None. **R.E. van Kesteren:** None. **W.D.J. van de Berg:** None. **J.C. van Swieten:** None. **P. Heutink:** None. **Y.A.L. Pijnenburg:** None. **A.B. Smit:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.27

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

Support: NS093097
NS122351
NS125845

Title: Circuit mechanisms of impaired consolation behavior in a mouse model of behavioral variant frontotemporal dementia

Authors: *A. M. KOBEISSI[1], F.-B. GAO[2], W.-D. YAO[1];
[1]SUNY Upstate Med. Univ., Syracuse, NY; [2]Univ. of Massachusetts, Worcester, MA

Abstract: Frontotemporal dementia (FTD) is a leading cause of dementia and is a progressive, fatal disease characterized by neurodegeneration of the frontal and temporal lobes. There is no cure and current treatments are limited. There are several forms of FTD, including behavioral variant FTD (bvFTD), where marked changes in personality occur, including disinhibition, social deficits, and loss of empathy. The most common genetic cause of FTD is a G_4C_2 repeat expansion located in *C9orf72*. Several pathogenic mechanisms have been proposed; 1) loss of function due to haploinsufficient C9orf72 mRNA, 2) gain of function due to RNA foci, and 3) gain of function due to dipeptide repeat protein (DRP) aggregates generated by RAN translation of repeat RNA. The arginine containing DRPs, poly(GR) and poly(PR), have been shown to be especially toxic. However, the neural mechanisms and circuitry underlying bvFTD behavioral deficits are largely unknown. In this study, we used an inducible tetracycline-based system to express 80 repeats of poly(GR) in forebrain CaMKII-containing neurons (GR80) in an age-dependent manner to assess distress-induced, other-directed affiliative or comforting behavior in mice. Male and female C57BL/6J bystander mice display comforting behavior, characterized by allogrooming, body-contact, and cuddling, towards a distressed conspecific. However, aged male and female GR80 mice showed decreased comforting behavior compared to littermate controls. To assess the neural population and circuits involved in comforting behavior, we conducted an activity-dependent c-Fos mapping assay and identified several frontal regions activated during behaviors. Additionally, preliminary electrophysiology experiments revealed altered intrinsic and synaptic properties in the prefrontal cortex in mutant mice. Ongoing experiments aim to delineate the underlying mechanisms and rescue/ reverse loss of comforting behavior in mutant mice by targeting the neuropathological deficits in identified frontal circuits of these mice.

Disclosures: **A.M. Kobeissi:** None. **F. Gao:** None. **W. Yao:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.28

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

Support: NIH-NINDS F99NS125815

Title: Impact of race and sex on the cognitive progression of multiple sclerosis

Authors: *S. MOODY[1,2], M. MANUEL[3], B. COPELAND[4], J. LOVERA[4], D. DEVIER[5]; [1]Neurol. | Cell Biol. & Anat., LSU Hlth. Sci. Center-NO, New Orleans, LA; [2]Human Develop. and Family Studies, Iowa State Univ., Ames, IA; [3]Cell. Biol. and Anat., Med. Col. of Georgia at Augusta, Augusta, GA; [4]Neurol., Louisiana State Univ. Hlth. Sci. Ctr., New Orleans, LA; [5]LSU Hlth. Sci. Ctr. Cell Biol. & Anat., New Orleans, LA

Abstract: Worldwide, prevalence of multiple sclerosis (MS) has increased from 2.3 million in 2013 to 2.8 million in 2020. Incidence in diagnosis of MS differs by race and gender. White populations have historically had higher rates of diagnosis than Black populations yet, this trend has shifted such that Black Americans show increased incidences of MS diagnosis, worse disease severity, and earlier likelihood of mortality due to MS. Variation in diagnosis, clinical outcomes, and severity are also linked to biological sex. As with Black individuals, the characteristics of diagnosis in women has shifted towards greater increases in MS prevalence. To examine this shift in diagnosis and progression, we investigated if race and sex had an impact on cognitive decline in MS by using the Symbol Digit Modality Test (SDMT), Montreal Cognitive Assessment (MoCa), and the King Devick (KD) test. Ninety-three Black (N=36) and White (N=57) participants with MS were recruited from a neurology clinic in the gulf south of the United States. Black participants had overall shorter disease duration, (MYears= B: 8.6, W: 12.8), $F(1,91) = [16.66]$, $p < .01$) and were younger than White participants, (MAge= B: 40.1, W: 50.3), ($F(1, 90) = [4.21]$, $p = .04$) . Yet regression and ANOVA models revealed that even when considering age, sex and disease duration, Black participants had worse cognitive outcomes as measured by the SDMT ($R2 = .12$, $F(3, 88) = 3.92$, $p < .01$) and MoCA ($R2 = .11$, $F(3, 88) = 3.64$, $p = .01$) and were more likely to be cognitively impaired as measured by T-scores for the SDMT ($F(1, 91) = [6.06]$, $p = .02$) and KD ($F(1, 86) = [4.70]$, $p = .03$). This suggests that Black participants may have more aggressive disease courses leading to earlier cognitive decline.Men had a longer disease duration than women (M= M: 15.21:, W: 10.20) ($F(1,90) = [4.05]$, $p = .05$) but showed no differences in cognition. Specifically, Black women had a slightly lower disease duration than Black men (Myears= BM: 14.39, BW: 7.40), ($F(1, 33) = [3.71]$, $p = .06$) and a lower disease duration (Myears= BW: 7.40, WW: 12.00), ($F(1, 72) = [.4.59]$, $p = .04$) as well as age (Mage= BW: 40.13, WW: 49.20), ($F(1, 72) = [.4.59]$, $p = .04$) than White women. Yet, Black women still had worse scores as measured by the SDMT ($R2 = .09$, $F(2, 71) = 4.57$, $p = .014$) and MoCA ($R2 = .17$, $F(2, 71) = 8.57$, $p < .01$) and were more likely to be cognitively impaired as measured by T-scores on the KD ($F(1, 69) = [4.86]$, $p = .03$) when compared to White women. Our findings indicate both between and within racial group differences in MS. Specifically, Black women fared cognitively worse than Black men and especially White

women, hinting that an intersection between race and gender may impact the cognitive progression of MS.

Disclosures: S. Moody: None. **M. Manuel:** None. **B. Copeland:** None. **J. Lovera:** None. **D. Devier:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.29

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

Support: NIH/NIGMS-Pilot: 1P20GM103653-01A1

Title: Increased expression of the small heat shock protein 27 may reduce aggregation of transactive response DNA binding protein 43

Authors: *M. ABEER[1], A. K. GAUFF[1], M. LIMA[1], S. A. DAVIS[2], M. A. GITCHO[1]; [1]Delaware State Univ., Dover, DE; [2]Delaware Tech. Community Col., Dover, DE

Abstract: A major pathological protein in both amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTD) is Transactive response DNA binding protein 43 (TDP-43). Recently, pathological TDP-43 was discovered as a secondary pathology in up to 50% of Alzheimer's disease (AD) cases. Several studies reported that TDP-43 binds to heat shock protein family B (small) member 1 (HSPB1 or HSP27) but no functional evaluation of this interaction has been explored. In response to stress, heat shock proteins work to help fold native proteins in order to reduce aggregation. Inducing expression of HSP27 has been shown to be protective of many other disease conditions and has been shown to reduce aggregation of amyloid in AD. The overall goal of the current project is to utilize both primary neuronal cultures and mice that are selectively expressing pathogenic TDP-43, HSP27, and apolipoprotein E (apoE) in the brain and spinal cord in order to characterize the effect of HSP27 overexpression on TDP-43 and apoE. This will give us a better model to understand TDP-43 proteinopathies. In the present study, we hypothesize that increased expression of HSP27 may reduce TDP-43 aggregation and alter mitochondrial morphology. A new transgenic mouse model was developed to selectively drive human HSP27 and pathological TDP-43 with a defective nuclear localization signal (ΔNLS) in the hippocampus and neocortex using the Ca^{2+}/calmodulin protein kinase (Camk2a) tetracycline inducible system. We evaluated the following genotypes: wild-type, Camk2a/NLS, Camk2a/HSP27 and Camk2a/HSP27/TDP43ΔNLS at 4 months of age for immunohistochemistry, biochemistry (solubility fractionation), and Western blot. Preliminary *in vitro* results show that cells overexpressing HSP27 reduce aggregation and protein levels of TDP43. Mice overexpressing HSP27 in a TDP43ΔNLS background in the hippocampus show a reduction of aggregated TDP43. We also examined protein changes altering processing of full length TDP-43 in to N- and C-terminal fragments. Besides, samples were treated against human

specific TDP; no differences were observed between CK2/NLS and CK2/HSP/NLS mice. Interestingly, HSP27 overexpression modulated endogenous apoE expression. We will also explore interactions between HSP27 and apoE. To identify HSP27-apoE interactions, we will induce HSP27 and apoE isoforms in cellular and mice models. Immunohistochemical and bioenergetic experiments will be carried out to evaluate the overall brain and mitochondrial morphology upon HSP27 overexpression. Overall, our initial data suggests that modifying HSP27 expression may provide a point of therapeutic intervention for TDP-43 proteinopathies.

Disclosures: M. Abeer: None. **A.K. Gauff:** None. **M. Lima:** None. **S.A. Davis:** None. **M.A. Gitcho:** None.

Poster

532. Alzheimer's Disease and Other Dementia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 532.30

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

Support: German Research Foundation (DFG) grant KA1675/3-2 (P.J.K.)

Title: Acetylation modulates TDP-43 nucleo-cytoplasmic shuttling and promotes liquid-liquid phase separation.

Authors: J. GARCIA MORATO[1,2], F. HANS[1,2], F. VON ZWEYDORF[1], R. FEEDERLE[5,6], S. ELSAESSER[7], A. SKODRAS[3], C. GLOECKNER[1,8], E. BURATTI[9], V. GIACHIN[4], M. NEUMANN[1,10], *P. KAHLE[11,4,1];
[1]German centre for neurodegenerative diseases, Tuebingen, Germany; [3]Mol. Imaging Unit, Dept. of Cell. Neurol., [4]Functional Neurogenetics, [2]Hertie Inst. for Clin. Brain Res., Tuebingen, Germany; [5]Inst. for Diabetes and Obesity, Monoclonal Antibody Core Facility, Munich, Germany; [6]German Ctr. for Neurodegenerative Dis., Munich, Germany; [7]Sci. for Life Laboratory, Dept. of Med. Biochem. and Biophysics, Karolinska Institutet, Stockholm, Sweden; [8]Core Facility for Med. Bioanalytics (CFMB), , Ctr. for Ophthalmology, Univ. of Tübingen, Tuebingen, Germany; [9]ICGEB, Trieste, Italy; [10]Dept. of Neuropathology, Univ. Hosp. Tübingen, Tuebingen, Germany; [11]Univ. of Tuebingen, Tuebingen, Germany

Abstract: Acetylation modulates TDP-43 nucleo-cytoplasmic shuttling and promotes liquid-liquid phase separation
Neuropathological cytoplasmic inclusions of the RNA-binding protein TDP-43 characterize amyotrophic lateral sclerosis and distinct types of frontotemporal dementia. TDP-43 found in pathological aggregates is heavily ubiquitinated, phosphorylated and acetylated. We found four novel acetylation sites at specific residues in the nuclear localisation signal (K79, K84) and in the RNA-recognition domain (K121, K136). The relevance for TDP-43 pathophysiology was assessed via site directed-mutagenesis and amber suppression with acetyl-lysine. We describe how acetylation at lysine-84, located in the nuclear localisation signal, dramatically decreases

nuclear import of TDP-43. In addition, acetylation at lysine-136 impairs TDP-43 RNA-binding capabilities and drives TDP-43 into phase separated condensates. To assess the impact of these acetylations in TDP-43 pathophysiology, we developed specific antibodies to detect them. With these newly developed tools, we identified the nuclear deacetylating enzyme Sirtuin-1 (SIRT1) as the responsible enzyme to deacetylate both lysine-84 and lysine-136. Overexpression of SIRT1 significantly reduced the proportion of cells with acK84-driven cytoplasmic mislocalisation, as well as the number of acK136 TDP-43 liquid droplets. In addition, an indirect modulation of SIRT1 activity via resveratrol seems to reduce TDP-43 phase separation. To further confirm the pathological relevance of these acetylation sites, different immunoassays are being developed.
In summary, in this study we report that acetylation within the NLS region modulates TDP-43 nucleo-cytoplasmic shuttling. In addition, we have characterized in detail the effect of acetylation at the RNA-recognition domain. Acetylation at lysine-136 impairs TDP-43 RNA-binding capabilities and drives its phase separation and further pathological aggregation. This process can be modulated by SIRT1 activity, an enzyme which in turn can be modulated with small molecules such as resveratrol. Once the pathological relevance of these findings is confirmed, the regulation of TDP-43 acetylation could offer a new approach to reduce TDP-43 aggregation process in ALS and FTD.

Disclosures: **J. Garcia Morato:** None. **F. Hans:** None. **F. von Zweydorf:** None. **R. Feederle:** None. **S. Elsaesser:** None. **A. Skodras:** None. **C. Gloeckner:** None. **E. Buratti:** None. **V. Giachin:** None. **M. Neumann:** None. **P. Kahle:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.01

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

Title: Modelling neurodegeneration using a human isogenic cell system: A next generation approach to study Huntington's disease

Authors: *T. OOSTERVEEN[1], O. DOVEY[1], S. SALIC[2], M. GAMPERL[2], T. BURCKSTUMMER[2], A. VASILYEV[3], K. FIRTH[1], S. POKORNY[1], A. SIORENTAS[1], A. CONSTANTINE[1], T. FROLOV[1], F. PATELL-SOCHA[1], T. MOREAU[1], M. KOTTER[1]; [1]bit.bio, Cambridge, United Kingdom; [2]bit.bio discovery, Vienna, Austria; [3]Aelian, Vienna, Austria

Abstract: The development of therapies to treat patients with neuronal indications is currently hampered by the use of animal models as less than 10% of findings derived from these preclinical models can be translated to humans. Patient-derived induced pluripotent stem cells (iPSCs) offer the possibility to generate in vitro systems to model neurological diseases that can recapitulate relevant human disease phenotypes. However, conventional human iPSC (hiPSC)

differentiation protocols are often lengthy, inconsistent, and difficult to scale. More importantly, the lack of genetically matched controls for patient-derived models further complicates the investigation of disease-relevant phenotype and study of molecular mechanisms underlying neurodegeneration. To overcome these problems, we developed a proprietary gene- targeting strategy (opti-ox^{TM}) that enables highly controlled expression of transcription factors to rapidly reprogram hiPSCs into any specific somatic cell type in a scalable manner. Our reprogramming approach together with CRISPR/Cas9- mediated genetic engineering enables us to introduce specific mutations in these hiPSC-lines and create isogenic disease models that will improve screen specificity and accelerate drug development. We used our opti-ox^{TM} induced ioGlutamatergic Neurons to generate a Huntington's disease (HD) model that carries a 50CAG expansion in the huntingtin (HTT) gene. Mutant HTT proteins containing elongated polyglutamine (PolyQ) stretches are aggregation-prone and have been reported to affect a range of neuronal subtypes, including cortical glutamatergic neurons. Characterisation of the ioGlutamatergic Neurons HTT 50CAG showed that the expression profile of pan-neuronal (MAP2 and TUBB3) and glutamatergic (VGLUT1 and VGLUT2) marker genes as well as of the HTT transcript itself is highly similar to that of the wild type (WT) ioGlutamatergic Neurons. We are currently performing an in-depth phenotypic characterisation of this disease model and the genetically matched control to determine the differences in their transcriptome, neuronal activity and mitochondrial functions. Beside the 50CAG mutation in HTT, we have generated mutations in the MAPT, TARDBP, GBA and PRKN to provide isogenic disease models for FTD, FTD/ALS and Parkinson's disease. Our novel strategy to use the opti-ox^{TM} technology for the scalable and consistent production of hiPSC-derived isogenic disease models, offers new avenues into drug discovery and can accelerate research and the development of new therapeutics.

Disclosures: **T. Oosterveen:** A. Employment/Salary (full or part-time):; bit.bio. **O. Dovey:** A. Employment/Salary (full or part-time):; bit.bio. **S. Salic:** A. Employment/Salary (full or part-time):; bit.bio discovery. **M. Gamperl:** A. Employment/Salary (full or part-time):; bit.bio discovery. **T. Burckstummer:** A. Employment/Salary (full or part-time):; bit.bio discovery. **A. Vasilyev:** A. Employment/Salary (full or part-time):; Aelian. **K. Firth:** A. Employment/Salary (full or part-time):; bit.bio. **S. Pokorny:** A. Employment/Salary (full or part-time):; bit.bio. **A. Siorentas:** A. Employment/Salary (full or part-time):; bit.bio. **A. Constantine:** A. Employment/Salary (full or part-time):; bit.bio. **T. Frolov:** A. Employment/Salary (full or part-time):; bit.bio. **F. Patell-Socha:** A. Employment/Salary (full or part-time):; bit.bio. **T. Moreau:** A. Employment/Salary (full or part-time):; bit.bio. **M. Kotter:** A. Employment/Salary (full or part-time):; bit.bio.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.02

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

Title: Mechanistic characterization of compounds that lower mutant huntingtin in cell-based assays

Authors: *N. PRIGOZHINA, T. TRAN, T. KARG, B. HOFFMAN;
Origami Therapeut., San Diego, CA

Abstract: The currently incurable Huntington's disease (HD) is a progressive and highly debilitating neurodegenerative disease caused by a mutation in the huntingtin (HTT) protein. Mutant huntingtin protein (mHTT) is characterized by an expanded poly-glutamine tract leading to improper protein folding and, consequently, toxic gain of function, compromised protein degradation, cytotoxicity and the hallmark HD pathology of HTT protein aggregates. To address the root cause of the HD pathology, we employed a high throughput high content screen and identified multiple compounds that prevent mHTT aggregation in a cell-based assay. Emerging structure-activity relationships (SAR) reveal multiple chemotypes that lower mHTT levels. Here, we present initial results demonstrating the diverse modes of action for several Origami chemotypes. We use multiple cellular models, including primary human wild type (WT) and HD fibroblasts, as well as cell lines expressing wtHTT or mHTT under control of constitutive or inducible promoters. Several key compounds were effective in lowering HTT, reducing mHTT aggregation, modulating autophagy and enhancing neurite outgrowth. Additionally, we present preliminary data suggesting selective reduction of mHTT vs wtHTT.

Disclosures: **N. Prigozhina:** None. **T. Tran:** None. **T. Karg:** None. **B. Hoffman:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.03

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

Support: Lundbeck Foundation (SG)
The Hereditary Disease Foundation (AB)

Title: Huntington disease mice exhibit a TCF7L2-responsive suppression of both homeostatic and compensatory remeylination

Authors: *A. BENRAISS[1], J. N. MARIANI[2], A. TATE[2], P. M. MADSEN[2], K. M. CLARK[2], K. A. WELLE[2], R. SOLLY[2], L. CAPELLANO[2], K. L. BENTLEY[2], D. CHANDLER-MILITELLO[2], S. A. GOLDMAN[2,3];
[1]Univ. of Rochester, Rochester, NY; [2]Ctr. for Translational Neuromedicine and the Dept. of Neurol., Univ. of Rochester Med. Ctr., Rochester, NY; [3]Univ. of Copenhagen Fac. of Hlth. and Med. Sci., Copenhagen, Denmark

Abstract: Huntington's disease (HD) is characterized by defective glial differentiation as well as by its characteristic striatal neuronal loss. In particular, HD is associated with both early

hypomyelination and white matter disease, which may be radiographically evident in patients long before symptomatic onset otherwise. Here, we investigated the cellular basis for dysmyelination in two mouse models of HD, focusing on the role of glial progenitor cell dysfunction in that process. We first noted a progressive, age-related loss of myelin in R6/2 mice, an mHTT exon1 120CAG repeat knock-in model of HD, compared to wild-type (WT) controls. Following cuprizone demyelination, diseased R6/2 mice displayed significantly delayed recovery and remyelination compared to WT mice, at both 2 ($p<0.001$) and 4 weeks ($p<0.01$) after the cuprizone cessation. RNA-Sequencing and proteomic analysis of both callosal white matter and isolated GPCs, derived from both R6/2 and zQ175 mice (the latter a full-length mHTT model, with later symptomatic onset), revealed in each the relative down-regulation of genes associated with oligodendrocyte differentiation and myelinogenesis, compared to WT controls. Gene co-expression and network analysis predicted repressed TCF7L2 signaling as a major driver of this expression pattern. Lentiviral TCF7L2 over-expression in the R6/2 striatum resulted in the significant upregulation of a cohort of myelinogenic genes, which included Myrf, Mag, Plp1, Mbp and Tf, as well as the lipid biosynthetic genes Srebf1, Srebf2, and Hmgcr, all consistent with TCF7L2 role in regulating both glial myelinogenesis and lipid metabolism. On that basis, we assessed the ability of lentiviral TCF7L2 expression to potentiate remyelination in cuprizone-demyelinated R6/2 mice. We found that in vivo TCF7L2 overexpression proved sufficient to accelerate both the restoration of myelin gene expression and remyelination in demyelinated R6/2 mice. These data causally link impaired TCF7L2-dependent transcription to the deficient developmental and compensatory myelination of HD, and provide a potential target for its therapeutic restoration.

Disclosures: **A. Benraiss:** None. **J.N. Mariani:** None. **A. Tate:** None. **P.M. Madsen:** None. **K.M. Clark:** None. **K.A. Welle:** None. **R. Solly:** None. **L. Capellano:** None. **K.L. Bentley:** None. **D. Chandler-Militello:** None. **S.A. Goldman:** A. Employment/Salary (full or part-time):; Sana Biotechnology. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Sana Biotechnology.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.04

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

Title: Triming htt away in a hes cell model of huntington’s disease

Authors: *M. HERVA MOYANO[1], A. O'NEILL[2], T. CAJIGAL LOZANO[2], E. THATCHER[2], J. A. BARD[3];
[1]Discovery Charles River, Saffron Walden, United Kingdom; [2]Discovery Charles River, Saffron WAlden, United Kingdom; [3]CHDI Management, Inc., Princeton, NJ

Abstract: TRIM21 is a cytosolic Fc receptor ubiquitously expressed in the body. After antibody binding to antigen and binding of the antibody Fc fragment to a TRIM21 dimer, which functions as a E3 ligase, the antibody-antigen-TRIM21 complex is ubiquitinated and directed to the proteasome for degradation. Antibodies represent one of the few modalities which demonstrate allele selectivity for mutant huntingtin protein (mHTT) over wild-type HTT (wtHTT) and it has been shown in cell models overexpressing HTT and TRIM21 that allele specific mHTT degradation can be achieved (Cliff et al, 2018; Zeng et al, 2021). In this work, we aimed to target mHTT for proteasome degradation by intracellular delivery of either total HTT-specific or polyQ-specific antibodies into Q48 Genea020 hESC and neurons as a cellular model of endogenous HTT- and TRIM21- expressing proteins. The data that we are presenting demonstrates that TRIM21 is present in the Genea020 cell models at detectable levels (RNAseq and Westerns) and that the polyglutamine length-independent HTT antibody D7F7, and with more variable results, PolyQ antibodies, can lower mHTT levels (using HTRF assay). Preliminary data on Genea020-derived striatal neurons show a similar mHTT lowering effect. Furthermore, knock down of TRIM21 by siRNA as well as proteasome inhibition in ES cells at least partially abrogates the mHTT lowering effect, supporting the mechanism of action by TRIM21-mediated proteasome degradation. Next steps will focus on the mHTT specificity and the delivery of the HTT antibodies as cargo in AAV vectors both in cell models and animal models of HD. Such animal model studies could allow us to determine TRIM21-mediated HTT aggregate-specific or mHTT-selective lowering and measure the impact on disease progression. Overall, this proof-of-concept work demonstrates the value of engaging TRIM21 by using HTT-specific antibodies as a suitable way to lower HTT, with an additional advantage of being allelic selective and serving an advantage over other non-protein-directed therapeutic alternatives. References • Clift D, et al. A Method for the Acute and Rapid Degradation of Endogenous Proteins. Cell. 2017;171:1692-1706.:e18 • Zeng T, et al. Target-induced clustering activates Trim-Away of pathogens and proteins. Nat Struct Mol Biol. 2021 Mar 1; 28(3): 278-289.

Disclosures: **M. Herva Moyano:** None. **A. O'Neill:** None. **T. Cajigal Lozano:** None. **E. Thatcher:** None. **J.A. Bard:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.05

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

Title: Profiling of a novel orally active and brain penetrant splicing modulator small molecule demonstrates CNS and systemic mHTT lowering in BACHD mice

Authors: *D. MAGNANI[1], K. MALAGU[2], S. CLIFTON[2], H. ATTON[2], H. PATEL[2], D. MOTA[2], C. DAVIE[2], M. STEBBEDS[2], A. BARNARD[2], I. MANCINI[1], M. VISSER[1], D. TODD[1], M. HERVA MOYANO[1], M. CHAMBERS[1], P. MITCHELL[1], G. MCALLISTER[3], L. LIU[3], V. KHETARPAL[3], D. MACDONALD[3], C. DOMINGUEZ[3], I. MUNOZ-SANJUAN[3];

[1]Charles River Labs., [2]Charles River, Saffron Walden, United Kingdom; [3]CHDI Mgmt. Inc./CHDI Fndn., Los Angeles, CA

Abstract: Several drug modalities aimed at lowering levels of mutant HTT (mHTT) are currently in development for the treatment of Huntington's Disease (HD). Importantly, mHTT mRNA is increasingly being considered an important therapeutic target, and HTT lowering pharmacodynamic data have been recently shown in clinical trials with the use of antisense oligonucleotide (ASO) targeting HTT mRNA. However, those trials have recently been stopped for insufficient safety over therapeutic margin. Notably, ASOs are delivered intrathecally into the spinal cord or directly into the striatum due to their poor distribution.Small molecules aimed at the same target will provide the potential advantages of greater distribution, non-invasive delivery methods and higher patient accessibility. In this context, a growing number of small molecules targeting RNA are being identified and this class of molecules are progressing into the clinic. One such molecule is Branaplam (LMI070) targeting directly HTT mRNA and is now moving forward to clinical trials for HD. Branaplam was originally designed to target splicing of SMN2 exon 7, but also targets splicing of the exon junction 49-50 of HTT pre-mRNA, promoting retention of a pseudo-exon within intron 49b which contains a premature stop codon, subsequently resulting in nonsense-mediated decay (NMD)-dependent mRNA and consequential protein lowering.CHDI, with Charles River Early Discovery, has discovered a novel compound (SMSM1) targeting the same mechanism. Herein, we describe the *in vitro* and *in vivo* profile of SMSM1. Potency was assessed in an HD-relevant human stem cell model and demonstrated concentration-dependent splicing modulation and HTT protein-lowering. Moreover, Compound Y exhibited favourable ADME and PK properties, as well as significant HTT mRNA splicing and protein-lowering *in vivo* in brain and peripheral tissues of an HD rodent model.

Disclosures: D. Magnani: None. **K. Malagu:** None. **S. clifton:** None. **H. Atton:** None. **H. Patel:** None. **D. Mota:** None. **C. davie:** None. **M. Stebbeds:** None. **A. Barnard:** None. **I. Mancini:** None. **M. Visser:** None. **D. Todd:** None. **M. Herva Moyano:** None. **M. Chambers:** None. **P. Mitchell:** None. **G. McAllister:** None. **L. Liu:** None. **V. Khetarpal:** None. **D. Macdonald:** None. **C. Dominguez:** None. **I. Munoz-Sanjuan:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.06

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

Support: CHDI Foundation S. A. Ament
CHDI Foundation J. B. Carroll

Title: Identifying links between somatic expansion of the Huntington's disease mutation, misfolded huntingtin protein isoforms, and disease-associated transcriptomic states in striatal neurons through single-cell multi-omics

Authors: *M. CORTÉS-GUTIÉRREZ[1], R. BRAGG[2], J. CANTLE[3], S. MALAIYA[4], E. WILDERMUTH[6], J. B. CARROLL[3], S. A. AMENT[5];
[1]Univ. of Maryalnd, Baltimore, MD; [2]Dept. of Psychology, [3]Psychology, Western Washington Univ., Bellingham, WA; [4]1.Institute for Genome Sciences,, [5]Dept. of Psychiatry and Inst. for Genome Sci., Univ. of Maryland Sch. of Med., Baltimore, MD; [6]Inst. for Genome Sci., Univ. of Maryland, Baltimore, MD

Abstract: Huntington's disease (HD) is the most common monogenic form of neurodegeneration and is caused by an expanded trinucleotide (CAG) repeat in the HTT gene. Here, we used single-cell multi-omic technologies to test hypotheses regarding the pathogenic mechanisms of HD mutations in vulnerable neurons. This study was carried out using the striatum of six-month-old $Htt^{Q111/+}$ mice, a relatively early timepoint at which cellular and behavioral differences are detectable but subtle. First, we established that in these mice, striatal spiny projection neurons exist along a trajectory from healthy to disease-associated transcriptomic states. Second, we tested the hypothesis that pathological changes occur initially in cells that have undergone further somatic expansion of the HTT CAG repeat. For this purpose, we performed single-cell long-read sequencing to correlate the HTT CAG length and transcriptomic states across thousands of striatal neurons. Third, we tested the hypothesis that HD pathogenesis is seeded by the formation of specific misfolded HTT (mHTT) protein isoforms, utilizing Cellular Indexing of Transcriptomes and Epitopes by Sequencing (CITE-seq) to quantify soluble (3B5H10) and insoluble (MW8) mHTT isoforms in striatal neurons in parallel with their transcriptomic states. Fourth, we tested the hypothesis that HD pathogenesis involves dysregulation of epigenomic states, utilizing single-nucleus Assay for Transposase Accessible Chromatin. Finally, we tested that disease-associated transcriptomic changes can be rescued by treating $Htt^{Q111/+}$ mice with a therapeutic antisense oligonucleotide targeting the HTT gene. Our findings will aid in the development of novel therapeutics targeting each of these pathogenic mechanisms. Funding: This study was supported by contracts from the CHDI Foundation to S.A.A. and J.B.C.

Disclosures: M. Cortés-Gutiérrez: None. **R. Bragg:** None. **J. Cantle:** None. **S. Malaiya:** None. **E. Wildermuth:** None. **J.B. Carroll:** None. **S.A. Ament:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.07

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

Support: Nebraska Biomedical Research Development Fund LB692

Title: Analysis of gene networks and pathways in the striatum of HD mice exposed to a super-enriched environment

Authors: *H. KIM[1], S. H. WEE[1], J.-C. CHAI[1], T. YOO[1], R. ZUKIN[2], J.-Y. HWANG[1]; [1]Dept. of Pharmacol. and Neurosciences, Creighton Univ. Sch. of Med., Omaha, NE; [2]Dominick P. Purpura Dept. of Neurosci., Albert Einstein Col. of Med., Bronx, NY

Abstract: Huntington's disease (HD) is an autosomal dominant neurodegenerative disorder characterized by progressive motor, psychiatric, and cognitive decline. Marked neuronal loss in the striatum and cerebral cortical structures is the main neuropathological hallmark of HD. Environmental factors can alter gene expression through epigenetic responses and can change the mechanisms of experience-dependent plasticity such as synaptic plasticity and neurogenesis. An enriched environment has been shown to delay the onset and progression of motor symptoms and to improve neurological function and cognitive deficits in HD mice. However, the impact of an enriched environment on transcriptional changes in the R6/2 transgenic mouse model of HD is yet unclear. To address this issue, we performed genome-wide transcriptome analysis using next-generation RNA-seq in striatal tissue from HD and wild-type (WT) mice reared in either an enriched (EE) or normal (NE) housing environment. We then performed bioinformatic analysis on the differentially expressed genes (DEGs, fold change of ±2 and FDR < 0.01) using the Ingenuity Pathway Analysis (IPA). We first investigated how gene expression is changed in HD mice during the development of HD symptoms. Canonical pathway analysis showed that the activity of the synaptogenesis signaling pathway is reduced in HD mice reared under the normal environment at 12 weeks *vs.* 4 weeks. An analysis in disease and functions showed that the gene network of these DEGs is implicated in movement disorders and the degeneration of neurons. These results indicate that genes changed in HD mice correlate with the pathogenesis of HD. Next, we analyzed 268 DEGs identified as 'EE'-common genes, which are changed in both HD and WT mice reared in an EE vs. NE. The IPA analysis of these common DEGs showed that genes associated with networks of long-term potentiation, the release of neurotransmitters, and learning and memory are activated, while genes associated with networks of behavior deficits, movement disorders, and neurological signs are inhibited. Taken together, we anticipate that the identification of genes and gene networks that are altered in HD mice in response to enriched environments will assist us in understanding how enriched environments can improve HD symptoms and neurological functions and will contribute to the development of novel therapeutic strategies to ameliorate motor and cognitive deficits associated with this debilitating and devastating disease.

Disclosures: H. Kim: None. **S.H. Wee:** None. **J. Chai:** None. **T. Yoo:** None. **R. Zukin:** None. **J. Hwang:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.08

Title: WITHDRAWN

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.09

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

Support: NIH/NINDS Grant RO1-NS110943
NIH/NIA Grant AG057509
CHDI Foundation

Title: Hap40 is a positive regulator of endogenous huntingtin and a potential modulator of huntington's disease pathogenesis

Authors: *S. M. FARMER[1], Y. YU[1], A. SOLBACH[1], S. XU[1], G. LI[1], X. YE[1], D. CHEN[1], Z. CHEN[1], Z. XU[1], M. DANIELE[3], S. TAMBONE[3], A. CECCACCI[3], L. TOMEI[3], L. YE[1], E. F. STIMMING[2], G. MCALLISTER[4], D. MARCHIONINI[4], S. ZHANG[1];
[1]The Brown Fndn. Inst. of Mol. Med., [2]Neurology, HDSA Ctr. of Excellence, Univ. of Texas Hlth. Sci. Ctr. at Houston, Houston, TX; [3]Translational and Discovery Res., IRBM SpA, Pomezia (RM), Italy; [4]CHDI Management/CHDI Fndn., New York, NY

Abstract: Huntington's disease (HD) is a devastating hereditary neurodegenerative disorder caused by an abnormal CAG repeat expansion encoding a polyglutamine tract (polyQ) in Huntingtin (HTT). Aberrations in HTT's function due to polyQ expansion is one postulated factor in HD pathogenesis; however, the molecular function and regulation of HTT remain to be elucidated. Using a proteomics-based approach, we identified a novel 40kDa protein encoded by an uncharacterized CG8134 gene as a strong interactor of *Drosophila* HTT and further demonstrated it was the functional ortholog of human HAP40, an HTT-associated protein shown recently to modulate HTT's conformation but with unclear physiological and pathologic roles. Molecular modeling suggested that the structural architectures and interaction domains are conserved between human and *Drosophila* HTT/HAP40 complexes. Validation experiments in flies and human cells supported conserved physical and functional interactions of HAP40 with HTT across the evolutionarily distant species. Further, genetic interaction assays showed that loss of HAP40 causes similar phenotypes as HTT knockout. More strikingly, HAP40 was found to strongly affect HTT's protein stability, as depletion of HAP40 significantly reduced the levels of endogenous HTT protein while HAP40 overexpression markedly extended its half-life. Conversely, in HTT-deficient cells, the majority of HAP40 protein was degraded potentially by the proteasome. Further, polyQ expansion did not significantly alter the affinity of the HTT/HAP40 complex, and there were no abnormal accumulations of endogenous HAP40 protein in HD cells from mouse HD models or human patients. Lastly, when tested in *Drosophila* models of HD, HAP40 modulated the neurodegenerative effects of full-length mutant HTT, but showed no apparent effect on the toxicity of mutant HTT exon 1 fragment. Taken together, our study uncovers a conserved mechanism governing HTT's protein stability and endogenous function. Further, our results support that mutant HTT is toxic regardless of its association with HAP40, but HAP40 might be a potential modulator of HD pathogenesis through its multiplex effect on endogenous HTT.

Disclosures: **S.M. Farmer:** None. **Y. Yu:** None. **A. Solbach:** None. **S. Xu:** None. **G. Li:** None. **X. Ye:** None. **D. Chen:** None. **Z. Chen:** None. **Z. Xu:** None. **M. Daniele:** None. **S. Tambone:** None. **A. Ceccacci:** None. **L. Tomei:** None. **L. Ye:** None. **E.F. Stimming:** None. **G. McAllister:** None. **D. Marchionini:** None. **S. Zhang:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.10

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

Title: Establishing a hiPSC derived Huntington's disease neuronal model suitable for phenotypic drug screening & to identify small molecule modulators of mutant HTT

Authors: L. BUTLER[1], **D. MAGNANI**[1], T. OOSTERVEEN[2], O. DOVEY[2], T. FROLOV[2], F. PATELL-SOCHA[2], M. STEBBEDS[1], M. BROWN[1], G. LANGLEY[1], J. ANTON[1], G. GULBAHACE[1], C. MANSAT[1], *M. IOVINO[1];
[1]Charles River, Saffron Walden, United Kingdom; [2]bit.bio, Cambridge, United Kingdom

Abstract: Lowering of the pathogenic mutant huntingtin (mHTT) protein in Huntington's disease (HD) patients is one of the leading approaches to ameliorate the fatal neurodegeneration caused by the poly-CAG expansion in the *Htt* gene. Current therapeutics in development involve small molecule splicing modulators or use of novel biological agents such as ASOs, RNAi, ZFTR and CRISPR/Cas9. Therefore, physiologically relevant and scalable models of HD are needed to improve outcomes and efficiency of drug development. The use of human induced Pluripotent Stem Cell (iPSC)-derived neurons would offer disease relevant in vitro system however can be hindered by low scalability, and long complex protocols. The cell reprogramming technology, opti-ox^{TM} , in combination with CRISPR-Cas9 gene editing has been used to develop iPSC-derived ioGlutamatergic Neurons carrying a HTT allele with an abnormal 50 CAG repeat expansion. We have used high content imaging analysis and branched DNA assay to characterise ioGlutamatergic Neurons HTT$^{50CAG/WT}$ together with their isogenic control. Moreover, we have used Multi Electrode Array (MEA) platform to study functional activity of the ioGlutamatergic Neurons HTT$^{50CAG/WT}$ compared to the isogenic control. Preliminary functional data showed that ioGlutamatergic Neurons HTT$^{50CAG/WT}$ demonstrated formation of synchronous activity at late stage of maturation, suggesting these neurons are electrophysiologically active and amenable to functional studies. Here we also established a neuronal HTRF assay using ioGlutamatergic Neurons HTT$^{50CAG/WT}$ that provides optimal conditions with robust assay statistics suitable for future compound screening. Counter-screen and orthogonal assay formats will be developed and applied to define specificity and putative mechanisms of action.

Disclosures: **L. Butler:** None. **D. Magnani:** None. **T. Oosterveen:** None. **O. Dovey:** None. **T. Frolov:** None. **F. Patell-Socha:** None. **M. Stebbeds:** None. **M. Brown:** None. **G. Langley:** None. **J. Anton:** None. **G. Gulbahace:** None. **C. Mansat:** None. **M. Iovino:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.11

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

Support: MOST 110-2320-B-001-018-MY3

Title: Contribution of a PIAS1 S510G variant to the late onset of Huntington's Disease

Authors: *Y. LEE[1], Y. CHERN[2];
[1]Inst. of Biomed. Sciences, Academia Sinica, Academia Sinica, Taipei, Taiwan; [2]Inst. Biomed Sci., Taipei, Taiwan

Abstract: Contribution of a PIAS1 S510G variant to the late onset of Huntington's Disease
AuthorsYan Hua Lee[1]**,** Hui-Mei Chen[1], Hsing-Lin Lai[1], Chia-Wei Lee[1], Yijuang Chern[1].
[1] Institute of Biomedical Sciences, Academia Sinica, Taipei, Taiwan.
DisclosuresY.H.L. and Y.C. are inventors on a pending patent (entitled "Identification of Common Genetic Modifiers for Neurodegenerative Disease") (63/071,903, USA). The authors have no additional financial interests.
AbstractPolyglutamine (PolyQ) diseases are progressive neurodegenerative disorders caused by the polyQ encoding CAG-repeat expansion in the disease-causing gene. The CAG-repeat length is the major determinant of the age-at-onset (AO) of polyQ diseases, including Huntington's disease (HD) and spinocerebellar ataxia type 3 (SCA3). Of note, some patients have an AO deviated from the average AO, suggesting the presence of genetic modifier(s). Genetic modifiers may have substantial impact on disease onset, severity, and/or progression. We have recently shown that a genetic variant of PIAS1 (A445T, Ser510Gly), identified from late onset polyQ disease patients, may delay the onset of HD by modifying the homeostasis of mutant Huntingtin (mHTT). Biochemical analyses revealed that $PIAS1^{S510G}$ has a reduced ability to interact with mHTT compared to $PIAS1^{WT}$, causing a lower SUMOylation levels of mHTT and diminished accumulation of insoluble mHTT. The expression of the naturally occurring variant, $PIAS1^{S510G}$, in a mouse model of HD (R6/2) markedly improved several HD symptoms (including shortened life spans, motor dysfunction and mHTT accumulation), validating our clinical finding. To gain further insight into the underlying protective effect of $PIAS1^{S510G}$ in HD, we carried out quantitative proteomic analysis to investigate the impact of $Pias1^{S510G}$ on the brain proteome of WT or HD mice expressing $Pias1^{S510G}$. Brain samples from 4 weeks of age (pre-HD stage) were analyzed. Because mHTT exists in monomeric and oligomeric forms in the pre-HD stage, changes in the brain proteome would mostly likely be $Pias1^{S510G}$-specific. Proteomic analysis revealed that there is no strong proteome alteration at the age of 4 weeks. Only two proteins

(protein shroom 4 (*Shroom4*) and FXYD domain containing ion transport regulator 7 (*Fxyd7*)) were found to be significantly down-regulated in the HD brain harboring Pias1^{S510G} when compared with that of Pias1^{WT}. Additional analysis on the mouse brain proteome at the age of 12 weeks (late-HD stage) will be very important. The resultant comprehensive understanding of the mechanistic contribution of PIAS1^{S510G} to HD pathogenesis may facilitate the development of new therapies for HD.

Disclosures: Y. Lee: None. **Y. Chern:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.12

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

Support: Huntington's Disease Society of America

Title: Mutant huntingtin protein interaction map for dysregulation of cellular pathways, including translation, signal transduction, and mitochondrial systems, in neurodegeneration of Huntington’s disease

Authors: *S. PODVIN[1], S. B. ROSENTHAL[2], W. POON[1], E. WEI[1], K. FISCH[2], V. HOOK[3]; [1]Skaggs Sch. of Pharm. and Pharmaceut. Sci., [2]Ctr. for Computat. Biol. and Bioinformatics, Dept. of Med., [3]Skaggs Sch. of Pharm. and Pharmaceut. Sciences, Dept. of Neuroscience, Dept. of Pharm., UC San Diego, La Jolla, CA

Abstract: Huntington’s disease (HD) is a genetic neurodegenerative disease caused by trinucleotide repeat (CAG) expansions in the human HTT gene encoding the huntingtin protein (Htt) with an expanded polyglutamine tract. Non-human HD models from yeast to transgenic mice have investigated proteins interacting with mutant Htt that may initiate molecular pathways of cell death. There is a paucity of datasets of published Htt protein interactions that include the criteria of (1) defining fragments or full-length Htt forms, (2) indicating the number of poly-glutamines of the mutant and wild-type Htt forms, and (3) evaluating native Htt interaction complexes. Therefore, this study evaluated Htt interactor data in the literature reported in ~193 articles to gain understanding of mutant Htt dysregulation of cellular pathways. This analysis compiled a data set of Htt interacting proteins from the literature that meet our criteria and were subjected to network analysis via clustering, gene ontology, and KEGG pathways using rigorous statistical methods. This data set of Htt interactors found that both mutant and wild-type Htt interact with more than 2971 proteins. Application of a community detection algorithm to all known Htt interactors identified significant signal transduction, membrane trafficking, chromatin, and mitochondrial clusters, among others. Binomial analyses of a subset of reported protein interactor information determined that chromatin organization, signal transduction and endocytosis were diminished, while mitochondria, translation and membrane trafficking had

enriched overall edge effects. These data support the hypothesis that mutant Htt disrupts multiple cellular processes that may lead to toxicity, including translation, signal transduction, and mitochondrial systems. This dataset is an open resource to benefit investigators in formulating hypotheses of HD mechanisms of pathogenesis.

Disclosures: S. Podvin: None. **S.B. Rosenthal:** None. **W. Poon:** None. **E. Wei:** None. **K. Fisch:** None. **V. Hook:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.13

Title: WITHDRAWN

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.14

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

Support: CIHR Grant 162326

Title: Wild type huntingtin is essential for synapse stability and plasticity in the adult hippocampus

Authors: *J. C. BARRON, F. NAFAR, M. P. PARSONS;
Mem. Univ. of Newfoundland, St. John's, NL, Canada

Abstract: Huntington disease (HD) is a fatal neurodegenerative disease that results in a triad of motor, cognitive and psychiatric symptoms. Being a monogenic disorder, it is an ideal candidate for genetic therapies, which can target the root cause of HD: the mutant huntingtin (HTT) gene. However, many of these HD therapies currently in clinical trials also result in lowered levels of wild type HTT (wtHTT), which has been suggested to play an important role in cellular functions that promote synapse stability and plasticity, such as fast axonal transport, neurotransmitter release and receptor localization. Synaptic disruption occurs early in HD and is known to precede and predict neuronal cell death. Consequences of wtHTT reduction in the adult brain remain poorly understood, and additional research at the preclinical level is required immediately to better understand the risk factors associated with HTT-lowering therapeutics. We investigated the consequences of wtHTT deletion in the adult hippocampus using both cell

culture and mouse models. wtHTT-lowered cultured hippocampal cells showed a significant reduction in synaptic connections, as well as attenuated calcium transients, in comparison to siRNA vector-treated control cells. When synapse function was investigated using electrophysiological methods in our wtHTT conditional knockout (cKO) mouse models, we found that cKO mice showed impairments in both short- and long-term hippocampal plasticity. A decrease in neurotransmitter release probability was also seen in cKO animals. Results described here indicate that wtHTT is required for maintaining proper synaptic stability and function in the adult mammalian hippocampus.

Disclosures: **J.C. Barron:** None. **F. Nafar:** None. **M.P. Parsons:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.15

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

Support: CIHR

Title: Synaptic Dysfunction and Atypical NMDA Receptors in Huntington Disease

Authors: *E. HURLEY[1], F. NAFAR[2], L. FANG[3], B. MUKHERJEE[3], M. PARSONS[4]; [1]Mem. Univ. of Newfoundland, Mem. Univ. of Newfoundland, St. John's, NL, Canada; [2]Mem. Univ. of Newfoundland, Mem. Univ. of Newfoundland, Stjohn's, NL, Canada; [3]Mem. Univ. of Newfoundland, St. John's, NL, Canada; [4]Mem. Univ. of Newfoundland, St John's, NL, Canada

Abstract: Huntington Disease (HD) is a fatal inherited neurodegenerative disease with a triad of cognitive, motor, and psychiatric symptoms. HD is caused by a CAG-triplet repeat expansion in the huntingtin gene (HTT), resulting in the pathogenic mutant HTT protein that affects cellular functions that result in the dysfunction and death of neurons. However, before overt neuronal death is observed, alterations in synaptic function occur, suggesting that HD is initially a synaptic disease. Using the powerful combination of the Airyscan super-resolution microscope and Imaris software, we conducted a large-scale synaptic analysis of several brain regions heavily hit in HD - the striatum and hippocampus - and revealed multiple synaptic alterations in HD mice including synapse density and distance. Healthy cognitive function relies on synaptic plasticity. N-methyl-D-aspartate receptors (NMDARs) assemble as functionally diverse heterotetramers with essential roles in synaptic plasticity. Of all the NMDAR subunits, GluN3A may be the most unusual. Incorporation of the GluN3A subunit into NMDARs alters conventional NMDAR properties. GluN3A-containing NMDARs have reduced magnesium (Mg2+) sensitivity and reduced calcium permeability. GluN3A expression peaks during the first 1-2 weeks of postnatal life, progressively declines and remains low into adulthood in most brain regions. In early experiments, we demonstrate that synaptic plasticity is impaired in adult brain regions that retain GluN3A expression. We show that GluN3A is abnormally elevated in the HD

hippocampus at synaptic and extrasynaptic locations. Whole-cell patching of CA1 pyramidal neurons revealed an increase in glycine-induced currents in the HD hippocampus. In addition, Mg2+ sensitivity is significantly reduced in the HD hippocampus. Together this suggests that GluN3A-containing NMDARs assemble as both combinations of GluN1/GluN2/GluN3A triheteromeric receptors and GluN1/GluN3A excitatory glycine receptors in HD. Synaptic plasticity is impaired in the HD hippocampus. Perhaps elevated GluN3A may contribute to the cognitive impairments in HD. In current experiments, HD mice are injected into the hippocampus with a short hairpin RNA to reduce GluN3A levels to determine if GluN3A knockdown can restore cognition in HD.

Disclosures: E. Hurley: None. **F. Nafar:** None. **L. Fang:** None. **B. Mukherjee:** None. **M. Parsons:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.16

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

Support: Brain Canada Foundation (RES0030547)
Alberta Prion Research Institute (RES0039430)
Glyconet (RES0050801)
SynAD (RES0049270)
Canadian Institutes of Health Research (RES0052715)
Canadian Institutes of Health Research Frederick Banting and Charles Best Canada Graduate Scholarships
SynAD ADRD Trainee Grant

Title: Gangliosides are novel modulators of extracellular vesicle biogenesis: implications for protein misfolding neurodegenerative diseases

Authors: *J. MONYROR[1], V. KADAM[2], A. K. ZAIDI[1], L. C. MORALES[1], D. ORDÓÑEZ[1], J. IBANGA[1], A. KRYSLER[1], B. HUBBARD[1,4], S.-A. MOK[3,2], E. POSSE DE CHAVES[1,2], S. SIPIONE[1,2];
[1]Dept. of Pharmacol., [2]Neurosci. and Mental Hlth. Inst., [3]Dept. of Biochem., Univ. of Alberta, Edmonton, AB, Canada; [4]Dept. of Pharmacol. and Toxicology, Univ. of Toronto, Toronto, ON, Canada

Abstract: Gangliosides are sialic acid-containing glycosphingolipids that are crucial players in cell signaling and cell-cell communication. In some protein misfolding diseases, brain levels of gangliosides are perturbed, and administration of exogenous gangliosides, particularly GM1, is neuroprotective. Gangliosides are present in extracellular vesicles (EVs), secreted nanoparticles which contribute to the clearance and spread of misfolded proteins. Here, we investigated how

exogenously administered or endogenously synthesized gangliosides modulate secretion of EVs and misfolded cargo proteins.
Cell models of Huntington's disease, Parkinson's disease and tauopathies were treated with various gangliosides to increase cell ganglioside levels. To deplete gangliosides, cells were treated with Genz-123346 (an inhibitor of ganglioside biosynthesis), or *B4galnt1* was knocked-out using CRISPR-Cas9. DiD-labelled EVs and their misfolded protein cargo were analyzed by imaging flow cytometry, fluorometry, ELISA, immunoblot and ExoView.
Ganglioside GM1 increased secretion of EVs from neuronal cells and primary human fibroblasts. GM1 also promoted the secretion of mHTT, A53T α-synuclein, tau, N279K tau and P310L tau within EV fractions. The ceramide tail of GM1 was required for these effects, as treatment of neuronal cells with the GM1 pentasaccharide alone did not recapitulate the stimulatory action of GM1 on EV secretion. Other gangliosides also increased EV secretion, except for GM3 and GD3, which had a partial inhibitory activity on EV secretion. These data suggest that the N-acetyl-D-galactosamine residue in the glycan headgroup, which discriminate stimulatory gangliosides from GM3 and GD3, is required for ganglioside-induced EV secretion. We also found that endogenous gangliosides play an important role in EV biogenesis. Depletion of gangliosides with Genz-123346 reduced EV and misfolded protein secretion. Similarly, knockout of the ganglioside biosynthetic gene *B4galnt1* resulted in impaired EV secretion that could be rescued by treatment with an equimolar mixture of major brain gangliosides GM1, GD1a, GD1b and GT1b.
Altogether, our data identify gangliosides as novel modulators of EV secretion and suggest that decreased ganglioside levels in protein misfolding diseases might decrease the secretion of misfolded proteins and exacerbate proteotoxic stress within neurons. Therapies that raise ganglioside levels might alleviate proteotoxic stress in neurodegenerative diseases by promoting clearance of misfolded proteins through EVs.

Disclosures: **J. Monyror:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); University of Alberta. **V. Kadam:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); University of Alberta. **A.K. Zaidi:** None. **L.C. Morales:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); University of Alberta. **D. Ordóñez:** None. **J. Ibanga:** None. **A. Krysler:** None. **B. Hubbard:** None. **S. Mok:** None. **E. Posse de Chaves:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); University of Alberta. **S. Sipione:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); University of Alberta.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.17

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

Support: CHDI Foundation (A-5552 to A.M.G.)
Saks Kavanaugh Foundation (to A.M.G.)
National Institutes of Health (R01 MH060379 to A.M.G.; R01 NS100802 to M.H.)
Nancy Lurie Marks Family Foundation (to A.M.G.)
Simons Foundation (SFARI 306140 and grant to the Simons Center for the Social Brain at MIT, to A.M.G.)
JPB Foundation (to H.L. and M.H.)

Title: Huntington's disease produces multiplexed transcriptional vulnerabilities of striatal D1-D2 and Striosome-Matrix Neurons

Authors: *A. MATSUSHIMA[1], S. S. PINEDA[1,2], J. R. CRITTENDEN[1], H. LEE[1,2], K. GALANI[1,2], J. MANTERO[1,2], M. KELLIS[1,2], M. HEIMAN[1,2], A. M. GRAYBIEL[1];
[1]MIT, Cambridge, MA; [2]Broad Inst. of MIT and Harvard, Cambridge, MA

Abstract: Striatal cell-type-specific vulnerability in Huntington's disease (HD) preferentially affects dopamine D2R-expressing projection neurons (SPNs), compatible with manifest motor symptomatology in HD. A second, less fully studied feature of striatal vulnerability involves the compartmental organization of the striatum, with neurochemically specialized labyrinthine 'striosomes' thought to be affected especially in relation to premanifest mood symptomatology.To disentangle the cell-type-specific vulnerability in HD, we performed single-nucleus RNA sequencing on striatal samples from two murine models (zQ175 and R6/2) and rare Grade 1 HD patient tissue, and examined striosome and matrix sub-clusters within parent D1 and D2 SPN clusters. In the Grade 1 human HD, striosomal SPNs were the most depleted SPN population. Surprisingly, for both mouse models, transcriptomic distinctiveness was diminished more for striosome-matrix SPNs than for D1-D2 SPNs. Compartmental markers tended to cancel endogenous identities of striosomal and matrix SPNs; striosomal makers were downregulated in striosomal SPNs and upregulated in matrix SPNs, and matrix markers were upregulated in striosomal SPNs and downregulated in matrix SPNs. On the contrary, markers for D1-D2 SPNs exhibited less identity obscuring; they appeared up- and down-regulated in a non-systematic way. The degree of dysregulation (i.e., absolute values of up- or down-regulations) was largest in D2R-expressing SPNs, recapitulating the D2-dominent vulnerability in HD, and reflected in genes upregulated in specific cell types and downregulated in others.These results suggest that striosomes are the first to die in human HD, and that striosome-matrix identities are more vulnerable than those of D1-D2, a pattern that could reflect a differentiation deficiency during development due to loss of function of normal Huntingtin, as proposed previously. Given that D2-dominent transcriptional dysregulation is observed from only about the age of onset, the two axes of striatal organization might be affected differentially in time and in nature, with the striosome-matrix axis affected during development, leading to deficient compartmental identities, and the D1-D2 axis affected later, around the age of onset of motor symptoms.

Disclosures: A. Matsushima: None. **S.S. Pineda:** None. **J.R. Crittenden:** None. **H. Lee:** None. **K. Galani:** None. **J. Mantero:** None. **M. Kellis:** None. **M. Heiman:** None. **A.M. Graybiel:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.18

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

Support: Internal funding from the Lieber Institute for Brain Development

Title: The transcriptome and proteome of medium spiny neurons in Huntington's Disease: a focused laser capture microdissection study in post-mortem human brain

Authors: E. S. EL-HALAWANI[1], A. M. MARTINS[1], R. BHARADWAJ[1], L. BARROW[2], R. TAO[1], L. R. DEVINE[3], R. N. COLE[3], Q. CHEN[1], J. E. KLEINMAN[1], D. R. WEINBERGER[1], *T. M. HYDE[1];
[1]Lieber Inst. for Brain Develop., Baltimore, MD; [2]Univ. of Washington, Seattle, WA; [3]Dept. of Biol. Chem., Johns Hopkins Sch. of Med., Baltimore, MD

Abstract: Huntington's Disease (HD) is an autosomal dominant, neurodegenerative disorder marked by aberrant expression of the expanded Huntingtin gene (HTT). Expression of mutant *HTT* results in selective atrophy of medium spiny neurons (MSN) in the striatum. These neurons represent 85% of the neuronal population in the caudate nucleus. Previous studies have analyzed the caudate transcriptome in HD with a lack of consistency and an absence of corresponding proteome characterization. We hypothesize that an MSN enriched transcriptome and proteome characterization will identify gene targets central to the neurobiology of HD. We utilized postmortem, human caudate from 20 individuals with Huntington's disease and 20 neurotypical controls. The HD subjects have been clinically and neuropathologically diagnosed with HD, ranging from a severity score of 1- 4 based on the United Huntington's Disease Rating Scale (UHDRS). Our samples are 55% female with a mean age of 57.6 (SD = 11.1, min = 33, max = 78) at time of death, which supports the interrogation of sex based molecular changes underlying HD in MSN. We employ laser capture microdissection (LCM) to obtain whole MSN cell bodies (~5000 neuron enrichments) from each of the 40 samples to allow analysis of the cytosol in addition to the nuclei of these neurons. We performed RNA sequencing and tandem mass tag - mass spectrometry (Orbitrap Fusion Lumos) on the same MSN lysates to compare the transcriptomic and proteomic profiles at the gene and isoform levels. This methodology has been previously employed in our studies of the human nigrostriatal circuit, where we isolated 7,500 dopamine (DA) neurons and subsequently characterized over 112,000 'unique' DA peptides, 9,000 protein groups, and 7,000 master proteins (5% FDR, Proteome Discoverer v2.5). We identified 91 protein-protein interaction networks (String v11.5) significantly enriched for mitochondrial and oxidative phosphorylation pathways. Applying the same methodology and analytic paradigm, we expect to find (i) molecular correlations with HTT polyglutamine repeat lengths and severity of HD (ii) differentially expressed pathways underlying HD in MSN populations (iii) MSN-specific gene and protein networks that have previously not been defined in controls or HD subjects and (iv) phosphorylation-driven signaling changes in specific protein

pathways that are closer to functional gene expression underlying HD. The findings from our study will potentially identify novel therapeutic targets for HD and other neurodegenerative diseases.

Disclosures: E.S. El-Halawani: None. **A.M. Martins:** None. **R. Bharadwaj:** None. **L. Barrow:** None. **R. Tao:** None. **L.R. DeVine:** None. **R.N. Cole:** None. **Q. Chen:** None. **J.E. Kleinman:** None. **D.R. Weinberger:** None. **T.M. Hyde:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.19

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

Support: NIH R01NS086452

Title: Targeting PTMs by CDKs inhibitor to reduce neuronal toxicity of mHTT

Authors: *J. JIN, A. KRISHNAPRAKASH, M. JIANG, T. SHI, T. RATOVITSKI, C. A. ROSS;
Johns Hopkins University, Sch. of Med., Baltimore, MD

Abstract: Huntington's disease (HD) is an autosomal dominant neurodegenerative disease caused by a single mutation in huntingtin gene (*HTT*). Normal HTT has a CAG trinucleotide repeat at its N-terminal of 36 or less. However, once the CAG repeats exceeded 37, the mutant gene (*mHTT*) will encode mutant HTT (mHTT) causing neurodegeneration in the striatum, and other brain regions. HTT is a large protein, with many posttranslational modification sites (PTMs). These PTMs can be enzymatically modified by phosphorylation, acetylation, methylation and SUMOylation etc. Some previously reported modifications reduced mHTT toxicity both in cell and animal model of HD. We aimed to find PTMs which enhance toxicity, so that kinase or other enzyme inhibitors would be predicted to reduce toxicity.
We performed in vitro kinase assay using several HTT peptides with PTM sites. Total of 369 kinases were screened. Among those kinases, CDKs affected the serine phosphorylation on the peptides which contain S1181-HTT and S1201-HTT. We tested the effect of a CDKs inhibitor, roscovitine, on mHTT induced toxicity. Roscovitine protected neurons from mHTT induced toxicity. We further confirmed that roscovitine reduced phosphorylation of S1181 and S1201 of mHTT. As roscovitine can inhibit both CDK1 and CDK5, we explored the effect of CDK1 and CDK5 on phosphorylation of S1181-HTT and S1201-HTT. Both CDK1 and CDK5 knockdown can reduce the phosphorylation of S1181 and S1201. Furthermore, we tested the effect of replacing the serine (S) of mHTT with alanine (A) at PTM sites of 1181 (S1181A) and 1201 (S1201A) on mHTT toxicity. Mutant HTT with S◊A had less toxic effect in our HD cell model, indicating that modifying S1181 and S1201 would reduce mHTT toxicity. We will further confirm if roscovitine's protective effect is through targeting these 2 serine sites. We concluded

that CDK inhibition (we hypothesize at least in part mediated by decreased phosphorylation of S1181 and S1201 of mHTT) may be a promising strategy to reduce the neuronal toxicity induced by mHTT.

Disclosures: J. Jin: None. **A. Krishnaprakash:** None. **M. Jiang:** None. **T. Shi:** None. **T. Ratovitski:** None. **C.A. Ross:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.20

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

Support: NINDS R24 201603716
NINDS 5R01NS102486-05
NIH NIGMS Predoctoral Fellowship T32GM099608
A Stewart's and Dake Family Gift
HELP4HD International
WeHaveAFace.org
The Dickenson's Catalyst Fund

Title: Use of dCas9 epigenetic editors for long-term and allele-specific downregulation of Huntingtin

Authors: *J. WALDO[1], J. A. HALMAI[1], J. L. CARTER[1], D. L. CAMERON[1], I. VILLEGAS[1], J. NOLTA[2], K. FINK[1];
[1]Ctr. for Interventional Genetics, MIND Institute, Stem Cell Program and Gene Therapy Center, Inst. for Regenerative Cures and Dept. of Neurology, Univ. of California Davis Hlth. Systems, Sacramento, CA; [2]Ctr. for Interventional Genetics, MIND Institute, Stem Cell Program and Gene Therapy Center, Inst. for Regenerative Cures, Univ. of California Davis Hlth. Systems, Sacramento, CA

Abstract: Huntington's disease (HD) is a rare, autosomal dominant neurodegenerative disorder caused by a trinucleotide expansion in exon 1 of the Huntingtin gene (HTT), which leads to neuronal dysfunction and cell death. Healthy HTT is implicated in axonal trafficking and trophic factor regulation, making targeted allele-specific reduction important. This study aimed to target single nucleotide polymorphisms (SNPs) in HD patient-derived cells to reduce the expression of mutant HTT using CRISPR epigenome editing. Heterozygous SNPs near regulatory regions of the HTT promoter were confirmed in HD patient cells, allowing for the design of allele-specific gRNAs for both dxCas9 and dCas9-VQR. gRNA screen was initially conducted in patient-derived fibroblasts, followed by assessment in patient-derived iPSC-derived neural stem cells. Knockdown was assessed at multiple loci in the HTT gene and significant downregulation was achieved using several of our gRNAs that were designed to be allele-specific. We also assessed

HTT knockdown using different effector domains, including those that induce histone methylation as well as DNA methylation. We saw significant downregulation of HTT in both patient-derived fibroblasts as well as iPSC-derived neural stem cells. We also assessed the length of downregulation using a potent DNA methylator, DNMT3A/L. We assessed knockdown over a 21-day period to determine the durability of knockdown at the HTT locus, as well as changes in DNA methylation at a nearby CpG island using bisulfite sequencing. Future directions include gRNA modifications to increase allelic discrimination as well as assessment of functional recovery in HD patient-derived NSCs and neurons.

Disclosures: **J. Waldo:** None. **J.A. Halmai:** None. **J.L. Carter:** None. **D.L. Cameron:** None. **I. Villegas:** None. **J. Nolta:** None. **K. Fink:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.21

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

Support: The CHDI Foundation
NIH/NINDS (1 R01 NS100802)
The Broderick fund for Phytocannabinoid Research at MIT
The JPB Foundation
NIH (R25MH101076)
NIH (5T32EB019940-05)
NIH NIA NIMH NINDS NHGRI (R01AG062335,R01AG058002, RF1AG054012, RF1AG062377, R01MH109978, U01MH119509, U01NS110453, andR01HG008155)

Title: Cell Type-Specific Transcriptomics Reveals that Mutant Huntingtin Leads to Mitochondrial RNA Release and Neuronal Innate Immune Activation

Authors: *H. LEE[1,2], R. J. FENSTER[1], S. S. PINEDA[3,2], W. S. GIBBS[5], S. MOHAMMADI[3,2], J. DAVILA-VELDERRAIN[3,2], F. J. GARCIA[4], M. THERRIEN[2], H. S. NOVIS[5], F. GAO[1,6], H. A. WILKINSON[7], T. VOGT[7], M. KELLIS[3,2], M. J. LAVOIE[5], M. HEIMAN[1,4,2];
[1]Picower Inst. for Learning and Memory, Cambridge, MA; [2]Broad Inst. of MIT and Harvard, Cambridge, MA; [4]Dept. of Brain and Cognitive Sci., [3]MIT, Cambridge, MA; [5]Ann Romney Ctr. for Neurologic Dis., Brigham and Women's Hosp. and Harvard Med. Sch., Boston, MA; [6]Bioinformatics Resource Ctr., The Beckman Inst. at Caltech, Pasadena, CA; [7]CHDI Management, Inc./CHDI Fndn., Princeton, NJ

Abstract: Huntington’s disease (HD) is a fatal neurodegenerative disorder caused by CAG trinucleotide repeat expansions in the huntingtin (*HTT*) gene. Although mutant huntingtin (*mHTT*) has been linked to both toxic gain-of-function and loss-of-function effects, it is still not

fully understood how *mHTT* leads to the death of striatal spiny projection neurons, the most vulnerable cell type in HD. To gain new molecular insights, we conducted a large-scale cell type-specific transcriptomic profiling study across both human HD and mouse models of HD at various stages of disease progression, using two complementary techniques: cell type-specific Translating Ribosome Affinity Purification (TRAP-seq, a methodology used to purify ribosome-bound mRNAs in bulk from a specific cell type) and single nuclear RNA sequencing (snRNA-seq, a methodology used to capture nuclear RNAs at the single-cell level).
Our systematic analyses of caudate/putamen (striatal) cell type-specific gene expression changes in human HD and mouse models of HD revealed a large number of both non-cell type-specific and cell type-specific responses that are induced by *mHTT*. Among these we observed the release of mitochondrial RNA (mtRNA, a potent mitochondrial-derived innate immunogen) and a concomitant upregulation of innate immune signaling in *Drd2*-expressing striatopallidal "indirect pathway" spiny projection neurons (iSPNs, the most vulnerable cell type in HD). Normally these mtRNAs are sequestered inside the mitochondria. In response to *mHTT*, however, mtRNAs were released from SPN mitochondria into the cytoplasm. When released to the cytosol, mitochondrial nucleic acids can be sensed by various innate immune sensors, including the double-stranded RNA-dependent protein kinase PKR (which senses mtRNA) or cGAS-STING (which senses mtDNA), which can then trigger downstream innate immune responses that may lead to cell death. The iSPN-enhanced release of mtRNA correlates with *mHTT* CAG repeat length, is associated with disease model age, and even occurs at very early stages of HD model progression when the level of gene expression dysregulation is small. Our work reveals a new mechanism that may contribute to *mHTT* toxicity in HD and points to new therapeutic opportunities.

Disclosures: H. Lee: None. **R.J. Fenster:** None. **S.S. Pineda:** None. **W.S. Gibbs:** None. **S. Mohammadi:** None. **J. Davila-Velderrain:** None. **F.J. Garcia:** None. **M. Therrien:** None. **H.S. Novis:** None. **F. Gao:** None. **H.A. Wilkinson:** None. **T. Vogt:** None. **M. Kellis:** None. **M.J. LaVoie:** None. **M. Heiman:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.22

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

Title: The investigation of stress susceptibility in Huntington Disease neurons following HTT lowering treatment

Authors: *J. AQUINO[1], E. MACHIELA[1], N. POTLURI[1], R. JELOKA[1], M. ROOK[1], Y. XIE[1], A. SOUTHWELL[2];
[1]Burnett Sch. of Biomed. Sci., [2]Col. of Med., Univ. of Central Florida, Orlando, FL

Abstract: Huntington Disease (HD) is an autosomal dominant progressive neurodegenerative disorder caused by a CAG tract expansion in exon 1 of the *HTT* gene to greater than 35 repeats.

HD patients live with the genetic determinant before experiencing symptoms typically in mid-life. Conditional genetic inactivation of the mutant HTT (*muHTT)* gene can reverse HD-like phenotypes in mice. This led to the development of therapies for non-selective HTT lowering or selective muHTT lowering, and these methods have shown preclinical efficacy. Unfortunately, the first HTT lowering clinical trial was halted due to worsening clinical outcomes. While it is unclear if this was the result of non-specific toxicity or too much total HTT loss, this demonstrates the need to further explore the consequences of lowering HTT and improve preclinical assessments. HTT is a vital protein that can counteract cellular stress, and muHTT interferes with proper stress response. Furthermore, most HD patients receive treatment in adulthood, and age causes additional decline in cellular stress resistance. Considering that current preclinical studies are performed in models that are not aged or challenged with stress or comorbidities, they may not accurately reflect the tolerability of experimental therapies in the aging HD brain. To address this, we are attempting to better model clinical HTT lowering in aging adults that encounter stressors and determine if HTT lowering alters stress resistance. Using primary neurons from HD mice, we have induced age-like changes using progerin, the mutant protein that causes Hutchinson Gilford Progeria syndrome. We have found that induced-aged HD neurons have an impaired ability to counteract exogenous oxidative stress and altered levels of proteins involved in response to oxidative stress, DNA damage, and hypoxia. Using this system, we have tested induced-aged neurons that underwent non-selective or selective HTT suppression followed by oxidative stress, allowing us to interrogate stress susceptibility in mature 'aged' neurons post-HTT lowering. We found that while both selective and non-selective HTT lowering provides protection from HD-like changes in cells, neurons with robust non-selective HTT lowering are more susceptible to age and stress-induced cell death. This increased susceptibility was not observed in cells after a more modest ~50% reduction of total HTT. These data suggest that in a nonselective approach, the benefit of lowering mutant HTT can outweigh potential detriment from loss of total HTT up until some threshold of total HTT loss, beyond which stress resistance is compromised.

Disclosures: **J. Aquino:** None. **E. Machiela:** None. **N. Potluri:** None. **R. Jeloka:** None. **M. Rook:** None. **Y. Xie:** None. **A. Southwell:** None.

Poster

533. Huntington's Disease : Molecular Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 533.23

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

Support: BrightFocus Foundation
R03 NS084386
R03 NS092024

Title: Huntingtin during axonal injury: A retrogradely co-migrating Huntingtin-Rab7-LAMP1-containing signaling endosome.

Authors: *S. GUNAWARDENA;
SUNY At Buffalo, Buffalo, NY

Abstract: Huntingtin (HTT), the protein involved in Huntington's disease is a 350kDa protein of unknown function. Despite more than 350 binding partners identified for HTT across a wide range of cellular processes, including trafficking, the molecular mechanisms by which HTT and its binding partners function remain elusive. Here we provide evidence for a retrogradely moving HTT-Rab7 vesicular complex using *Drosophila* genetics, *in vivo* imaging in living axons coupled with a custom particle tracking analysis, and pharmacological inhibitors. We identify that adaptors HIP1 and RILP aid the retrograde motility of LAMP-1 containing HTT-Rab7 late endosomes, but not autophagosomes. Reduction of Syntaxin17 and Chloroquine/BafilomycinA1-mediated pharmacological inhibition, but not reduction of ATG5, disrupted the *in vivo* motility of these vesicles. The retrogradely moving HTT-Rab7-LAMP1-containing late endosome can traffic long-distance signaling components such as the BMP-receptors TKV/WIT, neurotrophic factor BDNF and axonal damage response components WND/DLK and JNK following axonal injury. Taken together, our observations unravel a previously unknown role for HTT in the retrograde movement of a Rab7-LAMP1-containing signaling late endosome, which has functional relevance during axonal injury.

Disclosures: S. Gunawardena: None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.01

Topic: C.06. Neuromuscular Diseases

Support: KAKENHI JP 19K07970

Title: Emodin alleviates SBMA phenotype via down regulation of androgen receptor

Authors: *H. ADACHI[1], Q. QIANG[3], Z. HUANG[2], T. TOYOTA[1], M. KATSUNO[4], G. SOBUE[5];
[2]Neurol., [1]Univ. of Occup. and Envrn. Hlth., Kitakyushu, Japan; [3]Dept. of Neurology, Huadong Hosp., Shanghai, China; [4]Nagoya Univ. Grad Sch., Nagoya, Aichi, Japan; [5]Nagoya Univ. Sch. of Med., Nagoya, Japan

Abstract: Spinal and bulbar muscular atrophy (SBMA) is an x-linked hereditary neuromuscular disorder caused by the expansion of CAG repeats in the first exon of androgen receptor (AR) gene. The expanded CAG repeats encode a long polyglutamine tract in the N-terminal transactivation domain of AR protein. SBMA characterized clinically by limb weakness, fasciculation, and muscle atrophy. The pathologic feature of SBMA is selectively loss of motor neurons in the anterior horn of spinal cord and the bulbar region of brain stem, diffusely-accumulated mutant AR in the nuclei of the residual motor neurons. Because the expanded CAG

repeats in *AR* gene are likely linked to the toxic gain of function, decreasing the amounts of mutant AR protein is becoming an optional therapeutic avenue for the SBMA treatment.We examined the effects of emodin in cultured-cell models of SBMA. Cells were transfected using Lipofectamine 2000 with plasmids encoding mutant AR containing normal (24 CAGs) or expanded (97 or 112 CAGs) polyQ repeats. 4 or 40 mg/kg of emodin was administered intraperitoneally every day. Control mice received DMSO.We demonstrated that emodin, a natural compound extracted from Chinese herbs, could decrease the association of mutant AR with heat shock protein 90 (Hsp90) without blocking ATP binding, which in turn destabilized mutant AR protein through the proteasome pathway. Intraperitoneal injection of emodin into SBMA transgenic mice showed improved motor performance, alleviated body weight loss and enhanced survival rate in comparison with vehicle-treated SBMA transgenic mice. Moreover, emodin treatment also attenuated spinal cord and muscle pathology, decreased the monomeric and aggregated mutant AR protein levels in the tissues of SBMA transgenic mice.Collectively, these results indicate that emodin treatment might be a possible therapeutic approach for the manifestations of SBMA.

Disclosures: **H. Adachi:** None. **Q. Qiang:** None. **Z. Huang:** None. **T. Toyota:** None. **M. Katsuno:** None. **G. Sobue:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.02

Topic: C.06. Neuromuscular Diseases

Title: Relative contribution of skeletal muscle vs. motor neurons to neuromuscular decline in Spinal and Bulbar Muscular Atrophy

Authors: *N. NGUYEN[1], A. GROMOVA[1], B. CHA[1], C. CORTES[2], A. R. LA SPADA[1]; [1]Pathology, Univ. of California, Irvine, Irvine, CA; [2]Neurol., Univ. of Alabama At Birmingham, Vestavia Hills, AL

Abstract: Spinal and Bulbar Muscular Atrophy (SBMA) is a rare, X-linked neuromuscular disease caused by a CAG repeat expansion mutation in the Androgen Receptor gene (polyQ-AR). Previous work in our lab has generated a mouse model of SBMA in which the human AR gene with 121 CAG repeats is engineered to contain loxP sites around the first exon (fxAR121 mice) so that the disease causing gene can be excised in a tissue-specific manner by crossing to mice expressing Cre recombinase in disease-relevant tissues, such as skeletal muscle and motor neurons. The transgene construct was derived from a Bacterial Artificial Chromosome, thus also contains native regulatory elements to ensure physiological expression levels. This is important because it is known that supraphysiological expression of just wildtype AR is sufficient to cause neuromuscular disease in mice. We have established that fxAR121 mice recapitulate hallmarks of SBMA, including progressive muscle atrophy with prominent polyQ-AR aggregation and

degeneration of lower motor neurons, and die prematurely due to these phenotypes. Excising polyQ-AR only from skeletal muscle by crossing with mice that express Cre recombinase only in skeletal muscle (HSA-Cre/fxAR121 mice) is sufficient to rescue all neuromuscular phenotypes, even degeneration of motor neurons, despite retained expression of polyQ-AR in motor neurons. Here, we report latent phenotypes in HSA-Cre/fxAR121 mice aged out to 2 years, long past the lifespan of globally-expressing fxAR121 mice. We show that HSA-Cre/fxAR121 mice die by 22 months due to cardiomyopathy, a pathological feature that is just beginning to be recognized in SBMA patients. Lastly, we report findings of mice in which polyQ-AR was excised from motor neurons using 2 different motor neuron Cre drivers, VAChT-Cre and Hb9-Cre. Surprisingly, these mice do not show significant abrogation of disease phenotypes, further highlighting the prominent role of polyQ-AR expression in skeletal muscle as driving neuromuscular decline in SBMA, including degeneration of motor neurons.

Disclosures: **N. Nguyen:** None. **A. Gromova:** None. **B. Cha:** None. **C. Cortes:** None. **A.R. La Spada:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.03

Topic: C.06. Neuromuscular Diseases

Support: FRRB Grant CP2_20/2018

Title: An integrated approach for investigating the pathomechanisms underlying neurodegeneration in SCA2 and MSA-C

Authors: M. RIZZUTI[1], M. NIZZARDO[1], S. SALANI[1], V. MELZI[1], L. BRAMBILLA[1], S. MARCUZZO[2], S. MAGRI[3], C. MARIOTTI[3], C. CORDIGLIERI[4], A. DI FONZO[1,5], G. COMI[1,5,6], F. TARONI[3], *S. CORTI[1,5];
[1]Fndn. IRCCS Ca' Granda Ospedale Maggiore Policlinico, Neurol. Unit, Milan, Italy; [2]Neurol. IV - Neuroimmunology and Neuromuscular Dis. Unit, Fondazione Inst. Neurologico Carlo Besta, Milan, Italy; [3]U.O.C. Genetica Medica e Neurogenetica, Dept. di Diagnostica e Tecnologia Applicata, Fondazione IRCCS Inst. Neurologico Carlo Besta, Milan, Italy; [4]Inst. Nazionale di Genetica Molecolare Romeo ed Enrica Invernizzi, Milan, Italy; [5]Dino Ferrari Center, Dept. of Physiopathology and Transplants, Univ. of Milan, Milan, Italy; [6]Fndn. IRCCS Ca' Granda Ospedale Maggiore Policlinico, Neuromuscular and Rare Dis. unit, Milan, Italy

Abstract: Rare neurological diseases are a group of neurodegenerative disorders characterized by the selective degeneration of specific neuronal populations including cortical neurons (CNs). The complexity of the human brain has made difficult to study brain disorders, pointing out the need for *in vitro* models of human brain degeneration. The combination of 2D and 3D models is the most promising approach for studying human CNS disease, providing robust and consistent

phenotypes useful to increase knowledges in the perspective of a clinical translation.Here, we took advantage of optimized iPSC-derived CNs and brain organoid models to investigate the pathological mechanisms underlying Cerebellar Ataxia associated with Ataxin-2 expansion (SCA2) and Multisystem Atrophy with a cerebellar phenotype (MSA-C).We generated patient-specific iPSCs of MSA-C, SCA2 and healthy subjects. We differentiated them in CNs. The repertoire of iPSC-derived CN subtypes was assayed by immunocytochemistry, qPCR and bulk RNA-sequencing. In addition, we assessed the neuropathological features of iPSC-derived CNs using live tracking of individual neurons. Finally, we employed calcium imaging and electrophysiological studies to investigate the neural activity.The potential to achieve the translational step by generating new knowledge requires reliable disease model for therapeutic targets identification. In this context, brain organoids offer the unprecedented opportunity to study brain functions and features of complex human diseases that affect different cell types, their interactions and neuronal circuits. We generated patient-specific iPSC-derived brain organoids of MSA-C, SCA2 and healthy subjects. Mini brain organoids were deeply characterized by immunocytochemistry, qPCR and single cell sequencing. Functional and neuropathological features of our 3D cultures were assessed through calcium imaging and electrophysiological studies.Lastly, we explored the use of antisense oligonucleotides (ASO) designed with a Morpholino chemistry to modulate ATXN2 expansion and improve the pathological phenotype. Indeed, although there are currently no approved disease-modifying treatments for these two diseases, RNA-targeted therapies are promising for neurodegenerative disorders. Establishing new human reliable SCA2 models can be helpful for optimizing this antisense therapeutic strategy. On the other hand, MSA pathogenesis is still puzzling and comparing data obtained in 2D/3D MSA models with that originated from SCA2, may provide meaningful information for MSA pathogenesis and therapeutic development.

Disclosures: **M. Rizzuti:** None. **M. Nizzardo:** None. **S. Salani:** None. **V. Melzi:** None. **L. Brambilla:** None. **S. Marcuzzo:** None. **S. Magri:** None. **C. Mariotti:** None. **C. Cordiglieri:** None. **A. Di Fonzo:** None. **G. Comi:** None. **F. Taroni:** None. **S. Corti:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.04

Topic: C.06. Neuromuscular Diseases

Support: Support for this study was provided by CMU Neuroscience Program, CMU College of Medicine, the John G. Kulhavi Professorship in Neuroscience, and the E. Malcolm Field and Gary Leo Dunbar Endowed Chair in Neuroscience at CMU

Title: Huntingtin gene editing using CRISPR-Cas9 system to alleviate cognitive deficits in the YAC128 mouse model of Huntington’s disease

Authors: *S. KONERU[1,2,3], A. POUDEL[1,2,3], E. CRESPO[1,5,6], N. WEDSTER[1,2,3], E. LAUZON[1,2,3], J. WASSELL[1,2,3], R. SCHALAU[1,2,3], C. BUENO ALVAREZ[1,2,3], N. SHARMA[1,4], U. HOCHGESCHWENDER[1,3,5], J. ROSSIGNOL[1,2,3,5], G. L. DUNBAR[1,2,3,7];
[1]Central Michigan Univ., Mount Pleasant, MI; [2]Neurosci., Field Neurosciences Inst., Ascension St. Mary's, Saginaw, MI; [3]Neurosci., [4]Sch. of Hlth. Sci., Program in Neurosci., Mount Pleasant, MI; [5]Central Michigan Univ. Col. of Med., Mount Pleasant, MI; [6]Program in Biochemistry, Cell. and Mol. biology program, Mount Pleasant, MI; [7]Dept. of Psychology, Mount Pleasant, MI

Abstract: Huntington's disease (HD) is an autosomal dominant neurodegenerative disease caused by mutations in the huntingtin (HTT) gene containing a long polyglutamine (CAG) stretch, on chromosome 4. These mutations lead to production and accumulation of mutant huntingtin (mHTT) protein aggregates, which cause motor, and cognitive impairments. This increased amounts of mHTT protein undergoes inappropriate post-translational modifications that accumulate in neurons and leads to death. Currently, no disease-altering treatment has been approved for HD. However, the advent of increasingly sophisticated gene-editing tools, like CRISPR-Cas9, provides new approaches to knock down or block the transcription of mHTT. In our study, we regulated the abnormal production of mHTT, by constructing two CRISPR-Cas9 plasmids with gRNAs that target the open reading frame (uORF) of 5′ UTR. We injected the adeno-associated virus encapsulated CRISPR-Cas9 system intracranially, in the striatum of YAC128 mice to investigate its gene-silencing ability. This mouse model is genetically modified to mimic the human HD condition, having the full-length human HTT gene in one of their alleles. Assessments of motor deficits (using the rotarod task) and cognitive deficits (using the water-T-maze task) were performed in CRISPR-treated-, vehicle-treated- YAC128 mice and vehicle-treated wild type mice. Our results indicated that gene silencing with CRISPR-Cas9 did not reduce motor deficits on the rotarod task but reduced cognitive deficits on the water-T-maze task. Western blot results indicated a significant HTT and GFAP protein reductions in the striatum of CRISPR-treated mice, suggesting a reduction in HTT aggregates and neuroinflammation, respectively. These results provide a proof-of-concept that CRISPR-Cas9 can be used to effectively reduce HTT by targeting uORF of 5′ UTR.

Disclosures: S. Koneru: None. **A. Poudel:** None. **E. Crespo:** None. **N. Wedster:** None. **E. Lauzon:** None. **J. Wassell:** None. **R. Schalau:** None. **C. Bueno Alvarez:** None. **N. Sharma:** None. **U. Hochgeschwender:** None. **J. Rossignol:** None. **G.L. Dunbar:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.05

Topic: C.06. Neuromuscular Diseases

Support: NINDS Competitive Postdoctoral Fellowship Award / Intergovernmental Personnel Act Agreement
Kennedy's Disease Association Research Grant

Maryland Stem Cell Research Fund Launch Award #135408
Intramural Research Program of the NINDS, National Institutes of Health

Title: Ligand-free mitochondria-localized mutant AR interacts with F-ATP synthase to promote motor neuron-related mitochondrial dysfunction in SBMA

Authors: *X. FENG[1,2,3], X.-T. CHENG[4], P. ZHENG[2,3,6], Y. LI[5], J. HAKIM[3], S. Q. ZHANG[7], S. M. ANDERSON[8], K. LINASK[9], J. ZOU[9], Z.-H. SHENG[4], C. BLACKSTONE[2,3,6];
[1]Psychiatry and Behavioral Sci., Johns Hopkins Univ., Baltimore, MD; [2]Cell Biol. Section, [3]Neurogenetics Br., [4]Synaptic Function Section, [5]Protein/Peptide Sequencing Facility, Natl. Inst. of Neurolog. Disorders and Stroke, Bethesda, MD; [6]Movement Disorders Division, Dept. of Neurol., Massachusetts Gen. Hospital, Harvard Med. Sch., Charlestown, MA; [7]River Hill High Sch., Clarksville, MD; [8]Flow Cytometry Core, Natl. Human Genome Res. Inst., Bethesda, MD; [9]iPSC Core, Natl. Heart, Lung, and Blood Inst., Bethesda, MD

Abstract: Spinal bulbar muscular atrophy (SBMA) is an X-linked neuromuscular disorder involving CAG-repeat-expansion mutations in the androgen receptor (AR) gene. We utilized CRISPR-Cas9 gene editing to engineer novel isogenic human induced pluripotent stem cell (hiPSC) models, consisting of isogenic AR knockout, control, and disease lines expressing mutant AR with distinct repeat lengths, as well as control and disease lines expressing FLAG-tagged wildtype and mutant AR, respectively. Adapting a small-molecule cocktail-directed approach, we differentiate the isogenic hiPSC models into motor neuron-like cells with a highly enriched population to uncover cell-type-specific mechanisms underlying SBMA and to distinguish gain- from loss-of-function properties of mutant AR in disease motor neurons. We demonstrate that ligand-free mutant AR causes drastic mitochondrial dysfunction in neurites of differentiated disease motor neurons due to gain-of-function mechanisms, and such cytotoxicity can be amplified upon ligand (androgens) treatment. We further show that ligand-free mitochondria-localized mutant AR interacts with F-ATP synthase, and such interaction is associated with compromised mitochondrial respiration and multiple other mitochondrial impairments in disease motor neuron-like cells. These findings counter the established notion that androgens are requisite for mutant AR-induced cytotoxicity in SBMA, and point to vital contributions of mitochondrial dysfunction, mechanistically involving a preferential interaction between ligand-free mutant AR and F-ATP synthase, as a causative factor for motor neuron degeneration in SBMA.

Disclosures: X. Feng: None. **X. Cheng:** None. **P. Zheng:** None. **Y. Li:** None. **J. Hakim:** None. **S.Q. Zhang:** None. **S.M. Anderson:** None. **K. Linask:** None. **J. Zou:** None. **Z. Sheng:** None. **C. Blackstone:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.06

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

Support: NIH RO1 grant NS113612
Hereditary Disease Foundation

Title: Fan1 knockdown and Fan1-R510H knockin variant modify disease phenotypes in Q140 knockin mice

Authors: *L. DENG[1,2], J. RICHMAN[1], K. TAMAI[2], N. WANG[1], P. LANGFELDER[1], F. GAO[1,2], G. COPPOLA[1,2], C. S. COLWELL[2], X. W. YANG[1,2];
[1]Semel Inst. of Neurosci. and Human Behavior, [2]Psychiatry and Biobehavioral Sci., UCLA, Los Angeles, CA

Abstract: In Huntington's Disease (HD), the mutant Huntingtin (mHtt) CAG repeat length is inversely correlated with the age of onset of motor symptoms. However, this CAG repeat length does not account for all of the variance in HD onset, suggesting other genetic and environmental factors may be involved. Recent Genome Wide Association Studies (GWAS) revealed several loci that are significantly associated with modifying age of onset in HD. Several modifier alleles were identified on chromosome 15 adjacent to FAN1, which encodes a DNA repair protein involved in repairing interstrand cross-links (ICLs) and may also be participated in mismatch repair (MMR) through its interaction with MLH1. Here we undertook a mouse genetic approach to generate mutant mice with knockdown of murine Fan1 (reduced expression about 70%; Fan1-KD) or R510H knockin in Fan1 (Fan1-KI). The latter allele is analogous to the R507H coding variant in human FAN1 that is associated with hastened disease onset by about five years. We crossed these homozygous Fan1-KD and Fan1-KI alleles onto the mHtt knockin mice with 140 CAG repeats (Q140), and obtained two cohorts of mice with all the key genotypes. We analyzed aged 6-month old mice for their locomotor behaviors, neuropathology, mHtt CAG repeat instability, and transcriptomes. Overall, we see evidence of exacerbated behavioral impairment especially in the Q140-Fan1 KI/KI mice, enhanced striatal and liver repeat instability especially in Q140-Fan1 KD/KD (homo) mice, enhanced neuropathology in both Fan1-KI/KI and KD/KD mice crossed to Q140. Intriguingly, we did not observe marked changes in the mHtt-dysregulated striatal transcriptomes. Together, our study reveals Fan1-KD and KI alleles both exacerbate mHtt-induced striatal pathogenesis and uncovers an unexpected separation of modification of repeat instability and disease pathology from that of transcriptionopathy.

Disclosures: L. Deng: None. **J. Richman:** None. **K. Tamai:** None. **N. Wang:** None. **P. Langfelder:** None. **F. Gao:** None. **G. Coppola:** None. **C.S. Colwell:** None. **X.W. Yang:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.07

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

Support: Hereditary Disease Foundation
Lundbeck Foundation

Title: Bdnf/noggin induced striatal neurons integrate into the motor circuitry in a huntington's disease mouse model

Authors: J. C. CANO[1], A. BENRAISS[1], A. TATE[1], C. MANGIAMELE[1], *S. GOLDMAN[2,3]; [1]Univ. of Rochester Med. Ctr., Rochester, NY; [2]Univ. Rochester Med. Ctr., Rochester, NY; [3]Fac. of Hlth. and Med. Sci., Univ. of Copenhagen, Copenhagen, Denmark

Abstract: Huntington's disease (HD) is a progressive neurodegenerative disease characterized by the selective loss of neostriatal medium spiny neurons (MSNs). Intraventricular delivery of BDNF and noggin, whether by viral transduction or protein delivery, induces heterotopic recruitment of new MSNs from subventricular zone neural progenitor cells of normal adult mice, rats and monkeys. This process can also be elicited in R6/2 mice, a transgenic mouse model of HD whose survival is extended by BDNF/noggin delivery. Yet despite their clear functional contribution, it has remained unclear if newly generated striatal neurons fully integrate into striatal circuits. Here, using a transgenic mouse reporter for neural stem cells, Nestin-CreER^{T2}, both as wild-type (WT-Nestin) and bred to R6/2-120Q mice (R6/2-Nestin), we evaluated the circuit integration of new, striatal neurons induced by BDNF and noggin administration in young adult mice. At 5 weeks of age, both WT- and R6/2-Nestin mice were infused with intraventricular BDNF and noggin proteins for 2 weeks and treated with tamoxifen for 10 days beginning at 6 weeks of age. Using Cre-dependent rabies tract-tracing, we found that newly generated MSNs receive inputs from cortex, thalamus and substantia nigra, equally so in WT and R6/2 mice. We then evaluated whether the new neurons project functional efferents to the globus pallidus, using trans-synaptic serial tracing. We found that the new striatal neurons integrated into the cortico-striato-pallidal circuit in both WT and R6/2 mice. To evaluate whether cortical inputs elicit post-synaptic activity in new striatal neurons, we then used optogenetic stimulation of cortical neurons in slice preparations, while recording post-synaptic events and calcium signals in the striatum. Following BDNF-Noggin infusion, mice were injected intrastriatally with a rabies virus encoding Channelrhodopsin-2 for cortical stimulation, and a lentiviral Cre-dependent GCaMP7 to image striatal calcium events. Cortical stimulation elicited calcium activity and excitatory post-synaptic events in the new striatal neurons in both WT and R6/2 mice, with no significant difference between the groups (N=6 mice/group). These findings indicate that new striatal neurons generated in adulthood in response to BDNF and noggin can functionally integrate into adult striatal networks, thus highlighting the potential of induced neuronal addition as a means of circuit restoration in HD.

Disclosures: J.C. Cano: None. **A. Benraiss:** None. **A. Tate:** A. Employment/Salary (full or part-time):; Sana Biotechnology. **C. Mangiamele:** None. **S. Goldman:** A. Employment/Salary (full or part-time):; Sana Biotechnology. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Sana Biotechnology.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.08

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

Support: Association Huntington France

Title: Developmental defects of striatal D2-expressing medium spiny neurons in a mouse model of Huntington's disease

Authors: *M. LEBOUC[1], L. BONAMY[1], Q. RICHARD[1], M. GARRET[2], J. BAUFRETON[1]; [1]CNRS UMR 5293, Inst. des Maladies Neurodégénératives, Bordeaux, France; [2]CNRS UMR 5287, Inst. de Neurosciences Cognitives et Intégratives d'Aquitaine, Bordeaux, France

Abstract: Huntington's disease (HD) is an inherited neurodegenerative disorder that typically occurs in midlife with progressive alterations of motor and cognitive functions. This disease is due to the mutation of the gene encoding the Huntingtin protein (Htt) and leads to a severe neurodegeneration in the striatum and cortex. Even if the implication of the Htt mutation in HD is well known, recent studies suggest that Htt mutation is also linked to developmental impairment. Indeed early striatal developmental alterations as hypertrophy and impaired cytoarchitecture have been observed in humans or in rodent models of HD. However this issue remains largely unexplored and these findings raise the question of when do the first disease-related striatal alterations emerge in the disease. To answer this question, we are longitudinally comparing the striatal development between wild type (WT) and the R6/1 mouse model of HD. This mouse line is crossed with D1-GFP or D2-GFP mice in order to discriminate between direct-pathway (D1-expressing) and indirect-pathway (D2-expressing) MSN subpopulations. This study is performed during the two first postnatal weeks as this period has been shown to be crucial for the maturation of MSN's morphological and electrophysiological properties. Using ex vivo whole-cell patch clamp electrophysiology we are recording the intrinsic electrophysiological properties of MSNs as well as the establishment of the cortico-striatal glutamatergic transmission. We are also looking at the morphology of the neurons recorded, especially their dendritic length and complexity and their dendritic spines. Our results suggest that there is a specific alteration of D2-MSNs properties in R6/1 mice highlighted by an early decreased excitability and higher dendritic complexity at postnatal day (P)0-3. These anatomical and electrophysiological data provide an insight into striatal developmental alterations in a mouse model of HD at very early stages.

Disclosures: **M. Lebouc:** None. **L. Bonamy:** None. **Q. Richard:** None. **M. Garret:** None. **J. Baufreton:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.09

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

Support: CHDI Foundation

Title: Indirect striatal projection neuron activity in Q175 Huntington's disease mice

Authors: *E. LARA-GONZALEZ, J. W. CALLAHAN, M. D. BEVAN;
Neurosci., Feinberg Sch. of Medicine, Northwestern Univ., Chicago, IL

Abstract: Huntington Disease (HD) is an autosomal dominant neurodegenerative disorder caused by an expansion of trinucleotide repeats in exon 1 of the huntingtin gene. HD is characterized by the progressive degeneration of the basal ganglia and cerebral cortex and associated psychomotor dysfunction. Although, the terminal stage of HD is associated with the widespread degeneration of the basal ganglia and cortex, neurons in the indirect pathway of the basal ganglia exhibit the earliest and most profound susceptibility. Therefore, dysfunction of the indirect pathway is likely to be a major contributor to motor dysfunction in HD. To better understand this linkage, we recorded the activity of indirect pathway striatal projection neurons (iSPNs) in head-fixed wild type and Q175 HD mice during rest and spontaneous self-initiated locomotion on a linear treadmill. Neuronal activity was recorded using 32-64 channel silicon optrodes. iSPNs were identified through optogenetic stimulation of ChR2(H134R) that was virally expressed in a Cre recombinase-dependent manner in wild type and Q175 mice expressing Cre recombinase in A2A receptor-expressing neurons. We found differences between the neuronal activity of iSPNs in wild type and Q175 mice, both at rest and during bouts of locomotion. We are currently determining whether this abnormal iSPN activity can be ameliorated through viral expression of zinc finger proteins that suppress the expression of mutant huntingtin in iSPNs. Thus, this research will inform our understanding of the cell-autonomous and circuit mechanisms that underlie psychomotor dysfunction in HD, and the therapeutic effectiveness of viral-based mutant huntingtin lowering strategies.

Disclosures: E. Lara-Gonzalez: None. **J.W. Callahan:** None. **M.D. Bevan:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.10

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

Support: CIHR Fdn-143210
CIHR Fdn-143209

Title: An automated homecage-based system to characterize long-term goal-directed motor learning in a mouse model of Huntington's Disease

Authors: *D. RAMANDI[1,2], T. H. MURPHY[1], L. A. RAYMOND[1];
[1]Djavad Mowafaghian Ctr. for Brain Hlth. and Dept. of Psychiatry, [2]Grad. Program in Cell and Developmental Biol., Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Understanding the neurological changes underlying motor learning is crucial for making advances in treating movement disorders like Huntington's Disease (HD). While many studies in the past have investigated motor skill learning (MSL), few studies have explored these changes in the long term. Therefore, in this study, we developed a skilled lever-pulling task within an automated home-cage testing system, assessing the ability of male and female wild-type (WT) and zQ175 HD 6-month old mice to learn the task over a period of several weeks. The engagement and changes in their performance were measured 24/7. In this self-directed behavioral task, animals learn to hold a lever for increasingly longer duration up to 1s. The required hold-time for a successful trial in this task is individually set for each animal daily, based on their performance in the previous day (75th percentile of hold-times of all the trials). Over several weeks of experimentation, WT animals showed a steady increase in the average hold-time of the lever, while zQ175 mice could not adjust to the increasing demand of the task. Interestingly, there were no genotype differences in the number of daily trials, or the time spent in the behavior chamber attached to the home-cage. Additionally, we observed an increased jerkiness of the pull trajectories in zQ175 mice, as shown by an elevated amplitude of high-frequency movements. The constant change in the required hold-time through the learning process partially eliminates the habituation process. This is validated by the lack of change in trial-to-trial correlation, which is an indicator of habit formation and motor variability. Overall, our findings suggest an impairment in fine motor learning and performance in HD mice at an early stage of the disease.
Funded by Canadian Institutes of Health Research Fdn-143210 to LAR and Fdn-143209 to THM.

Disclosures: D. Ramandi: None. **T.H. Murphy:** None. **L.A. Raymond:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.11

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

Support: Canadian Institutes of Health Research Fdn 143209
Canadian Institutes of Health Research Fdn 143210
Brain Canada Vectorology Foundry
Canada Vanier Scholarship Award

Title: Longitudinal monitoring of altered cortical mesoscale activity and water-reaching behavior in a Huntington Disease mouse model

Authors: *Y. WANG, M. D. SEPERS, D. XIAO, D. RAMANDI, L. A. RAYMOND, T. MURPHY;
Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Synaptic and circuit changes that precede progressive striatal medium spiny neuron (MSN) and cortical neuronal loss in Huntington Disease (HD) result in characteristic motor dysfunction, cognitive impairment, and neuropsychiatric symptoms. Presently, early-stage HD behavioral phenotyping in mouse models has largely been concentrated on gross motor and balance deficits with limited examinations into the forelimb 'reach-to-grasp' movement, which is used often in our daily lives. Here, we use a high-throughput Go/No-Go water reaching task to simultaneously monitor progressive changes in widefield GCaMP6 cortical activity and forelimb coordination impairment from ~5.5 to ~7.5 months of age, comparing female zQ175 and wildtype (WT) littermate mice. By Day 8, both groups successfully learned to perform the task with near perfect performance rates during Go trials; however, zQ175 mice had not learned to suppress reaching movement and related cortical activity during No-Go trials. A progressive decline in forelimb coordination and grasping defects characterized by first an increase in failed trials was apparent on Day 30 in zQ175 mice, progressing to limited task engagement by Day 60. In contrast, WT mice maintained near perfect success rates across the 67 testing days. Kinematic analysis of the paw using markerless pose estimation further revealed more variable reaching trajectories in zQ175 compared to WT mice. Correlated with the loss of forelimb motor control over time was an increase in peak amplitude of the cortical activity on Day 45 compared to Day 8 for zQ175 mice, whereas there was little to no significant change in peak amplitude of the cortical activity seen over time in WT mice. These early and manifest stages of HD respectively, characterized by the absence and presence of failed forelimb water reaching, was confirmed using the tapered beam and rotarod tests. An increased time to traverse the beam was seen for zQ175 mice compared to WT at 8 months but not 5 months of age. Decreased latency to fall from the rotarod was seen in zQ175 mice at 8 months of age compared to WT. Post-mortem immunohistochemistry staining for striatal MSNs revealed decreased DARPP-32 expression in zQ175 mice, confirming HD disease pathology. The water reaching task therefore provides a useful tool to inform disease onset, therapeutic intervention windows to test novel drugs and to assess face validity of various HD mouse models and those pertaining to other neurological diseases. *Funding provided by Canadian Institutes of Health Research Fdn 143209 to THM and Fdn 143210 to LAR, and from Brain Canada Vectorology Foundry (LAR and THM). YW is supported by a Canada Vanier Scholarship Award.*

Disclosures: Y. Wang: None. **M.D. Sepers:** None. **D. Xiao:** None. **D. Ramandi:** None. **L.A. Raymond:** None. **T. Murphy:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.12

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

Support: NIH Grant: NS106305

Title: Cms121 partially attenuates disease progression in the r6/2 and yac128 mouse models of Huntington's disease

Authors: *G. ATES[1,2], T. TAGUCHI[2], P. MAHER[2];
[1]Vrije Universiteit, Brussels, Vrije Universiteit, Brussels, Brussels, Belgium; [2]Cell. Neurobio., Salk Inst. for Biol. Studies, La Jolla, CA

Abstract: Huntington's disease (HD) is an autosomal dominant neurodegenerative disease for which no treatment exists. The flavonol fisetin has been shown to reduce the impact of mutant huntingtin in several HD disease models[1]. CMS121 is a derivative of fisetin, yet with an improved pharmacology, designed to treat diseases of the central nervous system characterized by neuroinflammation, oxytotic/ferroptotic cell death and proteotoxicity[2]. The current study aims at testing the therapeutic efficacy of CMS121 in two distinct mouse models for HD, one with a rapid (R6/2 mice) and one with a slower (YAC128 mice) disease progression.Male mice (n=18-20) were treated with 0 (control), 200 or 400ppm CMS121 in their chow at six weeks (R6/2) or two months (YAC128). Assignment to the treatment groups was in a random manner. Mice of the same background, not carrying the mutation in the Huntingtin gene, were used as healthy controls. The general health of the mice, including body weight, grip strength and motor function, were monitored at different time points throughout their lifespan. A subpopulation of the R6/2 mice was sacrificed at 14 weeks for RNAseq analysis.The results show a moderate decrease in disease progression in CMS121 treated mice in both models. Treatment with 200ppm CMS121 also tended to increase the median lifespan, especially in the R6/2 model. RNAseq analysis of the striatum of 14-weeks old R6/2 mice demonstrated a moderate yet significant treatment-induced reversal effect at the transcriptome level, with the proteasome and oxidative phosphorylation as the main molecular pathways affected by CMS121 treatment. Thus, while CMS121 slowed disease progression, it was not sufficient to stop it in the mouse models used in this study. This suggests that testing combinations of drug candidates that together target a wider range of pathways implicated in HD could be a very promising approach to treating this disease.
References:1. Maher et al. 2011 doi: 10.1093/hmg/ddq460 2. Chiruta et al. 2012 doi: 10.1021/jm 2012563

Disclosures: G. Ates: None. **T. Taguchi:** None. **P. Maher:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.13

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

Title: Laquinimod combined with an essential fatty acid-rich diet enhances the neuroprotective effect in an acute rodent model of Huntington's disease

Authors: *Q. ANGELES-LÓPEZ[1,2], A. MORALES[3], A. SÁNCHEZ -CHINCHILLAS[4], M. H. CERÓN, Sr.[5], F. PEREZ[3], J. V. SEGOVIA-VILA[6];
[1]Cinvestav, Ciudad de México, Mexico; [2]Neurofarmacología Mol. y Nanotecnología, Inst. Nacional de Neurología y Neurocirugía, Ciudad de México, Mexico; [3]Neurofarmacología Mol. y Nanotecnología, Inst. Nacional de Neurología y Neurocirugía, Mexico, Mexico; [4]Alimentos y Biotecnología, Univ. Nacional Autónoma de México, Ciudad de México, Mexico; [5]Inst. Nacional De Neurología Y Neurocirugía, Mexico, Mexico; [6]Cinvestav-IPN, Mexico 07300 DF, Mexico

Abstract: Oxidative stress caused by free radicals and altered signaling pathways are distinct characteristics observed in several neurodegenerative diseases, including Huntington's disease (HD). In HD patients, oxidative damage and neuroinflammation lead to neuronal dysfunction and synaptic loss, in both cortex and striatum. Since oxidative stress and neuroinflammation contribute to the onset and progression of HD, pharmacological modulators that block or reduce the progress of oxidative damage offer potential therapeutic options. Among these agents we find laquinimod that acts as inflammation modulator, promoting in turn the expression of brain-derived neurotrophic factor, and decreasing IL-6 levels in serum. Additionally, it is known that an essential fatty acid (EFA)-rich diet exerts a neuroprotective effect against the striatal damage induced by the intrastriatal injection of quinolinic acid (QA). The purpose of this work was to determine whether the combined effects of laquinimod with an EFA-rich diet, enhanced their individual neuroprotective effects in a HD model induced by QA. We independently constructed dose-effect curves for both laquinimod and EFA -rich diet as a pretreatment against QA-induced damage, later the optimal effective doses of both agents were combined. Rotatory behavior, beam balance performance and oxidative damage were evaluated. Our results show a synergic effect of the combined therapy, since there was a 45% reduction in rotatory behavior, beam balance performance was improved, and striatal oxidative damage was significantly diminished. We showed the neuroprotective effect that laquinimod combined with an EFA -rich diet induced in an acute model of HD.

Disclosures: Q. Angeles-López: None. **A. Morales:** None. **A. Sánchez -Chinchillas:** None. **M.H. Cerón:** None. **F. Perez:** None. **J.V. Segovia-Vila:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.14

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

Support: CHDI Foundation, Inc.
UCLA Neurobehavioral Genetics T32 Training Grant

Title: Novel conditional mouse genetic tools for Hap40 (F8a) to study its role in development and disease

Authors: *L. E. DIONISIO[1], N. WANG[2], M. PLASCENCIA[3], X. GU[5], X. YANG[4]; [2]Semel Inst. for Neurosci. and Human Behavior, [3]Semel, [4]Dept Psych, [1]UCLA, Los Angeles, CA; [5]Ctr. for Neurobehavioral Genet., Semel Inst. for Neurosci. & Hum. Behavior, Univ. of California Los Angeles, Los Angeles, CA

Abstract: Huntingtin Associated Protein, 40 kDa (HAP40), encoded by the F8a gene on the X-Chromosome, has been identified as a key interactor of Huntingtin. It forms a 1:1 stable complex with wildtype or mutant Huntingtin and the structure of such complexes can be resolved by CryoEM. However, the normal function of HAP40 and its role in HD pathogenesis remains elusive. Here we used CRISPR/Cas9 mediated genome editing to generate two mouse alleles to interrogate murine Hap40's roles in biology and diseases: one is a Cre/LoxP conditional knockout allele of endogenous Hap40 and the other is a conditional overexpression of Hap40 from the *Rosa26* locus. Since studies thus far showed endogenous HTT has a role in cortical development in human (Barnat et al., 2020, PMID: 32675289; Molina-Calavita et al., 2014, PMID: 25057205), we performed a proof-of-concept study to examine the role of Hap40 in murine cortical development. We crossed the X-linked conditional F8a allele with Emx1-Cre, which is expressed from embryonic day 10.5 onward in the lineage of all the cortical and hippocampal excitatory projection neurons and glial cell types, but without expression in the GABAergic interneurons or microglia. We will present preliminary data to show the postnatal neurodevelopmental phenotypes of complete loss of Hap40 (F8a) in the male cortex. Although the brains of the mutant mice appear indistinguishable from those of the wildtype controls at birth, we observed a robust cortical atrophy phenotype accompanied by gliosis at about 1-month of age. We are currently performing additional behavioral, pathological and transcriptomics studies to discern whether Hap40 is playing a role in neuronal differentiation or prevention of neurodegeneration. Moreover, we also plan to compare the phenotypes of Hap40 in forebrain neurons to those of endogenous murine Huntingtin. Together, our study provided novel conditional mouse genetic resources to study the normal biological function of Hap40 and its potential role in HD pathogenesis.

Disclosures: L.E. Dionisio: None. **N. Wang:** None. **M. Plascencia:** None. **X. Gu:** None. **X. Yang:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.15

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

Support: CIHR Fdn-143210

Title: Age and sex-specific motor and cognitive deficits in the Q175/B6 mouse model of Huntington's disease

Authors: *J. CHENG, E. T. KOCH, L. A. RAYMOND;
Psychiatry, Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Q175/B6 is a knock-in mouse model of Huntington's disease, a neurodegenerative disorder characterized by loss of motor control and cognitive deficits. The primary site of neurodegeneration in HD is the striatum, a brain region important for motor learning. Studies have found hypoactivity and abnormal rearing in older HD mice during the open field, as well as motor and cognitive deficits on the rotarod and water T-maze tasks at ~10 months of age. This study aims to conduct more detailed behavioural pattern analyses in 2- and 10-month-old male and female Q175 mice during these tasks using the DeepLabCut tracking software and the new Behavioral Segmentation of Open Field in DeepLabCut (B-SOiD) machine learning program. We also examined paw kinematics during open field behaviours and rotarod learning. Genotype and sex differences were revealed in the older group for open field behaviours, such as decreased locomotion and increased rearing in 10-month-old Q175 males only. Older male Q175 mice also showed a greater number of paw slips below the bottom of the rotarod on the rotarod task, while the female cohort only showed increased paw slips in the later days of rotarod testing. In the water T-maze, 2-month-old Q175 mice took longer than WT to reach the hidden platform during the reversal phase. For the older male Q175 animals, only those that used a striatum-dependent response learning strategy showed a longer latency to reach the platform. These findings reveal that Q175 mice show motor learning and coordination deficits at 10 months of age, as well as reduced exploratory behaviour and potentially increased anxiety-like behaviours in the open field. Younger Q175 mice may show early signs of impairment in cognitive flexibility, and the specific strategy used during motor learning may be more relevant at the older age. Future studies that examine striatal signalling using *in vivo* imaging techniques, such as fiber photometry, during performance of these tasks will be useful for evaluation of potential therapeutic treatments in HD.

Disclosures: J. Cheng: None. **E.T. Koch:** None. **L.A. Raymond:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.16

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

Support: CIHR

Title: Investigating the contribution of tau to Huntington's disease pathology

Authors: *S. SALEM[1], M. ALPAUGH[1], M. SAINT-PIERRE[1], T. BELLANDE[2], R. MELKI[2], F. CICCHETTI[1];
[1]Ctr. de recherche Univ. Laval, Ctr. de recherche Univ. Laval, Québec, QC, Canada; [2]Inst. François Jacob (MIRcen) and CNRS, Fontenay-aux-Roses, France

Abstract: In primary and secondary tauopathies, tau becomes hyperphosphorylated and accumulates into various pathological forms. In Huntington's disease (HD) - a genetic neurodegenerative disorder - huntingtin (HTT) is the disease associated protein however, the presence of hyperphosphorylated tau, accumulation of neurofibrillary tangles and increased expression of 4R tau isoforms in post-mortem brain tissue from HD patients, suggests this disorder may be a secondary tauopathy. Given the presence of both mutant HTT (mHTT) and pathological tau in HD patients, our study aims to evaluate the interactions of these two proteins. We hypothesize that the introduction of tau to cell and animal models of HD exacerbates intracellular HTT aggregation, as well as modifying disease associated features. To test this hypothesis, human synthetic recombinant 3R/4R tau were introduced to a cellular model ($StHdh^{Q111/Q111}$) and a knock-in mouse model (zQ175) of HD, followed by assessment of cellular dysfunction and behavioral changes. For all experiments, untreated controls, protein controls (monomers) and experimental (fibrils) conditions were included. *In vitro,* control Q7 and HD Q111 cells were treated with various tau doses in order to identify the lowest dose with a significant effect on cell metabolism as measured by a MTT assay. The effect of tau on HTT aggregation was then assessed by filter retardation assays, demonstrating that both monomeric and fibrillar 3R tau are sufficient to increase HTT aggregation in HD cells. *In vivo,* bilateral intracerebral stereotaxic injections of tau were performed in the prefrontal cortex and hippocampus (2µg/site) of wild-type (WT) and zQ175 mice (n=11 - 16 per group) at 3 months of age. A battery of behavioral tests such as Barnes maze and Open field were performed at baseline and post-surgery. Our behaviour data show that tau aggravates cognitive and motor impairments in zQ175 mice at 9 months of age. Post-mortem studies revealed that behavioral changes are accompanied by an increase in mHTT aggregation in the prefrontal cortex of zQ175 mice treated with 3R fibrils as observed by both filter retardation assays and immunohistochemistry. Additionally, increased phosphorylation of tau at disease-associated residues is observed in the hippocampus of tau treated WT mice. Together, our results show that tau treatment alters cellular features associated with HD and worsens behavioral phenotypes in mice with a particularly striking effect of 3R fibrils on HTT aggregation. This supports the idea that tau pathology contributes to HD pathology, and further elucidation of the mechanisms by which this occurs could reveal new therapeutic strategies for HD.

Disclosures: S. Salem: None. **M. Alpaugh:** None. **M. Saint-Pierre:** None. **T. Bellande:** None. **R. Melki:** None. **F. Cicchetti:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.17

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

Support: R01 AG057896
Rainwater Charitable Foundation

Title: Development of a new, in vivo, screenable, split-luciferase based model of huntingtin multimerization

Authors: *M. G. THOMAS[1,2,3], S. A. LEVY[1,2,3], B. FROST[1,2,3];
[1]Cell Systems and Anat., Univ. of Texas Hlth. Sci. Center, San Antonio, San Antonio, TX; [2]Barshop Inst. for Longevity and Aging Studies, San Antonio, TX; [3]Glenn Biggs Inst. for Alzheimer's and Neurodegenerative Dis., San Antonio, TX

Abstract: Development of a new, *in vivo,* screenable, split-luciferase based model of huntingtin multimerization
Morgan G. Thomas, Simon A. Levy, Bess Frost
Huntington's disease is a genetic neurodegenerative disorder caused by a polyglutamine (polyQ) expansion in the gene that encodes for the protein huntingtin. PolyQ expansion in huntingtin causes aggregation of the protein into a range of multimeric species ranging from soluble oligomers to fibrillar, insoluble inclusion bodies. Huntingtin multimerization involves transition of the conformationally-flexible polyQ stretch to a beta-sheet rich structure, monomers of which interact with one another to form multimeric species. The precise mechanisms and biological regulators of the transition of huntingtin from beta-sheet rich monomers to oligomers, subsequent aggregation into inclusion bodies, and potential clearance strategies are incompletely understood. Studies to date rely predominantly on assays performed *in vitro* or in cultured cells. To better understand the cellular factors facilitating huntingtin aggregation in a living, aging, screenable model organism over time, we have developed htt^{LUM}, a split-luciferase-based detector of huntingtin-huntingtin interaction in adult neurons of the *Drosophila melanogaster* brain. Using this system, we can quantify the extent of huntingtin multimerization in the *Drosophila* brain in flies housed and measured in a 96-well plate. This system allows quantification of huntingtin aggregation in real time in a living, active model organism. We will use htt^{LUM} to identify genetic modifiers of huntingtin aggregation and pharmacological approaches to limit huntingtin multimerization and associated neurotoxicity.

Disclosures: M.G. Thomas: None. **S.A. Levy:** None. **B. Frost:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.18

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

Support: Annexon Biosciences

Title: C1q inhibition reduces neurodegenerative damage and improves survival in the HD R6/2 mouse model

Authors: *C. HUYNH, A. TASSONI, V. MATHUR, J. VEREEN, L. KUHN, S. SANKARANARAYANAN, E. CAHIR-MCFARLAND, L. MATTHEAKIS, T. YEDNOCK, Y.

ANDREWS-ZWILLING;
Annexon Biosci., Brisbane, CA

Abstract: Huntington's disease (HD) is an autosomal dominant neurodegenerative disorder caused by expansion of CAG repeats in the Huntingtin (HTT) gene. Increased expression of early classical complement components, including C1q, C4, and C3, has been observed in striatal tissue from HD patients, and C1q has been implicated in synapse elimination and neuronal damage in HD mouse models. We examined complement expression, neuronal damage, and the potential therapeutic benefit of classical complement inhibition in an HD animal model. Specifically, we used the R6/2 transgenic mouse model of HD expressing an ~120 CAG expansion and measured classical complement components in the plasma and cerebral spinal fluid (CSF) of transgenic vs wild-type mice. We also measured the levels of neurofilament light chain (Nf-L) as a biomarker of neuronal damage. Additionally, we measured pre-synaptic marker VGLUT1 and striatal Fluoro-Jade C labelling as an indicator of neurodegeneration in the brain. Levels of Iba1 and CD68 were assessed as measures of microglial reactivity. To assess the role of the classical complement pathway in neuronal damage, we pharmacologically blocked C1q activity with an inhibitory antibody (ANX-M1), administered via intraperitoneal injection, and assessed Nf-L, synaptic and neuronal changes, microglial reactivity, and animal survival. In this model of HD, we observed increased plasma levels of C1q and multiple complement components. Nf-L was increased in both the plasma and CSF of R6/2 mice compared to wild-type mice, and there was a significant positive correlation between CSF Nf-L levels and plasma C1q, suggesting a potential role of the classical complement cascade in neuronal damage. Treatment of animals with anti-C1q fully blocked C1q and inhibited the classical pathway in the plasma and brain. Consistent with a role for the classical cascade in neuronal damage, anti-C1q treatment significantly reduced neurodegeneration, inhibited microglial reactivity, and increased survival rates compared to those in untreated R6/2 mice. These results suggest that inhibiting C1q protects against neuronal damage in mice with expanded CAG repeats in the HTT gene and that C1q is a potential pharmacological target in HD. A Phase 2 study of ANX005, an anti-C1q therapy, in HD patients is ongoing (ClinicalTrials.gov: NCT04514367).

Disclosures: C. Huynh: A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **A. Tassoni:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **V. Mathur:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **J. Vereen:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **L. Kuhn:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **S. Sankaranarayanan:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **E. Cahir-McFarland:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership

Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **L. Mattheakis:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **T. Yednock:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **Y. Andrews-Zwilling:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.19

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

Support: CIHR FDN-143210
UBC Four Year Fellowship
CONP Scholar Award

Title: In vivo striatal activity during motor skill learning and spontaneous behaviour in Huntington's disease mice

Authors: *E. T. KOCH, J. CHENG, M. D. SEPERS, L. A. RAYMOND;
Dept. of Psychiatry, Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Huntington's disease (HD) is a neurodegenerative disorder characterized by motor, cognitive and psychiatric deficits. The dorsal striatum is the major site of neurodegeneration in HD, along with atrophy of cortex and other regions. Studies have shown aberrant cortico-striatal signaling in HD mice, including changes to the activity of D1-type dopamine receptor-expressing spiny projection neurons (D1-SPNs) and D2-type dopamine receptor-expressing SPNs (D2-SPNs), aberrant neurotransmitter signaling, and deficits in cortico-striatal plasticity. We were interested in how changes to cortico-striatal signaling, previously reported in brain slice, correlate with behaviour *in vivo*. We combined the accelerating rotarod and open field tasks with calcium imaging using fiber photometry in the striatum of HD mouse models (YAC128 and zQ175). This includes imaging of overall striatal activity with GCaMP7f, as well as population-specific imaging of D1-SPNs and D2-SPNs using green and red calcium sensors. When imaging activity of all striatal neurons, we found that both WT and YAC128 male mice showed increased striatal activity when they performed the rotarod, which reduced over training. 2-3 month-old male YAC128 mice showed no deficit in latency to fall from the rotarod, however, the correlation between striatal activity and behaviour on the rotarod and the open field in male YAC128 was significantly weaker. Interestingly, male YAC128 showed deficits in paw

kinematics, including increased paw slips below the rotarod, which was associated with aberrant striatal activity. At 6-7 months, male YAC128 mice were severely impaired on the rotarod and had significantly increased striatal activity. Male YAC128 mice also showed elevated striatal activity at rest in the open field. We are currently performing follow-up experiments in both male and female YAC128 and zQ175 HD mice to compare the two sexes and two models of HD. Interestingly, female, but not male, 10-month-old zQ175 HD mice showed a deficit in latency to fall from the rotarod. Using both green and red calcium sensors together, we found that the activity of D1-SPNs is inversely correlated with rotarod performance, and 6-7 month old male YAC128 and 9-11 month-old female zQ175 HD mice showed elevated activity in D1-SPNs during early stages of training compared to WT littermates. We are also using machine learning software to analyze specific behaviours in the open field and how D1- and D2-SPN activity are correlated with these behaviours. This work begins to bridge the gap between changes to striatal signaling determined from *in vitro* studies and the behavioural deficits observed *in vivo*.

Disclosures: E.T. Koch: None. **J. Cheng:** None. **M.D. Sepers:** None. **L.A. Raymond:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.20

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

Support: CHDI Foundation

Title: Robust multimodal integration of predictive white matter signals in premanifest Huntington's disease: longitudinal change detection and atrophy prognosis

Authors: *P. POLOSECKI[1], E. CASTRO[1], I. RISH[1], D. PUSTINA[2], J. H. WARNER[2], A. WOOD[2], C. SAMPAIO[2], G. A. CECCHI[1];
[1]IBM Res., Yorktown Heights, NY; [2]CHDI Management/CHDI Fndn., Princeton, NJ

Abstract: Objective: White matter (WM) microstructure deterioration is thought to be one of the earliest changes in premanifest Huntington's Disease (pre-HD), and could provide a sensitive measure of early progression in individuals. For applications, one challenge is robustness of effects across imaging sites and cohorts. Another is how to integrate WM patterns with other modalities, notably powerful genetic burden measures. Here we evaluate the robustness of multivariate models trained to detect pre-HD WM trajectories, as well as the power of baseline WM measures for predicting prospective brain changes in multimodal machine learning models. **Method**: We analysed a total of 220 (106 pre-HD) subjects from two cohorts. Using data from the Track-ON HD cohort on 164 subjects (87 pre-HD, 77 healthy controls) with 2-3 diffusion weighted imaging (DWI) visits in a 2-3 year period, we computed a white matter skeleton using FSL's TBSS method and projected fractional anisotropy (FA) and mean diffusivity (MD) maps to the common skeleton. FA and MD slopes were computed along the skeleton and were used for

classification of pre-HD vs controls by support vector machines (SVMs) via intra-site, cross-site cross-cohort validation (56 subjects from the IMAGE-HD study). Additionally, baseline FA and MD were used to predict prospective longitudinal atrophy rates for three large subcortical structures (caudate, putamen, lateral ventricles). We implemented customized multimodal machine learning methods for integrating white matter signals with genetic burden measures and baseline atrophy values in prognostic models of prospective subcortical atrophy, tested with the same cross-validation scheme.
Results: We found longitudinal WM pre-HD changes (vs. controls), detectable across imaging sites and cohorts. Moreover, WM microstructure robustly predicted prospective atrophy changes. Multimodal models that integrated genetic burden with baseline WM and atrophy often surpassed individual modalities and showed increased robustness. Prominent features of pre-HD WM patterns of change included distal posterior projections of the callosum, while those of fast declining subjects encompassed more proximal segments of the same projections.
Interpretation: Our results suggest subtle WM changes can be robustly detected in unseen individuals and provide a handle to the association of an observed WM trajectory subsequent brain atrophy. A possible role for WM signals in practical applications is their integration with well-established burden measures into multimodal predictive models.

Disclosures: P. Polosecki: None. **E. Castro:** None. **I. Rish:** None. **D. Pustina:** None. **J.H. Warner:** None. **A. Wood:** None. **C. Sampaio:** None. **G.A. Cecchi:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.21

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

Support: NIH R01 Diversity Supplement Grant

Title: Interrogating the origins of mutant huntingtin in cerebrospinal fluid

Authors: *R. BANOS[1], N. S. CARON[2], Y. XIE[1], N. POTLURI[1], M. R. HAYDEN[2], A. L. SOUTHWELL[1];
[1]Col. of Med., Univ. of Central Florida, Orlando, FL; [2]Med. Genet., Ctr. for Mol. Med. and Therapeut., Vancouver, BC, Canada

Abstract: Huntington disease (HD) is a fatal, neurodegenerative disease caused by a CAG repeat expansion in the huntingtin (*HTT)* gene encoding for an elongated polyglutamine tract in the HTT protein. The toxicity of expanded mutant HTT (mHTT) leads to regional atrophy and progressive neuronal loss in the brain, occurring earliest in the striatum. In pre-clinical trials, therapeutic lowering of mHTT in the central nervous system (CNS) prevents or rescues HD-like phenotypes, which has led to ongoing clinical development of multiple HTT lowering strategies. However, reliable biomarkers of brain HTT are needed to evaluate clinical target engagement.

Our lab has developed an ultrasensitive immunoprecipitation and flow cytometry (IP-FCM) assay that allows quantification of mHTT in HD patient and mouse cerebrospinal fluid (CSF). Using IP-FCM we have shown that CNS neural tissue is the major source of mHTT in the CSF and that levels of mHTT in the CSF correlates with brain mHTT after CNS HTT lowering. For this reason, quantification of mHTT in the CSF is being used as a clinical pharmacodynamic biomarker of CNS HTT lowering. However, HTT lowering strategies have different distributions in the CNS. Therefore, further understanding the origin(s) of CSF mHTT will allow for better interpretation of this biomarker. Using humanized HD mice (Hu97/18), we are using a genetic approach to inactivate expression of mHTT in selective tissues and cell types in the CNS through Cre-mediated recombination. Hu97/18 mice were generated using BACHD HD model mice, which have a floxed exon 1 in the mutant allele and YAC18 control mice that do not. Thus, in cells expressing the cre recombinase in Hu97/18 mice, mHTT is selectively inactivated. Recently we have demonstrated that inactivation of mHTT in the striatum of Hu97/18 mice significantly lowers CSF mHTT, suggesting that the striatum is a contributor to CSF mHTT. However, understanding the contributions of other brain regions and cell types will be necessary to interpret CSF mHTT as a biomarker for target engagement in the CNS. In ongoing studies, we have inactivated mHTT selectively in the cortex, in neurons, or in astrocytes of Hu97/18 mice. By comparing CSF mHTT in these lines to one another, to the line with striatal mHTT inactivation, and to the parent Hu97/18 line we can delineate regional and cell type contributions to CSF mHTT. Understanding the origin of CSF mHTT will provide critical information about which HD interventions could be accurately assessed by changes in CSF mHTT as well as better interpretation of CSF mHTT as a biomarker for disease progression.

Disclosures: R. Banos: None. **N.S. Caron:** None. **Y. Xie:** None. **N. Potluri:** None. **M.R. Hayden:** None. **A.L. Southwell:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.22

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

Title: The “eat me-don’t eat me” mechanism in Huntington’s disease mouse model

Authors: E. PALDINO[1,3], G. MIGLIORATO[2,4], S. BARATTUCCI[5], ***F. R. FUSCO**[1];
[1]Santa Lucia Fndn. Rehabil. Hosp., Roma, Italy; [2]Santa Lucia Fndn. Rehabil. Hosp., Rome, Italy; [3]Systems Med., Univ. of Rome Tor Vergata, Rome, Italy; [4]Univ. of Trieste, Trieste, Italy; [5]Santa Lucia foundation Rehabil. Hosp., Rome, Italy

Abstract: Background: Mechanisms of tissue damage in Huntington's disease involve excitotoxicity, mitochondrial damage, and neuroinflammation, including microglia activation. We have previously described NLRP3 inflammasome and the role of pyroptosis process in the striatal neurons of the R6/2 mouse model of Huntington's disease. CD47 is a membrane protein

that interacts with the myeloid inhibitory immunoreceptor SIRPα. Engagement of SIRPα by CD47 provides a downregulatory signal that inhibits host cell phagocytosis, and CD47 therefore functions as a "don't-eat-me" signal. These proteins are involved in the immune response and are downmodulated in MS (Han et al, 2012). In this study, we have focused on new factors that can be involved in the modulation of cell death in HD, namely the “eat me-don’t eat me” mechanisms expressed through the CD47 and SIRP alpha proteins, and of CD 206, CD 40 and CR3, on the other.**Methods:** Histological and immunohistochemical studies with antibodies against CD47, SIRP α, CD 206, CD 40 and CR3 performed at pre-symptomatic and fully symptomatic disease stages of transgenic mice brain. **Results:** We found that the don’t eat me signals of CD47 and SIRP α were intensely expressed in WT animals and in the early stages of R6/2 mice. Their protein expression was higher in striatal spiny neurons and in parvalbumin interneurons, which are prone to degenerate in HD Conversely, the pro-inflammatory “eat me” factors CD206 and CR3 were upregulated in the R6/2 mice brain, particularly in the later stages of the disease. **Conclusions** An important role of the “eat-me-don’t eat me” signals was described in HD, and to act on the balance between the two could be potentially beneficial in fighting neurodegeneration in HD

Disclosures: E. Paldino: None. **G. MIgliorato:** None. **S. Barattucci:** None. **F.R. Fusco:** None.

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.23

Title: WITHDRAWN

Poster

534. Triplet-Repeat Disorders and Related Disease

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 534.24

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

Support: NIH Grant 2220-6A06
EU Horizon 2020 Grant 721802

Title: Huntingtin secretion in a free form and in extracellular vesicles modulates neuronal activity and is a potential biomarker for Huntington disease

Authors: *I. CALDEIRA BRAS[1,2], R. XIE[1], T. FLEMING OUTEIRO[3], A. SOUTHWELL[1]; [1]Burnett Sch. of Biomed. Sci., Univ. of Central Florida, Orlando, FL; [2]Dept. of Exptl.

Neurodegeneration, Univ. Med. Ctr. Göttingen, Goettingen, Germany; [3]Dept. of Exptl. Neurodegeneration, Univ. Med. Ctr. Goettingen, Goettingen, Germany

Abstract: Huntington disease (HD) is a dominantly inherited genetic disorder characterized by psychiatric, cognitive and motor symptoms displayed with the progressive degeneration of striatal neurons. The trinucleotide repeat expansions in the huntingtin (HTT) gene produces the mutant protein with extended polyglutamine sequence. Observation of mutant HTT (mHTT) aggregates within fetal striatal transplants in HD patients suggested a new mechanism of toxicity based on HTT capacity to propagate between cells. Additionally, HTT detection in human cerebrospinal fluid (CSF) and plasma demonstrates that it can be secreted from cells to the extracellular space. The correlation of mHTT levels in CSF with motor and cognitive symptoms in patients, and the demonstration that HTT lowering in the brain of HD mice results in correlative decrease in CSF HTT, highlights the importance of measuring extracellular HTT. Several mechanisms have been proposed for HTT release from cells, as in a free form and extracellular vesicles (EVs). However, the relative contribution of these mechanisms to HTT levels in the extracellular space and eventually in the CSF, and the overall effect on neuronal function is still unclear.To address this, we exploited different *in vitro* and *in vivo* models to investigate the mechanisms of HTT release to the extracellular space and clearance to the CSF. We have previously demonstrated that healthy neurons secrete both wild-type and mHTT, which contributes to the deposition of HTT in CSF. Although neurodegeneration increases HTT levels in CSF, we demonstrated that HTT was present in the absence of neurodegeneration and that HTT can enter the CSF by both passive release and active secretion. Interestingly, HTT is also present in ectosomes and exosomes, EVs with singular proteomic profiles. Assessment of neuronal network activity using multi-electrode array recordings showed that spontaneous neuronal activity can be modulated by HTT in a free form and in EVs. Neurons treated with free mHTT display greater impairment in the coordinated network activity correlated with the toxic effects of polyglutamine expansion, in contrast with the wild-type protein. Internalization of ectosomes and exosomes in neuronal cells disrupts their regular synchronized bursting activity, resulting in overall lower and more disorganized spiking activity.A detailed understanding of the mechanisms involved in HTT secretion and its potential function in the extracellular space will be important for the development of improved therapeutic strategies. Furthermore, HTT may be exploited as a valuable CSF biomarker to evaluate treatment-induced changes in HD therapies.

Disclosures: I. Caldeira Bras: None. **R. Xie:** None. **T. Fleming Outeiro:** None. **A. Southwell:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.01

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Title: Development and characterization of a robust FTD disease cell line to study tauopathies

Authors: L. RITSMA[1], ***M. BSIBSI**[1], S. VAN HOPPE[1], F. STEVENHAGEN[1], S. COMPTE SANCERNI[1], E. DE KRAA1[1], A. POPALZIJ[1], M. IOVINO[1], T. OOSTERVEEN[2], O. DOVEY[2], T. FROLOV[2], A. TURNER[2], F. PATELL-SOCHA[2], D. FISCHER[1], M. VLAMING[1]; [1]Charles River, Leiden, Netherlands; [2]Bit.Bio, Cambridge, United Kingdom

Abstract: Development and characterization of a robust FTD disease cell line to study tauopathies

Laila Ritsma[1], Malika Bsibsi [1], Stephanie van Hoppe[1], Fleur Stevenhagen[1], Sara Compte Sancerni[1], Esther de Kraa[1], Abeera Popalzij[1], Mariangela Iovino[1], Tony Oosterveen[2], Oliver Dovey[2], Tonya Frolov[2], Amanda Turner[2], Farah Patell-Socha[2], David Fischer[1], Marijn Vlaming[1]

1 Discovery Charles River**2** bit.bio, Babraham Research Campus, Cambridge, UK

Tauopathies, such as frontotemporal dementia (FTD), are neurodegenerative diseases characterized by the pathological aggregation of hyper-phosphorylated Tau (pTau) protein, in the shape of intracellular paired helical filaments (PHFs) or neurofibrillary tangles (NFTs), within neurons and glia, leading to cell death. Mutations in the microtubule-associated protein Tau (MAPT) gene result in tauopathies. Here, we aimed to develop and characterize a physiologically relevant and robust *in vitro* FTD model, to aid the future development of FTD disease therapeutics. Using CRISPR-Cas9 gene editing technology, familial mutations P301S and N279K underlying FTD were engineered into an iPSC line that carries the opti-ox^{TM} technology and can rapidly be reprogrammed into glutamatergic neurons. By means of immunocytochemistry, ddPCR, HTRF and western blot we characterized neurons derived from the distinct clones for FTD phenotypes. Preliminary immunocytochemistry data shows that all clones give rise to mature glutamatergic neurons as they express the classical vGlut1, vGlut2, TUBB3 and MAP2 marker genes, along with minimal cell debris, indicating healthy cell cultures for all clones. One homozygous P301S clone showed a clear increase in pTau (pTau202/205, pTau217 and pTau404) to total Tau ratio compared to the wild type (WT). Moreover, preliminary ddPCR data indicated a significant increase of 4R Tau to total Tau ratio of one homozygous N279K clone compared to the WT.In conclusion, the elevated pTau to total Tau ratio acquired from the immunocytochemistry assay along with the elevated 4R to total Tau ratio acquired from the ddPCR assay indicates the potential of at least two clones as possible disease models to aid future research into developing FTD disease therapeutics.

Disclosures: L. Ritsma: None. **M. Bsibsi:** None. **S. van Hoppe:** None. **F. Stevenhagen:** None. **S. Compte Sancerni:** None. **E. de Kraa1:** None. **A. Popalzij:** None. **M. Iovino:** None. **T. Oosterveen:** None. **O. Dovey:** None. **T. Frolov:** None. **A. Turner:** None. **F. Patell-Socha:** None. **D. Fischer:** None. **M. Vlaming:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.02

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: NIH/NINDS 1R01NS119280
NIH/NIA 1R01AG059639
AARGD-591887 (AARFD-21-847663)
Precision Health Initiative in AD at Indiana University and the Stark Neurosciences Research Institute (SNRI)

Title: Downregulation of Bassoon: A potential therapeutic for tauopathies

Authors: *H. PATEL[1,2], P. MARTINEZ[1,2], Y. YOU[1,2], N. JURY GARFE[1,2], A. PERKINS[1,2], A. LEE-GOSSELIN[1,2], Y. YOU[1,2], G. VIANA DI PRISCO[1,3], X. HUANG[4], A. WIJERATNE[5], S. SHAHID[1,6], A. L. MOSLEY[5], Y.-C. WU[1,6], D. L. MCKINZIE[1,3], J. ZHANG[7,8], B. K. ATWOOD[1,3], C. A. LASAGNA-REEVES[1,2,8];
[1]Stark Neurosciences Res. Inst., [2]Anatomy, Cell Biol. and Physiol., [3]Pharmacol. and Toxicology, [4]Biostatistics and Hlth. Data Sci., [5]Biochem. and Mol. Biol., [6]Radiology & Imaging Sci., [7]Med. and Mol. Genet., [8]Computat. Biol. and Bioinformatics, Indiana Univ. Sch. of Med., Indianapolis, IN

Abstract: Abnormal aggregation of the protein tau is the underlying cause of several neurodegenerative diseases, termed tauopathies, which are associated with motor and cognitive impairments. Despite significant advances, the nature of the tau species involved in tau pathogenesis and functional decline remains unclear. To identify the association between tau pathology and the functional deficits manifested, we assessed multiple *in vivo* phenotypes in the PS19 tauopathy mouse model, overexpressing human tau with P301S mutation. At 9 months, these mice displayed a hyperactivity-associated increased locomotor activity in the open field, a decline in motor strength measured by 2-paw and 4-paw grip strength assays, and a deterioration in the overall physiological conditions marked by lower body temperatures, weight loss, and increase in the measures of frailty. Interestingly, soluble p-tau levels and tau seeding activity correlated with the functional impairment of motor strength and temperature homeostasis supporting the notion that the soluble tau species with seeding activity is central to tau pathogenicity. We next used Size Exclusion Chromatography and Mass Spectrometry to isolate and characterize the 'tau-seed' involved in the seeding activity in the PS19 model. We identified Bassoon (BSN), a presynaptic protein, as an important interactor of the tau-seed, associated with exacerbation of tau seeding and tau propagation. To evaluate the therapeutic potential of BSN in the context of tau pathology, we downregulated BSN in the PS19 mice using neonatal (P0) intracerebroventricular (ICV) injection of an AAV harboring a short-hairpin RNA (shRNA) against murine BSN (shBSN) or control 'scramble' shRNA. Downregulation of BSN rescued synaptic impairments associated with LTP, recovered several functional deficits observed in PS19 mice, and reduced brain atrophy as measured by MRI. Furthermore, we also observed a reduction in overall pathological tau burden in the PS19 mice. Our results demonstrate the beneficial effects of downregulating BSN and open an avenue to target BSN as a potential therapeutic for tauopathies.

Disclosures: H. Patel: None. **P. Martinez:** None. **Y. You:** None. **N. Jury Garfe:** None. **A. Perkins:** None. **A. Lee-Gosselin:** None. **Y. You:** None. **G. Viana Di Prisco:** None. **X. Huang:** None. **A. Wijeratne:** None. **S. Shahid:** None. **A.L. Mosley:** None. **Y. Wu:** None. **D.L. McKinzie:** None. **J. Zhang:** None. **B.K. Atwood:** None. **C.A. Lasagna-Reeves:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.03

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: NIH/NINDS 1R01NS119280
NIH/NIA 1R01AG059639
AARGD-591887
AARFD-21-847663
NINDS P50 NS38377
NIA P50 AG05146

Title: Bassoon as a novel tau-seed interactor in neurotoxicity and propagation

Authors: *P. MARTINEZ[1], H. S. PATEL[1], Y. YOU[1], N. JURY GARFE[1], Y. YOU[1], X. HUANG[2], S. DUTTA[4], A. WIJERATNE[3], J. REDDING[5], J. F. CODOCEDO[1], G. E. LANDRETH[1], A. L. MOSLEY[3], J.-C. ROCHET[4], J. ZHANG[2], J. C. TRONCOSO[5], C. A. LASAGNA-REEVES[1];
[1]Dept. of Anatomy, Physiol. and Cell Biol., [2]Dept. of Med. and Mol. Genet., [3]Dept. of Biochem. and Mol. Biol., Indiana Univ., Indianapolis, IN; [4]Dept. of Medicinal Chem. and Mol. Pharmacol., Purdue Univ., West Lafayette, IN; [5]Div. of Neuropath., Dept. of Pathology, Johns Hopkins Univ., Baltimore, MD

Abstract: Pathological tau aggregates are critical histopathological features of Alzheimer's disease (AD) and other tauopathies. An important focus of research has been to understand the pathological tau propagation in AD patient brains that follow neuronal networks. Despite the knowledge acquired, the cellular mechanism involved in tau propagation and seeding are still unclear. Considering the key role of tau propagation in disease pathogenesis, we performed an unbiased quantitative mass spectrometry-based (masspec) to identify the tau species involved in spreading and the protein interactome of this "tau-seed" in PS19 mice, as well as in human AD and PSP cases. TBS-soluble brain extracts were passed through a Size Exclusion Chromatography (SEC) column. We then measured the tau seeding activity of each SEC-fraction using a well-described biosensor cell line that relies on flow cytometry detection of the FRET signal. We determined that tau from fraction-9 (F9) is found as an HMW-tau complex *in vivo* (>2,000KDa), containing the strongest seeding activity. Interestingly, the tau present in F9 is less than 5.4% of the total tau in the brain of PS19 mice, suggesting that this HMW-tau seed interacts with proteins that give it the ability to form a pathological HMW-tau "*seed"*. The immunoprecipitated HMW-tau from F9 was characterized by Electron Microscopy, revealing that the tau seed adopts a protofibrillar morphology. After masspec and bioinformatic analysis, we found proteins that specifically interact with pathological tau oligomers. Importantly, we report the identification of Bassoon (BSN), a scaffolding protein of the presynaptic active zone, as a significant interactor of the tau-seed isolated from a mouse model of tauopathy and AD and Progressive Supranuclear Palsy (PSP) postmortem samples. We show that BSN specifically interacts with HMW-tau, and exacerbates tau seeding, spreading, and toxicity *in vitro* and *in*

vivo. Furthermore, BSN downregulation significantly decreases tau spreading and overall disease pathology *in vivo*, rescuing synaptic and behavioral impairment and ameliorating brain atrophy. Our findings improve the understanding of tau-seeds and highlight the importance of identifying interactors, such as BSN, that could stabilize these seeds. Inhibiting tau-seed interactors is a potential new therapeutic approach for neurodegenerative tauopathies.

Disclosures: P. Martinez: None. **H.S. Patel:** None. **Y. You:** None. **N. Jury Garfe:** None. **Y. You:** None. **X. Huang:** None. **S. Dutta:** None. **A. Wijeratne:** None. **J. Redding:** None. **J.F. Codocedo:** None. **G.E. Landreth:** None. **A.L. Mosley:** None. **J. Rochet:** None. **J. Zhang:** None. **J.C. Troncoso:** None. **C.A. Lasagna-Reeves:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.04

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: State of Arizona, DHS

Title: Cellular models for the investigation of the structure and activity of human tau protein aggregate formation

Authors: *C. J. HUSEBY[1], E. RANAWEERA[2], P. FROMME[2];
[1]Neurodegenerative Dis. Res. Center, Arizona State Univ. Biodesign Inst., Tempe, AZ; [2]Ctr. for Applied Structural Discovery, Arizona State Univ. Biodesign Inst., Tempe, AZ

Abstract: Alzheimer's disease (AD) is a complex neurodegenerative disease with clinical symptoms of mental decline and postmortem markers such as intraneuronal hyperphosphorylated tau neurofibrillary tangles (NFT) in the brain. Mounting evidence for the central role of tau protein in neurodegeneration highlights the need for deep investigation of intracellular tau aggregation pathways and formation of aberrant tau polymorphs within cellular models. Intrinsically disordered proteins with low sequence complexity like hyperphosphorylated tau protein can form condensed folded structures. Candidates for aberrant intracellular tau folding and aggregation have been identified including liquid-liquid phase separation (LLPS) and nanocrystal growth at ribosomes. Recent research discovered that the tau molecule fold at the core of pathological tau aggregates can differentiate tauopathies. It is hypothesized that disease specific changes in the cellular environment surrounding tau, such as patterns of hyperphosphorylation, could lead to differential core structures. This exciting new structural information showing tau aggregates are unique to each tauopathy opens the door to a structural approach for elucidating mechanisms of pathogenic tau aggregation pathways in cellular model systems and AD brain. Combining advanced structural determination methods including cryoEM, cryoFIB, and cryo-electron tomography, we can directly image intracellular tau aggregates in cells as well as autopsy tissue derived from human brain. Here we have developed

exciting new cellular models of human tau aggregation using both mammalian and insect cell lines which we employ to systematically test intracellular forces necessary to create unique pathological tau structures beginning with patterns of hyperphosphorylation.

Disclosures: C.J. Huseby: None. **E. Ranaweera:** None. **P. Fromme:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.05

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: NIH 2020 R01 AG

Title: Understanding Tau Propagation using Convolution Neural Networks

Authors: *S. WANG[1], J. LAMSTEIN[1], S. FINKBEINER[2];
[2]Ctr. for Systems and Therapeut., [1]Gladstone Inst., San Francisco, CA

Abstract: Understanding Tau Propagation using Convolution Neural Networks
Recent evidence implicates that the microtubule-associated protein tau (MAPT) plays an important role in the pathogenesis of multiple neurodegeneration diseases, including Alzheimer's disease (AD), frontotemporal dementia (FTDP), Parkinson's disease (PD), and progressive supranuclear palsy (PSP). Pathological aggregation and deposition of tau were observed in these diseases, although they present distinct clinical features and affect different brain regions. Pathological tau performs prion-like activities, can form different conformational strains (or "seed"), propagate, and spread through the brain. However, how the different isoforms of tau can causally contribute to one respective tau-related disease but not others, and how the propagation processes might differ inside a cell remains to be determined. In the presentation, we aim to address this issue by integrating longitudinal live-cell imaging of tau seeding and the deep learning model. We modeled in-vitro tau seeding and trained convolution neural networks (CNNs) to recognize the tau pattern inside the cell, with or without seeding. Using different preformed protein fibrils (PFF) of tau, we modeled Tau aggregation in vitro and tested the ability of CNN models to distinguish the strain differences in propagation. To further investigate the differences, we used Gradient-weighted Class Activation Mapping (Grad-CAM) to visualize the essential signal changes from live-cell imaging that the CNN models consider in distinguishing the tau strain effect. Moreover, we multiplexed with different biosensors to identify the affected pathways identified by the Grad-CAM. Our work will contribute to the understanding of strain differences in tau and potentially other prion-like proteins.

Disclosures: S. Wang: None. **J. Lamstein:** None. **S. Finkbeiner:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.06

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: P01 AG014449
P30 AG072931

Title: Tangle evolution within the default mode network during the course of Alzheimer's disease

Authors: *B. KARA[1], N. M. KANAAN[1], E. J. MUFSON[2], S. E. COUNTS[1];
[1]Translational Neurosci., Michigan State Univ., Grand Rapids, MI; [2]Neurobio., Barrow Neurolog. Inst., Phoenix, AZ

Abstract: The Default Mode Network (DMN) is a large-scale cortical brain network that mediates cognitive function in the absence of demanding external tasks and displays altered functional connectivity (fc) during the early stages of Alzheimer's disease (AD). From a clinical-pathological perspective, the DMN displays hypoconnectivity that correlates with cognitive decline once neurofibrillary tangles (NFTs) become detectable via PET imaging. However, growing evidence suggests that early, pre-tangle modifications of soluble tau (e.g., oligomers) confer tau toxicity. Hence, there is a critical knowledge gap about when pre-tangle tau pathological changes occur within the DMN that initiate fc alterations prior to the appearance of NFT pathology using PET tracers. We aimed to investigate the relationship between early pathological tau changes across the three main DMN hubs (medial frontal cortex, posterior cingulate cortex, and precuneus) and cognitive decline. In our pilot cohort (n=36), we quantified pre-tangle tau modifications in DMN hubs using pS422 (early pathogenic phosphorylation event), TOC1 (tau oligomers), TNT2 (aberrant N-terminus conformational change), and TauC3 (C-terminus truncation at amino acid 421) epitope-specific antibodies in postmortem fixed tissue from Rush ROS/MAP cases representing all six Braak NFT stages. For quantitative analysis, we used HALO Image Analysis software. Our preliminary results indicated the accrual of pS422 and TOC1 epitopes as early as Braak stage III (limbic stage). Although the distribution of tau load for the individual epitopes was not significantly different among the hubs, we found a strong correlation between pS422 load in the frontal cortex and the Mini-Mental State Exam score ($p=0.006$) as well as Braak stage ($p=0.009$). Our initial results indicate that pre-tangle soluble tau accumulation in cortical DMN regions is an early event during the progression of AD that may impact global cognitive function. We have expanded our study (n = ~100 cases) to include frozen and fixed tissue samples from the same donors to cross-validate our quantitative immunohistochemical results via custom ELISAs. We also plan to relate our findings to neuropsychological test scores and other clinical pathologic criteria. Collectively, this study will provide novel insights into early toxic tau pathological accrual in cognitive functional brain connectomes in the early stages of AD.

Disclosures: B. Kara: None. **N.M. Kanaan:** None. **E.J. Mufson:** None. **S.E. Counts:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.07

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: NIH/NINDS 1R01NS119280
AARGD-591887

Title: Phosphorylation at Serine 214 correlates with tau seeding activity in the progression of tauopathies

Authors: *A. MANABAT[1,2], P. MARTINEZ[1,2], A. PERKINS[1,2], Y. YOU[1,2], Y. YOU[1,2], H. PATEL[1,2], A. LEE-GOSSELIN[1,2], N. JURY GARFE[1,2], R. VIDAL[2,3], C. A. LASAGNA-REEVES[1,2];
[1]Dept. of Anatomy, Cell Biol. & Physiol., [2]Stark Neurosciences Res. Inst., [3]Dept. of Pathology and Lab. Med., Indiana Univ. Sch. of Med., Indianapolis, IN

Abstract: Under pathological conditions, the microtubule-associated protein tau is hyperphosphorylated. This leads to the abnormal aggregation of tau and the formation of neurofibrillary tangles (NFTs), which is a main neuropathological hallmark of Alzheimer's disease (AD) and other tauopathies. Increasing evidence supports the idea of "seeding," in which intermediate, soluble tau aggregates can propagate between cells and induce the misfolding of non-pathogenic tau monomers, thereby promoting the intracellular aggregation of tau. However, the nature of the tau species involved in seeding and propagation remains to be fully elucidated. For this reason, we aim to identify the nature of the tau seed in the context of disease progression in hopes of using this species as a future therapeutic intervention target or biomarker for disease progression. Using a FRET-based biosensor cell line, we profiled the temporal evolution of tau seeding activity in the brains of two mouse models of human tauopathies: hTau P301S (0N4R) and hTau P301S (1N4R). Utilizing a diverse set of phosphor-and conformational tau species antibodies, we also biochemically and histologically evaluated these tauopathy models. Our results demonstrated phosphorylation of tau at Serine 214 (pTauS214) occurs prior to the formation of conventional AT8 positive NFTs and has a distinct histological pattern that differs from AT8. Additionally, it was found that pTauS214 was the only tau species that strongly correlated with seeding activity during disease progression in both tauopathy models used. Taken together, this study demonstrated how pTauS214 is an early event of tau pathogenesis and that this pathology differentiates from AT8-detected NFTs-like pathology.

Disclosures: A. Manabat: None. **P. Martinez:** None. **A. Perkins:** None. **Y. You:** None. **Y. You:** None. **H. Patel:** None. **A. Lee-Gosselin:** None. **N. Jury Garfe:** None. **R. Vidal:** None. **C.A. Lasagna-Reeves:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.08

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: NIH Grant AG065925

Title: Role of phosphorylation in activity-dependent release of human tau from Drosophila larval neuromuscular junction

Authors: R. UDDIN, *D. LEE;
Ohio Univ., Athens, OH

Abstract: Alzheimer's disease (AD) is one of the most common forms of dementia that shows progressive memory loss. A key pathological hallmark of AD, neurofibrillary tangles (NFTs) are intracellular aggregates of hyper-phosphorylated tau protein. NFTs are initially formed in the brain's temporal lobes and then progressively spread throughout the brain. Based on these findings, it has been proposed that intracellular tau is released into extracellular space as oligomers from the affected neurons and then taken up by the nearby healthy neurons. Finally, transferred tau serves as a seed in a healthy neuron for its continuous propagation. However, mechanisms of tau propagation are still not well elucidated. In our study, we wanted to see how neuronal activity and phosphorylation affect tau release as both neuronal excitability and phosphorylation are increased in the early stage of AD. We used the 3rd instar *Drosophila* larval neuromuscular junction (NMJ) as an *in vivo* model system to investigate mechanisms underlying tau release. We have chosen *Drosophila* NMJ because it is glutamatergic and greatly resembles neuronal and synaptic functions in human central nervous system. To increase neuronal excitability, we expressed channelrhodopsin (ChR2) in addition to wild-type human tau (hTau) in the glutamatergic motor neurons of flies by using a driver D42-Gal4. ChR2 was activated by blue light (470nm, 3x 10 min with 1 hour interval) to induce hTau release. Our ELISA results showed a significant increase of hTau release into the hemolymph from NMJ compared to the control group. Furthermore, we tested the role of different phosphorylation sites (pSites) of tau protein on the activity-dependent release by using phospho-specific antibodies (e.g., AT8 or PHF-1). We have demonstrated that tau released by neuronal stimulation is highly phosphorylated at different sites in the proline-rich domain (PRD) and C-terminal. Based on our findings, it can be said that neuronal excitability and phosphorylation play a significant role in hTau release. We are currently examining the importance of individual pSites in PRD and C-terminal by using *Drosophila* transgenic lines carrying one or two mutations in the pSites (e.g., UAS-hTau[S202A/T205A]). Overall, our novel *Drosophila* NMJ model will provide an important *in vivo* experimental platform to examine molecular and cellular mechanisms underlying tau release and the impact of tau phosphorylation on its activity-dependent release.

Disclosures: R. Uddin: None. **D. Lee:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.09

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: The design, study conduct, and financial support for this research were provided by AbbVie

Title: Tau aggregates derived from phosphorylated and unphosphorylated tau have differential aggregation kinetics, seeding competency, and toxicity

Authors: H.-Y. WU, O. NAZARKO, X. YANG, N. BROWN, K. TAYLOR, D. NANAVATI, J. XU, L. HUANG;
AbbVie Bioresearch Ctr., Abbvie, Worcester, MA

Abstract: One of the pathological hallmarks of Alzheimer's disease (AD) is the accumulation of abnormally phosphorylated tau in neurons and eventual formation of neurofibrillary tangles (NFTs). Although tau molecules in NFTs are found to be highly phosphorylated, it is still unknown whether tau hyperphosphorylation in AD drives fibril formation and subsequently causes cellular toxicity due to aggregation. Here, we utilized human tau (htau) produced from sf9 cells containing high state of phosphorylation, while htau produced by E. coli is unphosphorylated. htau2N4R and htau2N3R were generated from both sf9 cells and E.coli to derive four types of tau aggregates or paired helical filaments (PHFs). To compare the dynamics of tau aggregate formation, we monitored tau aggregation in real time by Dynamic Light Scatter (DLS) and Thioflavin T fluorescence spectrometry, which indicated that in presence of aggregation inducer heparin, phosphorylated htau2N4R and htau2N3R exhibited slower progression of aggregate formation compared to unphosphorylated htau2N4R and htau2N3R. Analysis from circular dichroism spectroscopy (CD) demonstrated 59.6% ß-sheet structure in the aggregation solution of unphosphorylated htau2N4R while only 37.5% was measured for the phosphorylated htau2N4R. The differences in CD spectral and estimated beta-sheet content between unphosphorylated and phosphorylated hTau2N4R aggregates suggests underlying structural differences between the two tau species. Among these four types of tau aggregates, the unphosphorylated htau2N3R showed the most rapid progression of tau aggregate formation with 61.4% ß-sheet structure. By contrast, the aggregates formed from the phosphorylated tau2N4R demonstrated the highest potency in inducing seeding in tau biosensor HEK cells expressing tau 4-repeat domains, and cultured primary neurons expressing human tau compared to other three tau aggregates. To investigate toxicity of the different aggregates, cultured primary neurons were treated with tau aggregates. We observed that tau aggregates derived from the phosphorylated tau2N4R or tau2N3R induced more significant neurite regression (60-65%) compared to tau aggregates derived from the unphosphorylated tau2N4R or tau2N3R (33-55%). Both tau phospho-specific antibodies tested and N-terminal pan tau antibody effectively blocked seeding and neurite regression. Together, this study illustrated that although both hyperphosphorylated and unphosphorylated tau proteins can form aggregates *in vitro*, tau aggregates derived from

hyperphosphorylated tau possess slower kinetics of fibril formation and are more seed competent and toxic to neurons.

Disclosures: H. Wu: None. **O. Nazarko:** None. **X. Yang:** None. **N. Brown:** None. **K. Taylor:** None. **D. Nanavati:** None. **J. Xu:** None. **L. Huang:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.10

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Title: Transsynaptic spreading of corrupted proteins through the gut-brain axis

Authors: *A. MARTIN-PEÑA, J. LOPEZ-SCARIM, M. TANSEY;
Neurosci., Univ. of Florida, Gainesville, FL

Abstract: Alzheimer's disease (AD) is the most common form of dementia. However, although the clinical and pathological aspect of the disease are thoroughly characterized, the pathophysiological mechanism underlying the progressive neurodegenerative process is not well understood. In consequence, there is no cure and a disease-modifying treatment is lacking. It is therefore imperative to identify the earliest signs of disease pathology to pave the way to identify the triggering molecular factors of the disease and the mechanisms underlying molecular spreading of toxic assemblies. Our search for these key clues in the periphery is founded on the fact that peripheral dysfunction, particularly in the gastrointestinal (GI) system, can arise decades before the onset of the classic symptoms in neurodegenerative disorders. In fact, the risk of AD increases significantly in patients with inflammatory bowel disease (IBD) when compared with the non-IBD population. Furthermore, Tau accumulates in Crohn's Disease guts and in sigmoid colon of AD and FTD patients. Therefore, we are investigating the physiological and molecular mechanisms that support Tau spreading in the prodromal stages and, particularly, along the gut-brain axis. To test whether corrupted proteins expressed in the gut will spread to the brain, we employed the Gal4/UAS system and expressed wild type or mutant human Tau in selected populations of gut epithelium cells. We then monitor behavior on aging flies expressing Tau in the gut epithelium using *Drosophila* activity monitors (DAM) and analyzed their circadian patterns of activity along with several sleep parameters. We observed that flies selectively expressing full length (2N4R) human Tau in enteroendocrine cells of the gut exhibited a significant impairment in their daily activity pattern that specifically affected their behavior during the light phase (day) but not during the dark period (night). Interestingly, these flies also showed an increased sleepiness during the day without affecting their nocturnal sleep pattern. The onset of these behavioral changes appears to occur as early as 10 days after eclosion. Our results support the presence of spreading mechanisms for Tau from gut epithelial cells to the brain; hence, affecting the neuronal physiology that sustain daily activity patterns.

Disclosures: **A. Martin-Peña:** None. **J. Lopez-Scarim:** None. **M. Tansey:** A. Employment/Salary (full or part-time):; University of Florida. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NIH, MJ Fox Foundation, Parkinson's Foundation, Alzheimer's Association, ASAP/MJFF. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Biogen. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Xencor Inc. F. Consulting Fees (e.g., advisory boards); Weston Foundation, Quebec Parkinson's Network, Alzheimer's Association, INmune Bio, Jaya, Longevity, Biogen/IONIS, Amylyx, iMetabolicPharrma, Cerebral Therapeutics, Sanofi, Nysno Bio.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.11

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Title: Mutants associated with familial tauopathies display variations in oligomeric conformations and aggregation propensities

Authors: *A. BHOPATKAR[1], N. BHATT[2], R. KAYED[2];
[1]Univ. of Texas Med. Br., [2]Neurol., Univ. of Texas Med. Br., GALVESTON, TX

Abstract: The microtubule-associated protein, Tau (MAPT), is implicated in a multitude of neurodegenerative disorders, including Alzheimer disease (AD) and frontotemporal dementia (FTD). These disorders are characterized by the presence of the aggregated form of the protein. Despite this, recent studies assign a prominent toxic role to the soluble, oligomeric form of the protein instead of the insoluble aggregates. Within this context, it remains unanswered how mutations identified in familial tauopathies alter the aggregation and conformational dynamics of the protein and how they lead to the generation of structural polymorphs of the oligomers. Using biophysical and biochemical techniques, we characterized and compared the oligomeric forms of 2N4R wild-type Tau and its mutants A152T, P301L, P301S and R406W, which are associated with familial tauopathies. We investigated their structural and conformational characteristics using biochemical and immunochemical analysis, where results indicated that there exists variability amongst them. This was complemented by assays using fluorescent probes including thioflavin-T (ThT) and anilinonaphthalene-8-sulfonic acid (ANS), which revealed there also exists differences in the aggregation kinetics, further highlighting that the oligomeric forms of the protein adopt distinct conformations. These differences suggest that individual mutant forms of the proteins use disparate aggregation pathways to generate conformers. Our observations strengthen the hypothesis that structural polymorphism of amyloidogenic proteins underlies the phenotypic variations seen in neurodegenerative pathologies. Overall, our results represent a

preliminary step towards understanding the oligomeric landscape of tau mutants, and future studies will probe deeper into their biological effects using cellular and animal models.

Disclosures: **A. Bhopatkar:** None. **N. Bhatt:** None. **R. Kayed:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.12

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: NIH T32 NS082145
NIH K01AG066747
NIH R21AG072423
Alzheimer's Association AARG-21-846012
Dan & Kelley McNamara Alzheimer's Research Fund
curePSP 679-2021-12

Title: Microglia internalize tau multimers and fibrils of different sizes using macropinocytosis

Authors: *K. F. ODFALK[1], P. J. ESQUINCA[2], S. C. HOPP[1];
[1]Glenn Biggs Inst. for Alzheimer's and Neurodegenerative Dis. and Dept. of Pharmacol., UT Hlth. Sci. Ctr. At San Antonio, San Antonio, TX; [2]Voelker Biomed. Res. Acad., Univ. of Texas Hlth. San Antonio, San Antonio, TX

Abstract: Tauopathy is class of neurodegenerative disease characterized aberrant hyperphosphorylation and misfolding of the microtubule-associated protein Tau and includes more than 20 diverse clinicopathologies (e. g. Alzheimer's disease, AD). Tau is thought to drive neurodegeneration via multimerization into neurotoxic aggregates. Neurons are known to release these aggregates into the parenchyma as both free and vesicle-bound tau. Nearby neurons then internalize these aggregates via macropinocytosis, and this can lead to propagation of Tau pathology. Microglia are actively surveying the local milieu and are therefore likely to endocytose these aggregates. However, the mechanism for microglial uptake of Tau is unknown. We hypothesize that microglia (like neurons) take up take via macropinocytosis. Thus far, we have tested our hypothesis in immortalized murine microglial cell line (BV2). To model extracellular Tau in culture, we used recombinant human P301S mutant Tau monomers, preformed fibrils (PFFs), and sonicated PFFs (sPFFs). We measured the effects of pharmacological macropinocytosis inhibition on Tau uptake and tracked the movement of Tau into the microglial endocytic compartment. We optimized our inhibitor concentrations for specificity and lack of toxicity and found that several putative macropinocytosis inhibitors are nonspecific for macropinocytosis in BV2 microglia. Pretreatment with actin inhibitor (cytochalasin D) abrogated uptake of Tau sPFFs but only partially blocked uptake of monomers and PFFs. Conversely, pan phosphoinositide 3-kinase (PI3K) inhibitor (wortmannin), blocked

uptake of monomers and PFFs but not sPFFs. Further, monomers, PFFs, and sPFFs colocalized with macropinosomes which we labeled using fluorescent 70 kDa dextrans (Dex70). However, we also observed several Tau positive, Dex70 negative vesicles within these cells. Our findings suggest that Tau aggregates are taken up via macropinocytosis in BV2 microglia and some additional unknown endocytic mechanism(s). We plan to investigate other endocytic routes after we have repeated our experiments in primary microglia and organotypic brain slice microglia. Understanding mechanisms that promote microglial clearance of Tau may lead to novel therapies against the progression of Tau pathology.

Disclosures: K.F. Odfalk: None. **P.J. Esquinca:** None. **S.C. Hopp:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.13

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Title: Design optimizationof Tau repeat-domain probe allows detection of bioactive Tau in-situ in Alzheimer's disease brain tissues.

Authors: *R. C. PERBET[1], A. LATHUILIERE[2], C. GLYNN[1], A. ANASTASIE MATE DE GERANDO[1], B. T. HYMAN[1];
[1]Massachusetts Gen. Hosp., Charlestown, MA; [2]EPFL SV BMI LEN, Lausanne, Switzerland

Abstract: Prion-like properties of tau protein are a major mechanism in the pathophysiology of tauopathies. By this mechanism, pathological tau is able to recruit normal tau and convert it into pathological tau. Tau seeding bioactivity is usually quantified using a "biosensor" cell line that expresses tau repeat-domain with a disease-associated mutation fused to complementary fluorophores. Induced aggregation in cells that scores positive by fluorescence resonance energy transfer is next quantified by flow cytometry. Although this assay is specific/sensitive for quantification of bioactive tau, it is not discriminating for localization. Recently, in our laboratory, we tested the hypothesis that rational design of tau probes aligning the sequence of the probe with amino acid sequence incorporated in the central core of Tau aggregates observed by cryo-EM and mimicking post-translational modifications described in Alzheimer disease (AD) brains could improve the assay's sensitivity. Taking advantage of this new design, we aimed to develop a technique allowing localization of bioactive tau directly on tissues. We applied lysates from our next generation biosensor cells on frozen human brain tissue sections. We found that unlike the previous construct, our new fluorescent tau probes present in the cell lysates were recruited on AD tissue and were binding specifically to tau aggregates. Incubation of AD brain slices with formic acid inhibit the recruitment of the probe but not the binding of tau antibodies. The probe is recruited by neurofibrillary tangles, dystrophic neurites and neuritic plaques. Interestingly, positivity for Tau immunostaining and the probe were not strictly colocalized meaning that only some Tau aggregates were detected with the probe. Moreover, we

do not observe positive staining with the probe on brains from control, progressive supranuclear palsy, corticobasal degeneration and Pick disease patients, suggesting that the probe detects a unique conformation of tau present in AD tissues only. We next tried to incubate AD brain and control brains lysates with biosensor cell lysate and analyzed high molecular weight proteins extracted by fast protein liquid chromatography. By transmission electron microscopy, we observed colocalization of Tau probe with tau antibody on small protein aggregates but not on paired helical filaments. We also observed the presence of the probe by western blot on high molecular weight fraction of AD brains but not on low molecular weight fractions. This technique demonstrates the presence of seeding-competent tau in-situ in tissues and also demonstrates that the design of tau probe has to be specific to each tauopathy.

Disclosures: **R.C. Perbet:** None. **A. Lathuiliere:** None. **C. Glynn:** None. **A. Anastasie Mate de Gerando:** None. **B.T. Hyman:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Abbvie. F. Consulting Fees (e.g., advisory boards); Dewpoint therapeutic.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.14

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: JST university fellowships JPMJFS2139
Grant support from the Takeda Science Foundation

Title: Microtubule affinity-regulating kinase 4 implements pathological tau misbehavior through interaction with stress granules

Authors: *G. SULTANAKHMETOV, S. NAKAJIMA, A. FUKUCHI, A. ASADA, T. SAITO, K. ANDO;
Tokyo Metropolitan Univ., Tokyo, Japan

Abstract: Abnormalities in tau proteins lead to their accumulation and aggregation, which is believed to play a significant role in the pathogenesis of neurodegenerative diseases such as Alzheimer's disease. Although the association of tau toxicity with its phosphorylation in microtubule-binding repeats by microtubule affinity-regulating kinases (MARKs) was established, it is unclear whether and how each member of the human MARKs (MARK1-4) affects tau toxicity in vivo. In contrast to other human MARKs, the MARK4 level increases in response to stress, such as ischemic injury in mice (Hayden et al., 2019). One of the major stress responses is the formation of stress granules (SGs), temporary dense structures of RNA binding proteins (RBPs) and RNAs. In this study, we aimed to investigate the role of MARK4 in tau regulation by focusing on the interaction MARK4 with SGs and RBPs. First, to elucidate

similarities and differences among the effect of MARKs on tau toxicity, we established transgenic Drosophila expressing human MARK1-4 with a site-specific integration system. Each MARK was co-expressed with human tau that caused neurodegeneration in fly eyes using a pan-retinal driver GMR-GAL4. We found that tau co-expressed with MARK4 showed more prominent neurodegeneration than other MARKs. Only MARK4 increased the total level of tau observed by western blotting, while all MARKs promoted tau phosphorylation at Ser262 located in the microtubule-binding repeat. In HeLa cells, we found that MARK4 expression, but not MARK2, caused the formation of puncta colocalized with SG markers such as fragile X mental retardation 1 (FMR1) protein, Ras GTPase-activating protein-binding protein 1 (G3BP1), and T-cell intracellular antigen 1 (TIA1). Inhibition of liquid-liquid phase separation by 1,6-hexanediol reduced these puncta, suggesting they are SGs. When tau was co-expressed with MARK4, tau was sequestered in SG with MARK4. Finally, we analyzed the role of SG in adverse effects on tau-induced neurodegeneration caused by MARK4 in Drosophila. Knockdown of SG-associated RBPs such as Fmr1, Rox8 (Drosophila homolog of TIA1), and Ataxin-2 (Atx2) ameliorated neurodegeneration, to note Rox8 knockdown had a more negligible effect. Western blot analysis showed that Atx2 knockdown suppressed tau accumulation caused by MARK4. Interestingly, knockdown of Fmr1 and Atx2, but not Rox8, lowered MARK4 protein levels. Taken together, our results suggest that MARK4 promotes the formation of SGs, which play a role as a catalyzing medium for MARK4 and tau interactions. We propose a novel mechanism by which SG formation stimulates tau abnormality in the pathogenesis of neurodegenerative diseases.

Disclosures: G. Sultanakhmetov: None. **S. Nakajima:** None. **A. Fukuchi:** None. **A. Asada:** None. **T. Saito:** None. **K. Ando:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.15

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: 2ULTR001442-061
P30 AG062429-02S1

Title: Testosterone Supplementation modulates Tau Pathogenesis in female Tauopathy models

Authors: *D. MUNOZ-MAYORGA[1], J. GIRARDINI[1], A. S. KAUFFMAN[1], R. A. RISSMAN[1,2], X. CHEN[1];
[1]Neurosciences, UCSD, La Jolla, CA; [2]VA San Diego Healthcare Syst., La Jolla, CA

Abstract: Approximately 2/3 of patients living with Alzheimer's disease (AD) in the US are women. Women also present a higher pathological tau load, as demonstrated by a variety of well-established biomarkers. Why are women more vulnerable to tau pathology and how this vulnerability might be therapeutically exploited is currently unknown. Interestingly, recent work

has found a significant relationship between lower testosterone levels and higher levels of pathological tau in the cerebrospinal fluid of APOE4 carriers, particularly in women. Research in females has mostly focused on the modulation of tau pathology by the major female sex hormones estrogen and progesterone; whether testosterone can positively regulate tau pathology in the context of female physiology is poorly understood. To test the hypothesis that testosterone protects against tau pathophysiology in females, we performed testosterone supplementation in both a brain slice culture model and in 4-6 month-old P301S female mice. Our preliminary results showed a reduction of phosphorylated tau and total tau levels in both the ex vivo and in vivo models. Furthermore, using primary neuronal cultures, we found testosterone, but not estrogen (E2), reduces the amount of tau released to the media. These results highlight testosterone as a regulator of tau pathology in females and pave the way for further research into its potential protective mechanisms and interactions with risk factors such as ApoE4. Closing this gap in the field might explain the sex difference in the vulnerability of AD, and uncover a potential use of testosterone as a therapeutic strategy in AD and related dementia.

Disclosures: **D. Munoz-Mayorga:** None. **J. Girardini:** None. **A.S. Kauffman:** None. **R.A. Rissman:** A. Employment/Salary (full or part-time):; VA San Diego Healthcare System, La Jolla, CA. **X. Chen:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.16

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: NIH Grant 1RF1AG062171-01

Title: Nucleocytoplasmic Transport and Nuclear Pore Disruption in Tauopathies

Authors: *L. RUAN[1], J. CHEW[2], C. N. COOK[3], L. PETRUCELLI[4], J. D. ROTHSTEIN[5]; [1]Johns Hopkins Med. Institutions, Baltimore, MD; [2]Dept. of Neurology, Johns Hopkins Univ. Sch. of Med., Baltimore, MD; [3]Neurosci., [4]Mayo Clin., Jacksonville, FL; [5]Brain Sci. Inst., Johns Hopkins Univ., Baltimore, MD

Abstract: As human life expectancy continues to increase, the two most common dementias, Alzheimer's Disease (AD) and Frontotemporal Dementia (FTD), are becoming more prevalent with no cure and no effective medicine to significantly alter disease progression. When compared to intercellular amyloid plaques, intraneuronal accumulations of neurofibrillary tangles constituted by insoluble, filamentous, hyperphosphorylated Tau is better correlated with neuronal loss and the clinical progression of AD. In addition, the burden of Tau aggregation is also positively associated with the progression of Frontotemporal lobar degeneration (FTLD), the pathologic diagnosis for clinical FTD. However, it remains unclear how Tau accumulation contributes to the devastating neuronal dysfunction and/or death characteristic of these disorders.

Therefore, understanding the cellular mechanisms underlying Tau-mediated neuronal toxicity during aging may provide much needed therapeutic targets. Our previous studies discovered that hyperphosphorylated Tau interacts with nucleoporins and disrupts the nuclear pore complex (NPC) (Neuron, 2018). The pathological accumulation of Tau in mice and human patients impedes nuclear transport and may disrupt the NPC. Furthermore, our studies in Amyotrophic Lateral Sclerosis (ALS) have suggested that rescuing the NPC injuries prevents various defects in human disease models. Here, we sought to determine the cellular and molecular mechanisms underlying Tau mediated nuclear pore injuries in AD and FTD-Tau. We found that nucleoporin 210 (NUP210) mislocalized to cytosol in both AD patients and cell models with light-dependent Tau protein oligomerization. Remarkably, NUP210 accelerates the fibrilization of Tau protein AD core in vitro. Tau mutation carrying AD patients-derived pluripotent stem cells induced cortical neurons also showed reduced Nup210 in the NPC. In addition, we provide evidence suggesting that FG-repeat containing nucleoporin NUP62 also directly interacts with pTau both *in vivo* and *in vitro*, whereas other nucleoporins such as NUP50 and NUP54 were not affected. Currently, we are evaluating if reducing levels of NUP210 or NUP62 in rodent disease models would reduce/delay pTau accumulation. Moreover, we will determine how loss of NUP210 and NUP62 from NPC in tauopathies specifically affect nuclear transport of protein and RNA. Eventually, we will develop mechanism-based therapy to rescue NPC defects, in order to ameliorate neuronal injury in tauopathies.

Disclosures: L. Ruan: None. **J. Chew:** None. **C.N. Cook:** None. **L. Petrucelli:** None. **J.D. Rothstein:** None.

Poster

535. Tau: Cellular and Molecular Mechanisms II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 535.17

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

Support: NIH AG054345
NIH AG054753
IUSM, Precision Health Medicine Program Grant

Title: Inpp5d modulates tau pathology in vivo

Authors: *A. L. OBLAK[1], A. LEE-GOSSELIN[2], D. SONI[3], B. T. LAMB[1], C. A. LASAGNA-REEVES[4];
[1]Stark Neurosciences Res. Inst., Indianapolis, IN; [2]Indiana University-Purdue Univ. Indianapolis, Indianapolis, IN; [3]Med. neuroscience, [4]Anatomy, Cell Biol. and Physiol., Indiana Univ. Sch. of Med., Indianapolis, IN

Abstract: A non-coding variant of inositol polyphosphate-5-phosphatase D (INPP5D), rs35349669; OR=1.08, 95%CI=1.06-1.11, increases the risk of late-onset Alzheimer's disease

(AD). Recently, the interaction of INPP5D on beta-amyloid pathology has been investigated, but the effect of INPP5D on tau pathology is unknown. Here, we used an adeno-associated virus (AAV) *in vivo* model to drive tau expression with the human P301L mutation. INPP5D wild-type (WT) and heterozygous female and male mice were used. In this study, we aim to determine the role of INPP5D in tau pathogenesis, and its cellular and molecular mechanisms. At P0, mice were subject to intracerebroventricular bilateral injection of AAV-tauP301L or AAV-eGFP (control). Six months later, brain tissues were harvested for histological analysis, biochemical analyses, and gene expression using NanoString. In the thalamus and striatum, histological analysis revealed decrease PHF-1 (tau phosphorylation Ser396/Ser404) tau staining in $INPP5D^{+/-}$ compared to $INPP5D^{+/+}$ mice. We also show that phosphorylated tau at Thr231 and disease-specific conformational modification of tau (MC1) is reduced in $INPP5D^{+/-}$ mice. Next, in order to identify a pathway or mechanism involved in alterations of tau pathogenesis, we utilized the mouse neuroinflammation panel (NanoString) and found that multiple pathways (e.g. microglia and astrocyte function, inflammatory signaling, and adaptive immune response) were upregulated in the $INPP5D^{+/-}$ mice injected with AAV-tauP301L compared to the WT. Neuroprotective genes, such as TNF-β1, IL6RA, RAPGEF3 and HSPB1, were also upregulated. Our findings suggest that INPP5D modulates tau pathogenesis *in vivo* by upregulating microglial/astrocytic function and genes that serve a neuroprotective role. These findings provide more insight of the role of INPP5D in tauopathy.

Disclosures: A.L. Oblak: None. **A. Lee-Gosselin:** None. **D. Soni:** None. **B.T. Lamb:** None. **C.A. Lasagna-Reeves:** None.

Poster

536. Tau and Tauopathies: Human

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 536.01

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

Support: Medical Research Council Centres of Excellence in Neurodegeneration CoEN5025
European Research Council 681181
UK Dementia Research Institute

Title: Oligomeric, phosphorylated, and misfolded tau species accumulate in pre and post synapses of Alzheimer's disease brain even in areas without substantial neurofibrillary tangle pathology

Authors: M. COLOM-CADENA[1], C. DAVIES[1], S. SIRISI[2], L. JI-EUN[1], E. M. SIMZER[1], M. QUEROL-VILASECA[3], E. SANCHEZ-ACED[2], Y. CHANG[1], R. MCGEACHAN[1], J. ROSE[1], J. TULLOCH[1], C. SMITH[1], T. ANDRIAN[2], S. PUJALS[2], R. KAYED[4], M. HORROCKS[1], A. LLEO[5], ***T. L. SPIRES-JONES**[6];
[1]The Univ. of Edinburgh, Edinburgh, United Kingdom; [2]Hosp. de Sant Pau, Univ. Autònoma de

Barcelona, Barcelona, Spain; [3]- Hosp. de Sant Pau, Univ. Autònoma de Barcelona, Barcelona, Barcelona, Spain; [4]Univ. of Texas Med. Br., Univ. of Texas Med. Br., Galveston, TX; [5]Hosp. Santa Creu I Sant Pau, Hosp. Santa Creu I Sant Pau, Barcelona, Spain; [6]The Univ. of Edinburgh, Univ. of Edinburgh, Edinburgh, United Kingdom

Abstract: Deposition of fibrillar forms of tau -neurofibrillary tangles and neuropil threads- follow a hierarchical progressive pattern across brain regions in Alzheimer's disease (AD) and this progression is associated with cognitive decline. Synapses loss occurs early in disease progression and correlates even more strongly with cognitive decline, but whether pathological oligomeric or phosphorylated tau accumulates at synapses where it could contribute to synapse loss or spreads through the brain via synaptic connections remains unclear in human brain. We used sub-diffraction microscopy techniques (array tomography, electron microscopy, STORM and DNA-PAINT) to visualize single synaptic terminals in post-mortem human samples from temporal and occipital cortex of 29 AD and 24 control cases without dementia. Pre- and post-synaptic terminals (synaptophysin and PSD95) and oligomeric, misfolded, or phosphorylated tau (T22, Alz50 or AT8 antibodies) were labelled. To compare synaptic tau with total fibrillar tau burden, neurofibrillary tangles and neuropil threads were quantified from immunohistochemical sections (AT8 antibody) of the same cases. We observed oligomeric, phosphorylated, and misfolded tau accumulate in synapses in Alzheimer's disease brain. A subset of pre and postsynaptic paired synapses contain tau and synaptic tau was observed in areas without abundant tangles, indicating that tau pathology may spread through synapses. Together, these data indicate that targeting synaptic tau may prevent the spread of pathology through the brain.

Disclosures: M. Colom-Cadena: None. **C. Davies:** None. **S. Sirisi:** None. **L. Ji-Eun:** None. **E.M. Simzer:** None. **M. Querol-Vilaseca:** None. **E. Sanchez-Aced:** None. **Y. Chang:** None. **R. McGeachan:** None. **J. Rose:** None. **J. Tulloch:** None. **C. Smith:** None. **T. Andrian:** None. **S. Pujals:** None. **R. Kayed:** None. **M. Horrocks:** None. **A. Lleo:** None. **T.L. Spires-Jones:** F. Consulting Fees (e.g., advisory boards); SAB Cognition Therapeutics.

Poster

536. Tau and Tauopathies: Human

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 536.02

Title: WITHDRAWN

Poster

536. Tau and Tauopathies: Human

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 536.03

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

Support: NIH Grant R01AG058533-01A1

Title: Improved delineation of the reference region for 18F-PI-2620 Tau PET analyses

Authors: *N. N. LEE[1], K. V. WHEELER[1], M. A. TUBI[1], J. A. TERNER[1], B. J. HALL[1], A. M. TWISSELMANN[1], A. W. TOGA[1], S. E. O'BRYANT[2], N. ROÉ-VELLVÉ[3], M. N. BRASKIE[1]; [1]Imaging Genet. Ctr., USC, Los Angeles, CA; [2]Inst. for Translational Res., Univ. of North Texas, Fort Worth, TX; [3]Life Mol. Imaging GmbH, Berlin, Germany

Abstract: Alzheimer's disease is a progressive neurodegenerative disease characterized by the accumulation of extracellular β-amyloid plaques and intracellular hyperphosphorylated tau neurofibrillary tangles. Tau positron emission tomography (PET) quantitatively measures regional tau load by comparing tracer uptake in regions of interest to that of a reference region where signal is not expected to arrive at standardized uptake value ratios (SUVR). 18F-PI-2620 is a novel PET tracer that has improved binding specificity over its predecessors and exhibits high binding affinity for aggregated tau. Past 18F-PI-2620 PET work has used cerebellar gray matter excluding vermis and anterior lobe of the cerebellum as a reference region. In our larger study, we found that non-specific tracer uptake still occurred within the reference region in approximately 23% of brains out of an initial sample of 321 subjects, which may affect SUVR. Our aim was to identify a reference region that allowed maximum inclusion of subjects, without biasing results by non-specific uptake. To create our initial reference region mask, we used the Cerebellum-MNIfnirt-maxprob-thr25-2mm cerebellum template from the Montreal Neurological Institute (MNI) cerebellum atlas, and included only the posterior lobe of the cerebellum. We then coregistered the posterior cerebellum in MNI space to the cerebellar gray matter in Freesurfer space to create our posterior cerebellar gray matter mask. Our revised reference region includes posterior cerebellar gray matter, but excludes all regions superior to the left and right Crus I lobule. A two-tailed t-test between the medial temporal lobe SUVR of the uncontaminated initial and revised reference regions showed no significant differences ($t = -1.50$ $p = 0.13$), with an average SUVR difference of 0.014. This indicated that the new mask avoided contamination without significantly changing apparent tau SUVR. With our new reference region, we have achieved a 100% pass rate out of our final sample of 661 subjects, establishing a viable tau PET processing pipeline that restricts possibly confounding uptake in the reference region.

Disclosures: N.N. Lee: None. **K.V. Wheeler:** None. **M.A. Tubi:** None. **J.A. Terner:** None. **B.J. Hall:** None. **A.M. Twisselmann:** None. **A.W. Toga:** None. **S.E. O'Bryant:** None. **N. Roé-Vellvé:** A. Employment/Salary (full or part-time):; Life Molecular Imaging GmbH. **M.N. Braskie:** None.

Poster

536. Tau and Tauopathies: Human

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 536.04

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

Title: Characterization of the interplay between Musashi and Tau aggregation in human AD brains

Authors: *A. ADAY[1], M. MONTALBANO[2,3], L. FUNG[2,3], M. KIDD[2,3], C. JEREZ[2,3], N. N. BHATT[2,3], E. GEORGE[2,3], R. KAYED[2,3];
[1]M.D./Ph.D. combined degree program, [2]Dept. of Neurol., [3]Mitchell Ctr. for Neurodegenerative Disorders, Univ. of Texas Med. Br., Galveston, TX

Abstract: Tauopathies, such as Alzheimer's disease (AD), are characterized by the accumulation of toxic tau oligomers in the brain. Recent studies suggest that AD also exhibits pathology related to RNA binding proteins (RBPs), including Musashi proteins. Our lab has previously discovered that Musashi 1 (MSI1) and Musashi 2 (MSI2) mislocate and aggregate in human AD brains. We have also visualized co-localization between oligomeric tau and MSI 1 and MSI2, suggesting toxic direct cross-talk between tau and MSI proteins. Recently, it has been shown that tau aggregates represent distinct polymorphs (conformers). With this discovery and the growing evidence indicating RBPs as contributors to AD pathology, it is crucial to elucidate how the interactions between tau and MSI influence tau aggregation and perhaps formation of distinct polymorphs. In this study, we further characterize which tau oligomeric polymorphisms interact with MSI within the human AD brain. We investigate the abundance and cellular location of MSI and tau aggregates in AD and age matched control human cortical brain tissues though immunoblotting and immunofluorescence (IF). We performed western blotting, dot blotting, and filter-trap assays using commercial and in-house antibodies to qualitatively investigate MSI1, MSI2, and tau polymorphs across our sampled population. IF was performed to investigate the spatial and quantitative distribution of MSI1, MSI2, and tau polymorphs. 3D rendering and aggregate morphological characterization was conducted with IMARIS 9.9 Explorer software. For the first time, we show that the presence and quantity of tau oligomeric polymorphisms and MSI aggregates vary across the sampled patient population. We also identify the differential association of tau conformers with MSI1 and MSI2. Further investigations are warranted to determine the temporal and regional distribution of these conformers in association with MSI proteins as well as the mechanisms underlying their aggregation.

Disclosures: A. Aday: None. **M. Montalbano:** None. **L. Fung:** None. **M. Kidd:** None. **C. Jerez:** None. **N.N. Bhatt:** None. **E. George:** None. **R. Kayed:** None.

Poster

536. Tau and Tauopathies: Human

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 536.05

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

Support: The design, study conduct, and financial support for this research were provided by AbbVie. AbbVie participated in the interpretation of data, review, and approval of the publication.

Title: Examining selective vulnerability to tau pathology using human iPSC-derived neurons

Authors: *I. WEIDLING[1], C. PREISS[1], G. SRIVASTAVA[2], L. GIBILISCO[1], M. BRENNAN[1], S. LUDWIG[1], K. NAM[1], J. THOMAS[3], P. PHILIP[3], S. CHANCELLOR[1], T. KWON[1], H. LEE[4], P. REINHARDT[4], N. VENKAT[1], K. YANAMANDRA[1], A. WELKER[1], J. WU[1], X. LANGLOIS[1], J. MANOS[1];
[1]AbbVie, Cambridge Res. Center, 200 Sidney Street, MA 02139, [2]Data and Statistical Sci., AbbVie, Cambridge, MA; [3]Excelra Knowledge Solutions, Hyderabad, India; [4]AbbVie, Deutschland GmbH & Co. KG, Neurosci. Research, Knollstrasse, 67061, Ludwigshafen, Germany

Abstract: Alzheimer's disease (AD) is characterized by a stereotypical progression of tau pathology throughout the brain. Tau alterations typically appear first in the entorhinal cortex followed by the hippocampus and cerebral cortex, while midbrain and hindbrain regions are relatively spared. These neuropathological observations suggest certain regions have a selective vulnerability to the disease processes, but the molecular underpinnings of selective vulnerability have yet to be well defined. Human induced pluripotent stem cell (hiPSC) technology offers the ability to derive unique neuronal subtypes from the same genetic background. Here, we generated hiPSC-derived neuronal populations with varying propensities to form tau aggregates following seeding with sarkosyl-insoluble (AD) brain-derived pathological tau (SI-AD tau) to identify potential mediators underlying differential vulnerability to tau seeding. We differentiated commercially available hiPSC lines derived from apparently healthy donors (3 female, 2 male, ages 15-64) into forebrain, midbrain, and hindbrain populations resembling AD vulnerable and resilient neuronal subtypes. Protein and RNA markers specific to each neuronal subtype were measured via immunofluorescence (IF) and qPCR. An in-house, well-established tau aggregation high content imaging assay evaluated each neuronal population's propensity to establish tau pathology following seeding. Finally, we performed bulk RNA sequencing of hiPSC-derived neurons following treatment with SI-AD tau. qPCR and IF staining revealed distinct expression of brain region- and neuronal subtype-specific markers for each hiPSC-derived neuronal population. The unique neuronal subtypes showed differential vulnerability to tau aggregation following seeding with SI-AD tau, with the cortical inhibitory-like population showing the highest levels of endogenous tau aggregation and with high concordance between donor lines. Preliminary analysis of bulk RNA-seq data from susceptible and resilient populations found distinct transcriptional changes following seeding with SI-AD tau as well as inherent differences between subtypes. Future experiments aim to further define and evaluate the transcriptomic signatures associated with vulnerable and resistant neurons, which could provide more relevant targets for future drug discovery efforts in Alzheimer's disease.

Disclosures: I. Weidling: A. Employment/Salary (full or part-time):; AbbVie. **C. Preiss:** A. Employment/Salary (full or part-time):; AbbVie. **G. Srivastava:** A. Employment/Salary (full or part-time):; AbbVie. **L. Gibilisco:** A. Employment/Salary (full or part-time):; AbbVie. **M. Brennan:** A. Employment/Salary (full or part-time):; AbbVie. **S. Ludwig:** A. Employment/Salary (full or part-time):; AbbVie. **K. Nam:** A. Employment/Salary (full or part-time):; AbbVie. **J. Thomas:** B. Contracted Research/Research Grant (principal investigator for a

drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; AbbVie. **P. Philip:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; AbbVie. **S. Chancellor:** A. Employment/Salary (full or part-time):; AbbVie. **T. Kwon:** A. Employment/Salary (full or part-time):; AbbVie. **H. Lee:** A. Employment/Salary (full or part-time):; AbbVie. **P. Reinhardt:** A. Employment/Salary (full or part-time):; AbbVie. **N. Venkat:** A. Employment/Salary (full or part-time):; AbbVie. **K. Yanamandra:** A. Employment/Salary (full or part-time):; AbbVie. **A. Welker:** A. Employment/Salary (full or part-time):; AbbVie. **J. Wu:** A. Employment/Salary (full or part-time):; AbbVie. **X. Langlois:** A. Employment/Salary (full or part-time):; AbbVie. **J. Manos:** A. Employment/Salary (full or part-time):; AbbVie.

Poster

536. Tau and Tauopathies: Human

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 536.06

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

Title: Widespread brain neurofibrillary tau tangles and negative association with aging

Authors: *S. HOJJATI[1], F. FEIZ[1], S. NAYAK[1], J. SHTEINGART[1], S. OZORIA[1], A. FERNÁNDEZ GUERRERO[1], D. DEVANAND[2], J. LUCHSINGER[3], Y. STERN[4], Q. R. RAZLIGHI[1];
[1]Radiology, Cornell University: Weill Cornell Med. Col., New York, NY; [2]Psychiatry, [3]Geriatric psychiatry, [4]Neurol., Columbia Univ. Irving Med. Ctr., New York, NY

Abstract: The aggregation of neurofibrillary tau tangles is one of the well-documented pathologies for Alzheimer's disease (AD). However, the low-level tangle accumulation, particularly in the medial temporal lobe, is also reported in aging studies. Disentangling age-related increase in the tau deposition from the pathological alteration of tau aggregation in neurodegeneration is a major challenge in the field. More specifically, few studies reported an age-related decrease in tau deposition for clinical populations with abnormally elevated levels of tau and Ab depositions. This study aims to investigate the negative age association with tau tangles and introduce it as a distinctive biomarker for identifying pathological tau accumulation from normal aging. We used 394 healthy control (HC) and 52 mild cognitive impairments (MCI) participants with amyloid and tau positron emission tomography (PET) scans acquired at Columbia University Irving Medical Center and Weill Cornell Medicine. To replicate the results, we also took advantage of 151 HC and 116 MCI participants from Alzheimer's Disease Neuroimaging Initiative data with different PET tracers. For each dataset participants' regional amyloid and tau (26 regions) standard uptake value ratios (SUVRs) were fed into agglomerative clustering to identify groups with unique spatial-temporal characteristics. In both datasets, participants were clustered into three groups (with Euclidean distance measure>5). Next, a

regional multiple linear regression model is performed for each group to detect any negative age-related association with tau SUVRs (controlling for gender, intracranial volume, and regional Amyloid SUVR). In both datasets, regional tau in one cluster of participants showed a significant ($p<0.01$) negative age-related association (in regions like precuneus, and superior frontal) whereas another cluster showed a significant positive age-related association (in regions like middle temporal, Entorhinal cortex) and the last cluster showed almost no association with age. In comparison to the other two clusters, the group that showed the negative age-related association had a significantly higher tau and Ab burdens and also had a significantly higher number of MCI patients ($p<0.001$). These results suggest that while the initial accumulation of tau increases with age, at the higher level of Ab and tau deposition age becomes associated with a decrease in tau deposition. In other words, age-related tau accumulation might be discriminated from the pathological accumulation of Tau where the negative age-related association is observed.

Disclosures: S. Hojjati: None. **F. Feiz:** None. **S. Nayak:** None. **J. Shteingart:** None. **S. Ozoria:** None. **A. Fernández Guerrero:** None. **D. Devanand:** None. **J. Luchsinger:** None. **Y. Stern:** None. **Q.R. Razlighi:** None.

Poster

536. Tau and Tauopathies: Human

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 536.07

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

Title: Isolation and characterization of tau oligomers

Authors: *C. JEREZ[1,2], L. FUNG[1,2], M. KIDD[1,2], N. BHATT[1,2], U. SENGUPTA[1,2], R. KAYED[1,2];
[1]Neurol., [2]Mitchell Ctr. for Neurodegenerative Dis., Univ. of Texas Med. Br., Galveston, TX

Abstract: Isolation and Characterization of Tau Oligomers
Cynthia Jerez[1,2]**,** Leiana Fung[1,2], Madison Kidd[1,2], Urmi Sengupta[1,2], Nemil Bhatt[1,2], Urmi Sengupta[1,2,] Rakez Kayed[1,2]
[1]Department of Neurology, Neuroscience and Cell Biology, University of Texas Medical Branch, Galveston, TX, 77555, USA
[2]Mitchell Center for Neurodegenerative Diseases, University of Texas Medical Branch, Galveston, TX, 77555, USA
Tau oligomers have been shown to be the main toxic tau species in thea diverse group of age-related neurodegenerative diseases, collectively known as tauopathies such as Alzheimer's disease (AD), progressive supranuclear palsy (PSP), and dementia with Lewy Bodies (DLB). The characteristics of oligomers that exist in human disease, and that may be most closely associated with disease propagation and toxicity, are not fully established because of the lack of standardization of the preparation of small oligomeric tau aggregates and the methods for their

characterization. T Standardization of these oligomers can be achieved by isolating tau aggregates from authentic human tauopathy cases such as AD. By rigorously correlating their biophysical and biochemical properties with biological activity, developing probe sets for their selective detection, and disseminating reliably examined samples, lab-ready established protocols will be available to the broader research community. To acquire biologically relevant tau oligomers for the study of tau aggregation and mechanisms of toxicity, we have designed and optimized protocols for the preparation and characterization of tau oligomers *in vitro* using other amyloid oligomeric seeds, as well as for the isolation of tau oligomers from biological samples using immunoprecipitation and sucrose gradient fractionationcentrifugation.. We have also used commercially available and created novel antibodies and optimized techniques for the detection of tau oligomers using common biochemical techniques including ELISA, dot blot, western blot, filter trap assay, as well as immunohistochemistry, Fluorescent Amyloid Multi Emission Spectra (FLAMES), and proteolytic digestion by proteinase- K enzyme, were used to characterize oligomeric tau from recombinant protein and isolated from AD brain. Establishing defined characteristics of biologically active disease-relevant tau oligomers will bewill be invaluable for developing oligomer specific diagnostics and therapeutics.

Disclosures: C. Jerez: None. **L. Fung:** None. **M. Kidd:** None. **N. Bhatt:** None. **U. Sengupta:** None. **R. Kayed:** None.

Poster

536. Tau and Tauopathies: Human

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 536.08

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

Support: Sarah Roush Memorial Fellowship in Alzheimer's Disease
Rainwater Foundation Tau Leadership Award
IU Precision Health Initiative
NIH Grant 1R01AG059639
NIH Grant 1R01NS119280
NIH Grant 1K22NS092688
Alzheimer's Association (AARG-D591887)

Title: Resilience to Alzheimer's pathology is associated to a decrease Tau pathological features and distinct neuroinflammatory response

Authors: *N. JURY GARFE[1], Y. YOU[1], J. REDDING[3], P. MARTINEZ[1], H. KARAHAN[2], A. MANABAT[1], J. KIM[2], J. TRONCOSO[3], C. A. LASAGNA-REEVES[1];
[1]Anatomy, Cell Biol. and Physiol., [2]Med. and Mol. Genet., Stark Neurosci. Res. Inst., Indianapolis, IN; [3]Pathology and Neurol., Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: The neuropathologic hallmarks of Alzheimer disease (AD) are Aβ-amyloid deposits and neurofibrillary tangles (NFTs). Asymptomatic AD (AsymAD) is described as the status of subjects with preserved cognition assessed but with identifiable AD pathology at autopsy. Currently there is no clear understanding of the mechanisms underlying resilience. Considering that decreased inflammatory response and the tau pathology seems to be uncoupled from neuron death in AsymAD compared to typical demented AD, we conducted a detailed histological and biochemical analysis using postmortem brain samples from age-matched control, demented AD, and AsymAD subjects. Histological examination reveals a lower accumulation of filamentous Thioflavin S structures and an increase in glia reactivity around core plaques in AsymAD cases compared to AD. Biochemical characterization identified a significant decreased in phospho-tau species and seeding activity in AsymAD compared to AD cases. To assess the molecular weight and filament structure of the tau species involved in seeding activity, we performed size exclusion chromatography (SEC) on TBS-soluble brain extracts followed by co-immunoprecipitation and electron microscopy. Using subcellular fractionation, we found accumulation of seed-competent tau predominantly in cytosolic compartment in AsymAD samples, while in AD cases the distribution between cytosolic and synaptic fractions was similar. AsymAD and typical demented AD does not show differences in cytosolic nor synaptic Aβ42. Multiple Nanostring neuroscience panels indicate healthier neuronal context in AsymAD than AD. Our data suggest that the main differences between non-demented individuals with high loads of AD's pathology and typical demented AD patients lies in the structural diversity of tau aggregates, its ability to reach the synapse, and distinct neuroinflammatory response.

Disclosures: **N. Jury Garfe:** None. **Y. You:** None. **J. Redding:** None. **P. Martinez:** None. **H. Karahan:** None. **A. Manabat:** None. **J. Kim:** None. **J. Troncoso:** None. **C.A. Lasagna-Reeves:** None.

Poster

536. Tau and Tauopathies: Human

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 536.09

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

Support: NIH/NIH K23 Award 5K23AG059919-04
Alzheimer's Association Award 2019-AACSF-644153

Title: Focal Amygdalar Tau in Preclinical Alzheimer's Disease

Authors: *S. TUN[1], T. TOYONAGA[2], B. FICEK-TANI[3], E. C. MORMINO[4], C. FREDERICKS[1];
[1]Neurol., Yale Med. Sch., New Haven, CT; [2]Radiology & Biomed. Imaging, [3]Neurol., Yale Sch. of Med., New Haven, CT; [4]Neurol. and Neurolog. Sci., Stanford, Stanford, CA

Abstract: One of the first brain regions impacted by tau pathology in early-stage Alzheimer's disease (AD) is the amygdala, which plays a critical role in human social and emotional functioning. While pathologic series show that abnormal tau can be found in the amygdala as early as Braak stage 3, the presence of focal amygdala tau and its implications for functional connectivity and neuropsychiatric symptoms in living early-stage patients are unknown. The most heavily impacted nuclei by tau tangles include the medial division nuclei, which participate in the default mode network (DMN), a network which is implicated in mnemonic and social function and shows early abnormalities in AD. Conversely, amygdalar regions spared by AD pathology include the dorsal division nuclei, which are functionally related to a social salience network that shows increased activity in early-stage AD (Bickhart et al. 2012). We hypothesized that tau PET would reveal significant increases in tau binding in whole amygdala, and specifically in the medial division of the amygdala, between participants with preclinical AD and healthy controls (HC). We examined n=297 individuals (242 preclinical, 55 HC) with anatomical MRIs and [18F]flortaucipir tau PET (scan time window: 80-110 min) from the A4 Study, a clinical trial focused on individuals with preclinical AD. Preclinical subjects were those with a positive amyloid scan (determined per A4 study guidelines as a quantitative standardized uptake value ratio (SUVr) of 1.15 (or 1.10 with positive qualitative visual read)). All anatomical MRIs underwent Freesurfer cortical reconstruction to generate customized anatomical surfaces for each subject. Regional SUVrs for tau PET were derived using PETsurfer, an implementation added in Freesurfer 6.0. We additionally performed high-resolution segmentation of the amygdala using the amgydalar subfields module included in Freesurfer 7.2 (Saygin and Kliemann 2017). Using independent t-tests, we found increased tau signal in whole left ($t(295) = 3.47$, $p = 0.0005$) and right ($t(295) = 3.15$, $p = 0.001$) amygdala tau deposition in preclinical AD participants compared to HC. Elevated tau was specifically seen in the medial group nuclei (left ($t(295) = 2.89$, $p = 0.004$) and right ($t(186) = 3.58$, $p=0.0003$)) and not in the dorsal nuclei ($p>0.10$ bilaterally), demonstrating that selective accumulation of tau is already present in medial group nuclei during preclinical illness. Future analyses will explore the impact of focal amygdalar tau on functional connectivity within the DMN and neuropsychiatric symptomatology in preclinical AD.

Disclosures: S. Tun: None. **T. Toyonaga:** None. **B. Ficek-Tani:** None. **E.C. Mormino:** None. **C. Fredericks:** None.

Poster

536. Tau and Tauopathies: Human

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 536.10

Title: WITHDRAWN

Poster

536. Tau and Tauopathies: Human

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 536.11

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

Support: NRF-2020R1A5A1019023
NRF-2022R1A2C1004913
KHIDI-HU21C0071
BK21 Four Biomedical Science Program
NRF-2021R1F1A1049169

Title: Pathological tau from Alzheimer's brain disrupts axon initial segment structure and polarized trafficking

Authors: *S. LEE[1,2,3], Y. SUH[1,2,3];
[1]Dept. of Biomed. Sci., [2]Neurosci. Res. Inst., [3]Transplantation Res. Inst., Seoul Natl. Univ. Col. of Med., Seoul, Korea, Republic of

Abstract: Tau is predominantly distributed to the axon and stabilizes microtubules as a microtubule-associated protein. However, tau accumulation, a pathological hallmark of Alzheimer's disease (AD) and other tauopathy, is observed in the somatodendritic compartment. The AD tau filament core (AD tau core) discovered in the brain of AD patients is a tau fibril strain composed of R3 and R4 domains with a propensity for spontaneous self-assembly, and retains the ability to drive aggregation of endogenous tau or itself. The axon initial segment (AIS) is a specialized structure that forms diffusion barrier between the axon and the somatodendritic compartments. AIS plays a critical role in polarized trafficking that distributes the cytoplasmic cargo to distinct neuronal compartments. We therefore hypothesize that the AD tau core may compromise AIS integrity, which may trigger tau missorting and eventually lead to dendrite dysfunction. Using confocal imaging technology in rat primary cultured neurons, we show that the AD tau core is missorted to the dendritic branches and spines. We find that the AD tau core shortens the AIS length and impairs KCl-induced distal shift in the AIS location, suggesting that the AD tau core disrupts AIS structure and function. We then screen the AIS components that are essential for missorting of the AD tau core. Further studies on the function of the AD tau core in dendritic spines will provide a pathologic basis for tauopathy and AD.

Disclosures: S. Lee: None. **Y. Suh:** None.

Poster

536. Tau and Tauopathies: Human

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 536.12

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

Title: Regional population differences of Corpora amylacea in postmortem hippocampal human brain tissue

Authors: *J. DALLMEIER[1], R. T. VONTELL[1], D. DAVIS[1], R. GOBER[1], C. WANDER[2], A. BARREDA[1], X. SUN[1], T. J. COHEN[2], W. K. SCOTT[1];
[1]Dept. of Neurol., Univ. of Miami Miller Sch. of Med., Miami, FL; [2]Dept. of Neurol., Univ. of North Carolina Chapel Hill, Chapel Hill, NC

Abstract: Introduction: Alzheimer disease (AD) is the most prevalent neurodegenerative disorder, affecting over 50 million people worldwide. Severity and distribution of aggregated tau and neurofibrillary tangles (NFT) are strongly correlated with the clinical presentation of AD. Clearance of aggregated tau could decrease rate of NFT formation and delay onset of AD. Recent studies implicate corpora amylacea (CoA) activity in pTau aggregation. Normally, CoA clear brain waste products by amassing cellular debris and then they are extruded into the CSF where they travel to the cervical lymph nodes to be phagocytosed. The proper functioning of CoA may slow progression of AD-associated NFT pathology, and this relationship may be influenced by amount and distribution of pTau produced, age, sex, and genetic risk.**Objective**: In this study, we investigated regional population differences of CoA and determined if area occupied by CA was correlated with the area occupied by pTau pathology in the CA1 & CA3 region of Alzheimer's disease postmortem hippocampal tissue. **Methods**: Postmortem brain hippocampal tissue sections from 11 AD and 4 control donors were immunohistochemically stained with AT8 (pTau Ser202, Thr205) and counter stained with periodic acid schiff (PAS). Stained sections were digitized at 40x using the Motic Easy Scan System. QuPath and ImageJ were used for slide analysis. The percent area occupied (%AO) of each was then calculated by dividing the sum of the areas for all positively stained objects by the area of the ROI, then multiplying by 100. A correlation matrix consisting of CoA %AO, pTau %AO, and neurofibrillary tangle %AO was calculated. A paired t-test was performed between CoA %AO in the CA1 and CA3 hippocampal regions. **Results**: CoA %AO was significantly higher in the CA3 region compared to CA1 in AD donors (mean difference = 0.21 +/- 0.16, p = 0.015). In controls no difference was noted (mean difference = 0.05 +/- 0.11, p = 0.88). Although not statistically significant, we found a negative correlation of CoA %AO with pTau %AO (r = -0.14, p = 0.35), and neurofibrillary tangle %AO (r = -0.28, p = 0.21) in the CA1 region of the hippocampus. **Conclusions**: Our results suggest that more numerous CoA in the CA3 region (compared to CA1) in the hippocampus of AD donors but not controls may suggest CoA play a role in AD pathology. This could be due to increased pTau load in individuals with AD. Although the correlations in this small sample are not statistically significant, a negative correlation would support the idea that CA may play a role in AD pathologic progression by influencing innate tau clearance. These trends are currently being tested in a larger dataset.

Disclosures: J. Dallmeier: None. **R.T. Vontell:** None. **D. Davis:** None. **R. Gober:** None. **C. Wander:** None. **A. Barreda:** None. **X. Sun:** None. **T.J. Cohen:** None. **W.K. Scott:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.01

Topic: C.06. Neuromuscular Diseases

Title: Functional deficits and neuropathology in wild-type and ALS/FTD mutant cyclin F mice

Authors: *A. VAN HUMMEL, M. SABALE, G. CHAN, A. CHAN, L. M. ITTNER, Y. D. KE; Macquarie Univ., Sydney, Australia

Abstract: Amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTD), whilst being distinct neurodegenerative diseases, share underlying genetic causes and histopathological features, with a subset of patients (~15%) experiencing symptoms of both diseases. Approximately 5-10% of patients with ALS and/or FTD have a known family history of hereditary inheritance of disease, and of these, an estimated 0.6-3% are linked to mutations in the *CCNF* gene. *CCNF* encodes the Cyclin F protein, which plays a role in maintaining homeostasis through protein degradation via mediation of ubiquitination. Expression of pathological mutant CCNF ($CCNF^{S621G}$) in cellular models has been shown to lead to overactive ubiquitination and disrupted caspase 3-mediated cell survival pathways. Disrupted caspase 3-mediated cell viability was also observed in a $CCNF^{S621G}$ zebrafish model, along with abnormal axonal outgrowth and motor deficits. We have developed two CCNF mouse models using adeno-associated virus (AAV) to over-express wildtype or mutant (S621G) CCNF in the brains of neonatal C57Bl/6 mice. These mice underwent behavioural and motor testing at 3- and 6-months of age, followed by cognitive testing at 8-months of age. Whilst no motor deficits were present at 3- or 6-months of age, FTD-like symptoms of disinhibition, hyperactivity and altered exploratory patterns were observed in CCNF over-expressing mice in the elevated plus maze and open field tests when compared to GFP-injected controls from 3-months of age. Both wildtype and mutant CCNF over-expression models showed delayed spatial learning acquisition in the Morris Water Maze at 8-months of age and used less hippocampal-dependent swim strategies compared to GFP-injected controls, suggesting impaired cognition. Histological and biochemical post-mortem analyses showed an increase in ubiquitination in aged CCNF over-expressing mice compared to GFP-injected controls, in line with previously published cellular models. Cyclin F has several interaction partners which are associated with ALS/FTD, including splicing factor proline glutamine rich (SFPQ), recently shown to be aggregated and mislocalized in cellular models expressing mutant CCNF. In our CCNF over-expressing mice, we found an increase in insoluble SFPQ, recapitulating findings from neuropathological analyses of ALS/FTD cases. Taken together, our results indicate for the first time in mice that AAV-induced over-expression of wildtype or mutant CCNF is sufficient to drive FTD-like behaviors and ALS/FTD neuropathology and may provide a novel therapeutic target.

Disclosures: A. Van Hummel: None. **M. Sabale:** None. **G. Chan:** None. **A. Chan:** None. **L.M. Ittner:** None. **Y.D. Ke:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.02

Title: WITHDRAWN

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.03

Topic: C.06. Neuromuscular Diseases

Support: NIH Grant 5T32AG000255
NIH Grant 5R01NS11068803

Title: Seizures in a doxycycline-regulatable mouse model of TDP-43 proteinopathy.

Authors: *W. RODEMER, H. JUUL, S. MEHTA, M. FARAG, S. PORTA, D. M. TALOS, V. M.-Y. LEE;
Univ. of Pennsylvania, Philadelphia, PA

Abstract: Neuronal hyperexcitability is a pathophysiological hallmark of ALS yet its relationship to the underlying pathology remains poorly understood. Electrophysiological studies using transcranial magnetic stimulation have consistently identified neuronal hyperexcitability both in the brain and in the periphery of ALS patients. Hyperexcitability is observed early in disease and correlates with cognitive impairment. Importantly, aberrant neuron activity may contribute to neuron death via excitotoxicity. Postmortem studies have revealed that phosphorylated TDP-43 aggregates are a primary component of the cytoplasmic neuronal inclusions found in the majority of sporadic ALS cases. Emerging evidence in tissue slices and dissociated cell models suggest TDP-43 pathology may induce hyperexcitability. However, *in vivo* evidence in animal models has been lacking. To address this gap, we performed long-term continuous EEG in bigenic rNLS8 mice, which express the mutant cytoplasmic human TDP-43 (hTDP-43ΔNLS) under the control of the NEFH promoter in a Tet-OFF inducible fashion. Briefly, adult rNLS8 mice were bilaterally implanted with screw electrodes and connected to a tethered EEG acquisition system (Pinnacle) for up to 8 weeks off-DOX. We observed a robust electrographic seizure phenotype in 66% of the rNLS8 mice recorded (n=6). Seizure onset occurred approximately 2 weeks after transgene induction (range, 11 - 17d), correlating with hTDP-43ΔNLS expression but preceding gross cell death. Interestingly, mice that failed to develop seizures within 3 weeks of transgene induction remained seizure free even into later disease stages (6-8 wk off-DOX). Compared to non-bigenic littermates, rNLS8 mice displayed enhanced hippocampal cFOS immunoreactivity consistent with neuron hyperactivity. Ongoing studies will further assess rNLS8 mice for histological markers of seizures and determine whether seizure activity correlates with neuron loss. Results from these studies will advance our

basic understanding of TDP-43 pathophysiology and may uncover novel pathways for therapeutic development in ALS and related TDP-43 proteinopathies.

Disclosures: **W. Rodemer:** None. **H. Juul:** None. **S. Mehta:** None. **M. Farag:** None. **S. Porta:** None. **D.M. Talos:** None. **V.M. Lee:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.04

Topic: C.06. Neuromuscular Diseases

Support: CGS-M CIHR
CIHR

Title: Development of a zebrafish Stathmin-2 (STMN2) knockout model

Authors: *T. GURBERG[1], G. A. ARMSTRONG[2];
[1]McGill Univ. Integrated Program in Neurosci., Montreal, QC, Canada; [2]Dept. of Neurol. and Neurosurg., Montreal Neurolog. Inst., Montreal, QC, Canada

Abstract: Amyotrophic lateral sclerosis (ALS) is a progressive and fatal neurodegenerative disease characterized by the loss of upper and lower motor neurons leading to paralysis and death within 2-5 years of diagnosis. There is no known cure and FDA-approved treatments have little impact on increasing the quality of life or survival of afflicted patients. The majority (~97%) of ALS cases display mislocalization of the nuclear RNA binding protein TAR-DNA Binding Protein (TDP-43), a vital protein in RNA regulation. Stathmin-2 (STMN2) is a microtubule-associated protein encoded by the *STMN2* gene which contains TDP-43 binding sites in the first intron. Cytoplasmic mislocalization of TDP-43 is believed to result in the mis-splicing of *STMN2* resulting in the production of a truncated and non-functional form of STMN2. Our lab is interested in developing a zebrafish STMN2 knockout (KO) to help characterize STMN2's impact on motor neuron degeneration arising in ALS. In the fish, Stathmin-2 is encoded by two genes, *stmn2a* and *stmn2b*. Using the CRISPR/Cas9 system we injected several hundred zebrafish embryos with guide RNAs targeting both genes. Larvae were raised to sexual maturity and then outcrossed to determine if germline transmission of indels occurred using High Resolution Melting (HRM). Electropherograms of mutants confirm the identification of several lines with the following mutations: an 11 nucleotide deletion (*stmn2a*) and a 5 nucleotide deletion (*stmn2b*), both of which results in the presence of premature stop codons. We will incross these lines to generate a double *stmn2a* and *stmn2b* KO model. Using these models we will characterize the organization of ventral root projections in two day old larvae by immunofluorescent labelling of acetylated tubulin. We will also investigate neuromuscular junction defects through immunolabelling of pre- and post-synaptic markers synaptic vesicle protein 2 (SV2) and α-bungarotoxin conjugated to rhodamine, respectively. We

will compare the STMN2 KO ventral roots with previously developed TDP-43 KO and disease-associated knock-in (KI) models as well as healthy controls. We will characterize motor defects through two day old larval touch responses as well as determine changes in survival. Finally, we will attempt to rescue any motor phenotype through injection of full length *STMN2* mRNA into developing STMN2 KO blastocysts, as well as through drug treatments delivered through the zebrafish's aquatic environment. New treatment options are a desperate need for an ALS community lacking meaningful options, and our model hopes to address this issue while also contributing a novel characterization of STMN2's impact in ALS pathogenesis.

Disclosures: T. Gurberg: None. **G.A. Armstrong:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.05

Topic: C.06. Neuromuscular Diseases

Title: Characterization of TDP-43 delta NLS mice: behavior and biomarker analyses

Authors: *H. B. FERNANDES, I. MORGANSTERN, J. BELTRAN, M. KWAN, J. SHEA, A. GHAVAMI, D. HAVAS, T. HANANIA;
Psychogenics Inc., Psychogenics, Paramus, NJ

Abstract: Amyotrophic lateral sclerosis (ALS) is a fatal neurodegenerative disease characterized by progressive loss of upper and lower motor neurons producing muscle denervation, motor impairments and brain atrophy. Deposition of insoluble cytosolic inclusions of TAR-DNA binding protein (TDP-43) correlates with ALS-related pathology in affected tissues. To study the progression of ALS phenotypes and establish a model for testing therapeutic interventions, we characterized TDP-43ΔNLS mice (Walker et al, 2015). These inducible rNLS8 mice were generated by crossing transgenic mice expressing tTA under the control of the human neurofilament heavy chain (NEFH) promoter with tetO-hTDP-43ΔNLS mice containing a defective nuclear localization motif (ΔNLS). Administration of Dox suppresses expression of total and phosphorylated forms of hTDP-43ΔNLS, rescuing the disease phenotype. TDP-43ΔNLS mice showed dramatic loss of body weight following Dox cessation, increased tremors and hindlimb clasping, impaired gait and muscle strength and decreased survival compared to tTA control mice. EMG assessment of muscle function 4 weeks after DOX removal showed an increase in the latency of muscular contraction and decreased response amplitudes of muscle contractions following motor nerve stimulation in TDP-43ΔNLS animals, impairments whose increasing severity correlated with the amount of time spent off Dox. Histological analysis revealed strong overexpression of TDP43 in perinuclear cytoplasmic inclusions along with deposition of pTDP43 aggregates in multiple brain regions including hippocampus, cerebral cortex, dorsal striatum and cerebellum. This model recapitulates deregulated translocation of TDP43 from nucleus to the cytoplasm, a major pathology seen in ALS patients.TDP-43

pathologies were accompanied by increased expression of inflammatory marker transcripts in cortex, astrogliosis, and microglial activation in affected brain regions. Similar pathologies were detected in spinal cord but at lower level of severity than in the brain. Dramatic elevations in neurofilament light chain, a biomarker of neurodegeneration, were seen in plasma and CSF of TDP-43ΔNLS animals at 10 weeks of age, 5 weeks after Dox withdrawal. In summary, expression of human TDP-43ΔNLS in this mouse model of ALS resulted in the development of cytoplasmic inclusions in multiple brain regions and spinal cord, a loss of murine nuclear TDP-43, progressive loss of muscle strength and function, and motor impairments leading to death. These impairments were observed in both genders. This mouse model of ALS provides a rapid progressive phenotype for testing novel therapeutic strategies.

Disclosures: H.B. Fernandes: None. **I. Morganstern:** None. **J. Beltran:** None. **M. Kwan:** None. **J. Shea:** None. **A. Ghavami:** None. **D. Havas:** None. **T. Hanania:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.06

Topic: C.06. Neuromuscular Diseases

Support: NIH grant AG051513
NIH grant AG047612

Title: Developmental mortality and motor neuron terminal physiology of Drosophila SOD1 mutants are modified by mutations linked to axonal transport

Authors: T. C. D. G. O'HARROW[1,2], *A. UEDA[1], X. XING[1,3], C.-F. WU[1];
[1]Dept. of Biol., Univ. of Iowa, Iowa City, IA; [2]Raymond G. Perelman Ctr. for Cell. and Mol. Therapeut., The Children's Hosp. of Philadelphia, Philadelphia, PA; [3]Univ. of California, Davis, Davis, CA

Abstract: Mutations in the human Cu/Zn superoxide dismutase (SOD1) gene are associated with the motor neuron disease amyotrophic lateral sclerosis (ALS). The *Drosophila melanogaster* SOD1 gene (*Sod*) shares a highly conserved sequence with the human homolog, and *Sod* mutant *Drosophila* display elevated pupal mortality, decreased adult lifespan, and attenuated motor function. The *Drosophila* larval neuromuscular junction is well-adapted for the study of defects in neuromuscular physiology, due to its accessibility for *in vivo* tissue studies, and to a long history of study of neuromuscular transmission-altering genetic mutations. This study examined structural and physiological defects at the neuromuscular synapses of *Drosophila* larvae expressing the hypomorphic *n108* allele (Sod^{n108}) or a knock-in construct of the human ALS-linked *G85R* allele (Sod^{G85R}). Further, we detail substantial alterations of *Sod* mutant mortality and physiology by mutations in the epilepsy and axonal transport-linked gene *Prickle* (*Pk*). Immunostaining of neuronal membrane (anti-HRP) at neuromuscular synapses

in *Sod* mutant larvae revealed presynaptic terminals of abnormal, swollen morphology. Pharmacological treatment of *Sod* mutants with K^+ channel blockers revealed allele-specific motor neuron terminal hyperexcitability, characterized by focal synaptic transmissions of extended duration. These extended transmissions were accompanied by increased Ca^{2+} influx at the presynaptic terminal, imaged with a genetically encoded Ca^{2+}reporter (GCaMP6f). Mutations in the epilepsy and axonal transport-linked gene *Pk* had clear effects on the phenotypes of *Sod* mutants. The hyperexcitability observed at the NMJ of *Sod* mutant larvae was heavily suppressed by a copy of the *prickle-spiny-legs* (Pk^{Sple}) allele. Pk^{Sple} and the *prickle-prickle* (Pk^{Pk}) allele also improved survival or aggravated mortality of *Sod* mutants in a genetic dosage-dependent manner. Altogether, this study highlights alterations in motor synapse morphology and physiology at a developmental stage prior to the early death of *Sod* mutant organisms, along with an influence of axonal transport on the maintenance of neuronal health and survival of the organism throughout developmental stages.
1

Disclosures: **T.C.D.G. O'Harrow:** None. **A. Ueda:** None. **X. Xing:** None. **C. Wu:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.07

Topic: C.06. Neuromuscular Diseases

Support: NS111414

Title: A genetic screen of human neurodegenerative disease alleles identifies factors that drive rapid neuromuscular junction degradation in Drosophila

Authors: *I. SANGHVI[1], S. WU[1], S. PERRY[1], N. TRAN[1,2], D. DICKMAN[1];
[1]Dept. of Neurobio., [2]USC Neurosci. Grad. Program, USC, Los Angeles, CA

Abstract: A variety of neurological and neurodegenerative diseases have been modeled in *Drosophila*, with human disease-associated alleles causing phenotypes that may recapitulate disease etiology. While much insight has been gained through transgenic expression of these alleles in all neurons or the visual system, synaptic function and degeneration in these models has not been systematically assessed at a common synapse. We have systematically screened human neurodegenerative disease models at the *Drosophila* neuromuscular junction (NMJ), a powerful model glutamatergic synapse that permits sophisticated genetic, electrophysiological, and imaging approaches. We expressed disease-related transgenes in motor neurons corresponding to a variety of human disease including Alzheimer's, Parkinson's, Frontotemporal Dementia, ALS, Huntington's, and Myotonic Dystrophy, assessing synaptic bouton number and retractions. Out of ~100 transgenes screened, a handful of lines exhibited significant NMJ degeneration. Detailed morphological and electrophysiological analyses, as well as secondary

behavioral and learning assays, were consistent with severe NMJ degeneration. Finally, we probed whether rapid NMJ degeneration was protected or exacerbated following activation of the dual leucine zipper kinase (DLK) and SARM signaling programs, associated with neural injury and degeneration. Ultimately, we hope to establish well defined *Drosophila* NMJ models of human disease to fully characterize their impacts on synaptic degeneration, function, and plasticity.

Disclosures: I. Sanghvi: None. **S. Wu:** None. **S. Perry:** None. **N. Tran:** None. **D. Dickman:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.08

Topic: C.06. Neuromuscular Diseases

Support: 5R01NS089585-08

Title: Ribosomal profiling of motor neurons degenerating in ALS suggests the involvement of FGF21.

Authors: *W. STANSBERRY[1], E. NEWELL[1], J. SHADRACH[2], B. PIERCHALA[1];
[1]Med. Neuroscience, Anat. Cell Biol. and Physiol., Indiana Univ. Sch. of Med., Indianapolis, IN;
[2]Biol. & Material Sci., Univ. of Mchigan, Ann Arbor, MI

Abstract: The neuromuscular junction (NMJ) is a chemical synapse that is the site of skeletal muscle innervation by spinal motor neurons. This connection between the nervous system and muscle allows for coordinated motor function, and the maintenance of the NMJ is critical for maintaining musculoskeletal homeostasis. Under normal physiological conditions, spinal motor neurons have significant regenerative potential and can regrow axons in response to peripheral nerve injury. In diseases such as amyotrophic lateral sclerosis (ALS), the NMJ is dismantled and motor neurons selectively degenerate resulting in progressive muscle wasting and eventual fatal paralysis. We adapted the RiboTag methodology developed by Sanz *et al*. to perform ribosomal profiling of motor neurons in mice to assess how nerve injury and ALS affect motor neurons *in vivo*. We purified motor neuron-specific transcripts after the sciatic nerve crush model of acute injury, and in the *Sod1*^{G93A} ALS model. We identified 267 transcripts that were upregulated following sciatic nerve crush, and of those transcripts, 38% were also upregulated in 4-month-old *Sod1*^{G93A} ALS mice, demonstrating some translational overlap between regenerative and degenerative processes. However, the majority of upregulated genes in injured (58%) and ALS (78%) conditions were specific for either injury or neurodegeneration, respectively. One of the most highly upregulated transcripts was *Fgf21*, which was only induced in *Sod1*^{G93A} mice. FGF21 is a stress-inducible hormone that is critically involved in glucose turnover, and its expression may cause metabolic disturbances in ALS. Immunolabeling experiments in *Sod1*^{G93A}

and *Tdp43*^{A315T} mice revealed that FGF21 protein is increased both in motor neuron cell bodies and in the periphery in motor axons and muscle. We are evaluating the functions of FGF21 in normal motor neuron/muscle homeostasis and in degenerative mechanisms in motor neurons with the ultimate goal of identifying new therapeutic strategies for ALS.

Disclosures: W. Stansberry: None. **E. Newell:** None. **J. Shadrach:** None. **B. Pierchala:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.09

Topic: C.06. Neuromuscular Diseases

Support: 1 RF1 NS120992-01

Title: Tdp-43 knockdown in mouse model of als leads to dsrna deposition, gliosis, and neurodegeneration in the spinal cord

Authors: *R. A. MILSTEAD[1], C. D. LINK[1], Z. XU[2], C. A. HOEFFER[1];
[1]Univ. of Colorado Boulder, Univ. of Colorado, Boulder, Boulder, CO; [2]Univ. Mass Med. Sch., Univ. Mass Med. Sch., Worcester, MA

Abstract: Transactive response DNA binding protein 43 kilodaltons (TDP-43) is a DNA and RNA binding protein associated with severe neurodegenerative diseases like amyotrophic lateral sclerosis (ALS), which primarily affects motor neurons in the brain and spinal cord. Partial knockdown of TDP-43 expression in a mouse model (the amiR-TDP-43 mice) leads to progressive, age-related motor dysfunction as observed in ALS patients. Work in *C. elegans* suggests that TDP-43 dysfunction can lead to deficits in chromatin processing and the accumulation of double-stranded RNA (dsRNA), potentially activating the innate immune system and promoting neuroinflammation. To test this hypothesis, we used immunostaining to investigate dsRNA accumulation and other signs of CNS pathology in the spinal cords of amiR-TDP-43 mice. Compared to wild-type (WT) controls, TDP-43 knockdown (KD) animals show increases in dsRNA deposition in the dorsal and ventral horns of the spinal cord. Additionally, animals with heavy dsRNA expression show markedly increased levels of both astro- and microgliosis. Interestingly, areas of high dsRNA expression and microgliosis overlap with regions of heavy neurodegeneration, indicating that activated microglia could contribute to the degeneration of spinal cord neurons. Taken together, this study suggests that loss of TDP-43 function could contribute to neuropathology by increasing dsRNA deposition and subsequent innate immune system activation.

Disclosures: R.A. Milstead: None. **C.D. Link:** None. **Z. Xu:** None. **C.A. Hoeffer:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.10

Topic: C.06. Neuromuscular Diseases

Support: Science Foundation Ireland [18/CRT/6214 and 17/JPND/3455]

Title: The spectrum of tRNA derived small RNAs in amyotrophic lateral sclerosis and FTD mouse models

Authors: S. BAINDOOR[1], H. A. Y. GIBRIEL[1,2], P. DONOVAN[1,2], M. T. VENO[3], J. KJEMS[3], E. JIRSTROM[1,2], A. KENNY[1,2], G. NARDO[4], C. BENDOTTI[4], M. C. HOGG[1,2], T. ENGEL[1,2], *J. H. M. PREHN[1,2];
[1]Dept. of Physiol. and Med. Physics, Royal Col. of Surgeons in Ireland, 123 St. Stephen's Green, Dublin 2, Ireland; [2]FutureNeuro SFI Res. Ctr. for Chronic and Rare Neurolog. Diseases, Royal Col. of Surgeons in Ireland, Dublin 2, Ireland; [3]Interdisciplinary Nanoscience Centre, Dept. of Mol. Biol. and Genetics, Aarhus Univ., DK-8000 Aarhus C, Denmark; [4]Lab. of Mol. Neurobiology, Dept. of Neuroscience, IRCCS - Mario Negri Inst. for Pharmacol. Res., Via, La Masa, 19, 20156 Milan, Italy

Abstract: tRNA derived small RNA (tsRNA) are a recently recognised class of small noncoding RNA that play a role in protein translation and regulation of gene expression. tsRNAs are produced when ribonucleases such as angiogenin or dicer cleave tRNA under stress conditions. Based on cleavage site and size, tsRNA can be classified into tRNA-derived stress-induced RNA (tiRNA) and tRNA-derived fragments (tRF). While the complete biological functions of tsRNAs are yet to be established, several studies have implicated differential expression of tsRNAs in neurodegenerative disorders where they may represent novel diagnostic or prognostic biomarkers. This study was aimed to evaluate and compare the expression patterns of tsRNAs in mouse models of amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTD). We performed a small RNA sequencing analysis and employed a novel bioinformatics pipeline developed by our group, tsRNAsearch for the analysis of differentially expressed tsRNAs and other small non-coding RNAs. p-values less than 0.05 were considered significant for all statistical tests. Firstly, we analysed small RNA-seq data from sciatic nerve, muscle, spinal cord and serum samples obtained from $SOD1^{G93A}$ mice associated with ALS, with a slow (C57) and fast (129Sv) disease progression (n=4 in each case). Overall tRNA concentrations was similar in wild type (WT) and 129Sv samples and higher in C57 samples for all tissue types compared to WT. Strongest difference between samples was observed for 5'tiRNA-Gly-CCC in muscle samples with significantly higher expression in C57 vs WT. This effect was not seen when comparing muscle samples from 129Sv vs. WT. Secondly, we analysed small RNA-seq data from spinal cord samples from mice with a $TDP43^{A315T}$ mutation responsible for ALS and FTD. Samples were collected at 60 days pre-symptomatic and 100 days from onset of disease (n=4 in each case). The results showed that the mutant sample from both time points had an overall higher concentration of tRNA compared to the WT. Strongest difference between WT and 60 days pre-symptomatic samples was observed for 5'tiRNA-Val-CAC, with significantly lower

levels in WT samples. Interestingly, this trend was not seen when comparing WT to samples collected at 100 days from disease onset. Here, 5'tiRNA-Glu-TTC showed strongest difference between WT and samples collected 100 days after disease onset, with significantly lower levels in WT samples. Samples from hippocampi of Tau^{P301S} mutant mice are currently being analysed. We demonstrate that tsRNAs are differentially expressed in mouse models of ALS and FTD, highlighting their potential as diagnostic or prognostic biomarkers.

Disclosures: S. Baindoor: None. **H.A.Y. Gibriel:** None. **P. Donovan:** None. **M.T. Veno:** None. **J. Kjems:** None. **E. Jirstrom:** None. **A. Kenny:** None. **G. Nardo:** None. **C. Bendotti:** None. **M.C. Hogg:** None. **T. Engel:** None. **J.H.M. Prehn:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.11

Topic: C.06. Neuromuscular Diseases

Title: Molecular basis of motor neuron degeneration in SCYL1 deficiency syndrome

Authors: *A. CASSIDY[1], E. KULIYEV[2], D. THOMAS[1], R. KAEFER[1], H. CHEN[1], S. GINGRAS[3], S. PELLETIER[1];
[1]Med. and Mol. Genet., Indiana Univ. Sch. of Med., Indianapolis, IN; [2]St. Jude Children's Res. Hosp., Memphis, TN; [3]Immunol., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: SCYL1 is a multidomain protein thought to regulate several essential cellular functions including membrane protein trafficking, nucleocytoplasmic shuttling of tRNAs, and transcription via interactions with several molecular complexes. In both humans and mice, inactivating *SCYL1* mutations cause a multisystem disorder characterized by recurrent episodes of liver failure, growth retardation and progressive motor dysfunction resulting from the loss of spinal motor neurons. However, the molecular mechanisms underlying this syndrome has remained elusive. Here we report the characterization of an allelic series of *Scyl1* separation-of-function mutations in mice and show that although defective protein trafficking and faulty transcription were viewed as major disease mechanism, mice harboring mutations in *Scyl1* that prevented interaction between SCYL1 and various factors involved in protein trafficking or DNA binding showed no overt abnormalities. Importantly, however, mice expressing mutant forms of SCYL1 that disrupted the oligomerization, lipid binding, or subcellular localization of SCYL1 along the secretory pathway exhibited loss-of-function phenotypes. Together, these findings suggest that defective lipid trafficking and/or metabolism may represent a major disease mechanism in SCYL1 deficiency syndrome.

Disclosures: A. Cassidy: None. **E. Kuliyev:** None. **D. Thomas:** None. **R. Kaefer:** None. **H. Chen:** None. **S. Gingras:** None. **S. Pelletier:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.12

Topic: C.06. Neuromuscular Diseases

Title: TDP-43 shows an age-dependent susceptibility to aggregate under the absence of selective autophagy

Authors: *R. KIMURA, H. PHATNANI, A. YAMAMOTO;
Columbia Univ., New York, NY

Abstract: Protein aggregation is a major pathological hallmark spanning many age-related neurodegenerative disorders. TAR DNA binding protein 43 (TDP-43), a ubiquitously expressed RNA/DNA binding protein involved in transcription and splicing, forms abnormal, hyperphosphorylated aggregates in nearly all cases of amyotrophic lateral sclerosis (ALS) and in up to 50% of frontotemporal lobar degeneration (FTLD). Yet, it remains unclear how TDP-43 aggregation is regulated and how such processes may relate to the highly age-dependent component of disease. To address these two questions, we perturbed a highly-conserved autophagic pathway known as aggrephagy, which selectively targets pre-formed aggregates for lysosomal degradation. We report that inducible knockout mouse models of the autophagy-linked FYVE protein (ALFY), an adaptor protein required for aggrephagy, show both: 1) a drastic accumulation of insoluble, hyperphosphorylated TDP-43 in brain lysates (N=3 for both control vs. KO; t-test); and 2) a strong age dependence for aggregation, in which older mice (11- and 7-month-old compared to 3-month-old) are more susceptible to autophagic burden (N=3 per group across 5 timepoints*; two-way ANOVA). Interestingly, our preliminary screen of other common disease-associated aggregates (alpha-synuclein in Parkinson's disease, SOD1 in ALS, phospho-tau in Alzheimer's disease) do not show this phenotype. Importantly, given that our Alfy inducible KO models exist in the absence of a TDP-43 mutant or ALS/FTLD disease background, we show that wildtype TDP-43 displays an increased propensity to aggregate under natural aging. Taken together, our results suggest the importance of the selective autophagy pathway in maintaining TDP-43 integrity, in which compromised aggrephagy results in a phenotype that mirrors the proteinopathy commonly observed in ALS and FTLD patients.
*Timepoints were assessed based on the age- and duration- of KO. To understand the effects of age, we induced Alfy KO in 3-, 7-, and 11-month-old mice. To understand the effects of duration, we aged each group for a subsequent 4- or 8- months post-KO.

Disclosures: R. Kimura: None. **H. Phatnani:** None. **A. Yamamoto:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.13

Topic: C.06. Neuromuscular Diseases

Support: Lundbeckfonden LF-Experiment Grant (2020 call - project: Treating ALS disease by rescuing interneuronmotor neuron synaptic connectivity)

Title: Stabilization of V1 interneuron-motor neuron connectivity ameliorates motor phenotype in a mouse model of ALS

Authors: *S. MORA[1], R. VON HUTH-FRIIS[1], A. STUCKERT[1], G. NOES-HOLT[1], R. SELVAN[1,2], A. TOFT SØRENSEN[1], I. ALLODI[1];
[1]Dpt. Neurosci., [2]Dpt. Computer Sci., Univ. of Copenhagen, Copenhagen N, Denmark

Abstract: Amyotrophic lateral sclerosis (ALS) is a fatal disorder characterized by a progressive inability to execute movement. Although the disease is characterized by the loss of spinal motor neurons (MNs), novel evidence from our laboratory has pointed towards the implication of interneurons (INs) in the progression of the disease. Specifically, V1 spinal inhibitory INs (positive for Engrailed 1 - En1) lose their connections to MNs at early pre-symptomatic stages, which might lead to a MN hyperexcitability and cell death. The present study aimed to investigate whether forced overexpression of the presynaptic protein Extended Synagtotagmin 1 (ESYT1) in INs could stabilize MN-IN connectivity and ameliorate associated behavioral deficits. ESYT1, which is downregulated in spinal neurons early in disease, has been shown to enhance neurotransmission and stabilize synaptic growth. Here, intraspinal injections of a cre-dependent AAV8-hSyn-DIO-hESYT1-W3SL viral construct were performed in lumbar segments 1 to 3 of SOD1G93A mice crossed with En1cre mice. Four genotypes were investigated: SOD1, SOD1;En1cre, En1cre and wild-type. Cre-dependent ESYT1 overexpression was confirmed by RNAscope in situ hybridization using a probe recognizing viral sequence upon cre-recombinase. Upon treatment, we observed increased inhibitory synaptic density on lumbar MNs (measured by vesicular GABA transporter -VGAT-) in SOD1;En1cre mice when compared to untreated littermates. Moreover, the number of spared MNs was increased in the SOD1;En1cre mice. These anatomical changes also led to improvement of behavior: assessment between postnatal day 49 and 112 showed amelioration of the motor phenotype in SOD1;En1cre mice compared to SOD1 littermates, in parameters such as speed, step frequency, and stride length. Thus, our results suggest that interneurons can be a potential therapeutic target for ALS treatment and that ESYT1 might play a role in promoting MN survival.
This study was supported by the Lundbeck foundation.

Disclosures: S. Mora: None. **R. von Huth-Friis:** None. **A. Stuckert:** None. **G. Noes-Holt:** None. **R. Selvan:** None. **A. Toft Sørensen:** None. **I. Allodi:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.14

Topic: C.06. Neuromuscular Diseases

Support: NIH Grant R21AG064159
NIH Grant R01NS088627
NIH Grant R01AG066395
NIH Grant U19AG069701

Title: Trem2 interacts with tdp-43 and mediates microglial neuroprotection against tdp-43-related neurodegeneration

Authors: M. XIE[1,2], Y. LIU[1,9], S. ZHAO[1], L. ZHANG[3], D. BOSCO[1], Y.-P. PANG[4], J. ZHONG[5], U. SHETH[2,6], Y. MARTENS[7], N. ZHAO[7], C.-C. LIU[7], Y. ZHUANG[3], L. WANG[3], D. DICKSON[7], M. MATTSON[10], G. BU[7], L.-J. WU[1,7,8];
[1]Dept. of Neurol., [2]Mayo Clin. Grad. Sch. of Biomed. Sci., [3]Dept. of Mol. Pharmacol. and Exptl. Therapeut., [4]Computer-Aided Mol. Design Lab., [5]Dept. of Lab. Med. and Pathology, [6]Dept. of Neurosci., Mayo Clin., Rochester, MN; [7]Dept. of Neurosci., Mayo Clin., Jacksonville, FL; [8]Dept. of Immunol., Mayo Clin., Rochester, MN; [9]Lab. for Neurosci. in Hlth. and Disease, Guangzhou First People's Hosp. Sch. of Med., South China Univ. of Technol., Guangzhou, China; [10]Dept. of Neurosci., Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: Triggering receptor expressed on myeloid cell 2 (TREM2) is exclusively expressed on microglia in the central nervous system and is critical for microglial proliferation, migration and phagocytosis. TREM2 variants are linked to neurodegenerative disease risk. However, the function of TREM2 in neurodegeneration is still not fully understood. Here we investigated the role of microglial TREM2 in TAR-DNA binding protein 43 kDa (TDP-43)-related neurodegeneration using viral-mediated and transgenic mouse models rNLS8. We found that TREM2 deficiency impaired phagocytic clearance of pathological TDP-43 by microglia, and enhanced neuronal damage and motor impairments. In addition, TREM2 deficiency significantly increased mortality rate in both models. Mass cytometry analysis revealed that hTDP-43 induced a TREM2-dependent subpopulation of microglia with high CD11c expression and phagocytic ability. Using mass spectrometry and surface plasmon resonance analysis, we further demonstrated an interaction between TDP-43 and TREM2 *in vitro* and *in vivo* as well as in ALS patient tissues. We computationally identified regions within hTDP-43 that interact with TREM2. Our data highlights that TDP-43 is a possible ligand for microglial TREM2 and that this interaction mediates neuroprotection of microglia in TDP-43-related neurodegeneration. We conclude that microglial TREM2, as a key modulator of TDP-43 pathology, may present a therapeutic target for treating TDP-43-related neurodegenerative disorders.

Disclosures: M. Xie: None. **Y. Liu:** None. **S. Zhao:** None. **L. Zhang:** None. **D. Bosco:** None. **Y. Pang:** None. **J. Zhong:** None. **U. Sheth:** None. **Y. Martens:** None. **N. Zhao:** None. **C. Liu:** None. **Y. Zhuang:** None. **L. Wang:** None. **D. Dickson:** None. **M. Mattson:** None. **G. Bu:** None. **L. Wu:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.15

Topic: C.06. Neuromuscular Diseases

Support: CIHR
ALS Canada-Brain Canada Trainee Award-Doctoral
Weston Brain Grant

Title: Loss of C9orf72 induces neurodegeneration in a mouse model of TDP-43 proteinopathy

Authors: *L. LIN[1,2], J. ROBERTSON[1,2];
[1]Dept. of Lab. Med. and Pathobiology, [2]Tanz Ctr. for Res. in Neurodegenerative Dis., Univ. of Toronto, Toronto, ON, Canada

Abstract: In amyotrophic lateral sclerosis (ALS) and frontotemporal lobar degeneration (FTLD), transactive response DNA-binding protein 43 (TDP-43), a mainly nuclear DNA/RNA binding protein, is mislocalized to the cytoplasm of diseased neurons, where it forms abnormally phosphorylated and ubiquitinated inclusions, known as TDP-43 proteinopathy. TDP-43 proteinopathy is characteristic of patients with chromosome 9 open reading frame 72 (C9orf72) G4C2 repeat expansions, the most common genetic cause of ALS/FTLD. C9orf72 is a differentially expressed in normal and neoplastic cells domain containing protein and is reported to have roles in autophagy. Evidence has shown that there is a loss of C9orf72 transcripts and protein expression in disease affected brain regions of ALS/FTLD patients. We hypothesize that loss of C9orf72 could lead to autophagic deficits promoting TDP-43 pathology. To test this, we determined if loss of C9orf72 exacerbates TDP-43 proteinopathy in mice expressing an EGFP-tagged pathological isoform of TDP-43, TDP-25. AAV9-viral vector encoding EGFP-TDP-25 was delivered by intracerebroventricular injection into C9orf72-knock out (C9KO) and wildtype (WT) neonatal mice. Mice were aged to 8 months and extent of TDP-43 proteinopathy was assessed. Immunohistochemical labeling revealed large cytoplasmic EGFP-TDP-25 inclusions in neurons of the motor cortex, entorhinal cortex, visual cortex, primary somatosensory area, piriform area, amygdala, and olfactory bulb. EGFP-TDP-25 inclusions co-labeled with antibody to phosphorylated TDP-43, p62 and ubiquitin, recapitulating key features of TDP-43 proteinopathy. Autophagic deficits, as assessed by p62 immunoreactivity and LC3II:I ratio, were not detected in brain of control C9KO mice compared to WT. Expression of EGFP-TDP-25 however led to increased p62 immunoreactivity, and this was exacerbated by loss of C9orf72. Increased expression of ULK1 was also found in control C9KO mice when compared to WT mice; however, this increased in ULK1 levels was not present in TDP-25 expressing C9KO mice, indicating altered or impaired autophagy response. EGFP-TDP-25-C9KO mice exhibited a reduced number and size of EGFP-TDP-25 inclusions compared to EGFP-TDP-25-WT mice. Furthermore, EGFP-TDP-25-C9KO mice exhibited neuronal loss and developed mild motor phenotypes compared to EGFP-TDP-25-WT mice. These findings demonstrate that there is a combined effect of EGFP-TDP-25 expression together with C9orf72 deficiency that leads to neuronal loss and motor behavioral deficits, suggesting a two-hit model causing neurodegeneration in ALS/FTLD.

Disclosures: **L. Lin:** None. **J. Robertson:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.16

Topic: C.06. Neuromuscular Diseases

Title: A novel assessment of fine-motor function reveals early hindlimb and detectable forelimb deficits in an experimental model of ALS

Authors: *S. KHADEMULLAH, Y. DE KONINCK;
Psychiatry & Neurosci., Laval Univ., Quebec City, QC, Canada

Abstract: Amyotrophic lateral sclerosis (ALS) is a progressive neurodegenerative disorder that results in catastrophic muscle weakness and degeneration. In the preclinical setting, functional tests that can detect early changes in motor function in rodent models of ALS are critical to understanding the etiology of the disease and treatment development. However, current motor-performance tasks used to assess symptoms associated with ALS in rodent models (e.g., grip strength, gait analysis, and the rotarod) commonly measure gross motor function, and, thus, cannot detect fine-motor changes associated with forelimb and/or early hindlimb deficits. Moreover, despite the SOD1G93A mouse model being the most widely used in ALS research to date due to its ability to closely mimic the clinical phenotype of humans, it is still the center of much criticism because of its apparent lack of cortical involvement and relatively limited onset of forelimb dysfunction. However, recent evidence has emerged indicating that the cortex plays a larger role than initially acknowledged. Here, we established a string-pulling paradigm that can detect forelimb and hindlimb motor deficits in the SOD1 mouse model of ALS earlier than traditional motor performance tasks. Additionally, our findings indicate that early loss of forelimb and hindlimb function is correlated with cortical and spinal motor neuron loss, respectively. This task is not only ecological, low-cost, efficient, and non-onerous, it also requires little animal handling and reduces the stress placed on the animal. It has long been a concern in the field that the SOD1 mouse does not display forelimb motor deficits and does not give researchers a complete picture of the disease. Here, we provide evidence that the SOD1 model does in fact develop early forelimb motor deficits due to the task's ability to assess fine-motor function, reconciling this model with the various clinical presentations of ALS. Taken together, the string-pulling paradigm may provide novel insights into the pathogenesis of ALS, offer nuanced evaluation of prospective treatments, and has high translational potential to the clinic.

Disclosures: **S. Khademullah:** None. **Y. De Koninck:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.17

Topic: C.06. Neuromuscular Diseases

Support: 2R01NS081303-01A1

Title: Drosha is a genetic modifier of FUS-mediated neurodegeneration in vivo

Authors: S. KOUR[1], T. FORTUNA[2], E. ANDERSON[1], C. WARD[1], ***U. PANDEY**[1];
[1]Pediatrics, [2]Univ. of Pittsburgh Med. Ctr., Pittsburgh, PA

Abstract: Pathogenic mutations in FUS have been linked with aggressive and juvenile form of amyotrophic lateral sclerosis (ALS). FUS, a nuclear DNA/RNA binding protein, is involved in transcription, splicing, transport, and miRNA processing. However, the molecular mechanisms underlying FUS-mediated ALS are not fully known. Through an unbiased *in vivo* genetic screen in *Drosophila*, we discovered Drosha as a strong suppressor of mutant FUS toxicity. Drosha, a ribonuclease III enzyme and a major component of microprocessor complex, is involved in the regulation of microRNA biogenesis. FUS interacts with Drosha and modulates its specificity and efficacy of cleavage. However, the implication of mutant FUS on Drosha function as well as the involvement of Drosha in FUS-mediated toxicity in ALS has not been examined yet. Using *drosha* RNAi fly lines and classical mutants, we found that the depletion of drosha significantly suppresses FUS-mediated degenerative phenotypes including motor dysfunction and reduced lifespan. Accumulation of mutant FUS in cytoplasmic stress granules is the hallmark of FUS pathogenesis. Using CRISPR/Cas9 mediated *Drosha* knockout mammalian cells, we found a significant reduction in cytoplasmic FUS inclusions and FUS-positive stress granules. Drosha activity is largely dependent on phosphorylation at its serine 300/302 by GSK3beta. We found that RNAi knockdown of shaggy, a fly homolog of GSK3beta, strongly suppresses mutant FUS toxicity in flies and in patient-derived iPSCs motor neurons. In HEK293T cells the expression of phospho-dead drosha with S300A/S302A mutation alleviates mutant FUS cytoplasmic mislocalization. Furthermore, knockdown of drosha in FUS expressing flies reduces shaggy expression. Thus, our studies suggest that pathogenic mutations in FUS disrupt the regulatory interaction between Drosha and GSk3beta, thereby, altering microRNA biogenesis and turnover and their downstream targets.

Disclosures: S. Kour: None. **T. Fortuna:** None. **E. Anderson:** None. **C. Ward:** None. **U. Pandey:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.18

Topic: C.06. Neuromuscular Diseases

Support: The William Leech Charity

Title: A Macaque Model of Motor Neurone Disease

Authors: *R. H. A. JONES, F. BALEZEAU, I. SCHOFIELD, M. R. BAKER, S. N. BAKER; Newcastle Univ., Newcastle Univ., Newcastle Upon Tyne, United Kingdom

Abstract: Motor Neurone Disease (MND) is a rapidly progressive and ultimately fatal neurodegenerative disease, characterised by the loss of upper and lower motor neurons. The primary proteinopathy found in approximately 97% of all cases involves cytoplasmic mislocalisation and aggregation of the ubiquitous nuclear protein, TDP-43. Despite the identification of many implicated genes during the last few decades, our understanding of the mechanisms involved in the onset and propagation of pathology have advanced very little. Translatable advancements have likely been limited due a lack of reliable animal models which accurately recapitulate this complex disorder. No model has been created to date which replicates the progressive motor weakness; characteristic histopathologies; extended pre-symptomatic phase and subsequent rapid deterioration. Rodents have been the dominant species in MND research; however, their anatomy and genetic profile differs fundamentally to humans. Crucially, they lack the direct monosynaptic connection between the upper and lower motor neurons, unique to primates. We have harnessed a novel intersectional genetics approach to induce the overexpression of the human TDP-43 protein in a selective spinal motoneuron population in two Rhesus macaques. Focal overexpression of TDP-43 in a spinal motor pool was sufficient to induce the expression of pathological phosphorylated TDP-43 (pTDP-43) throughout the motoneurons of the cervical spine and corticomotoneuronal cells of the primary motor cortex. The detection of this histopathology in the distant giant cells of Betz in the primary motor cortex supports the idea of an axon mediated ‘prion-like’ spread, likely involving the corticospinal tract.

Disclosures: R.H.A. Jones: None. **F. Balezeau:** None. **I. Schofield:** None. **M.R. Baker:** None. **S.N. Baker:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.19

Topic: C.06. Neuromuscular Diseases

Support: NSF 1944247
Alzheimer's Association 2018-AARGD-591014
NIH U19-AG056169
McCamish Parkinson’s Disease Innovation Program at Georgia Institute of Technology

Title: Forecasting ALS combination therapeutics utilizing literature based discovery

Authors: *J. DENG, S. BI, A. LEE, C. S. MITCHELL;
Georgia Inst. of Technol., Georgia Inst. of Technol., Atlanta, GA

Abstract: Successful therapeutic discovery for Amyotrophic Lateral Sclerosis is painstakingly slow and arduously difficult. The objective of this study is to predict the most promising ALS combination pathophysiological treatment targets and to match them to corresponding repurposed drugs or substances. Quantitative data extracted from approximately 3,000 SOD1 G93A ALS mouse experimental studies is used to perform insilico temporal modeling using dynamic meta-analysis and machine learning. The temporal model optimizes the modulation of combinatorial pathophysiological factors (categorized by function: apoptosis, bioenergetics, cellular chemistry, excitability, inflammation, oxidative stress, proteomics). Based on the homeostatic instability hypothesis previously published by Mitchell and colleagues, which states homeostatic instability is the cause of ALS disease progression, mathematical re-stabilization criteria are used for treatment selection. The five most stabilizing three-way pathophysiological factor treatments are used to perform literature-based discovery (LBD) to match their desired modulatory function to real-world repurposed drugs or substances. LBD is performed using the recently published SemNet 2.0 biomedical knowledge graph and relationship ranking software, which contains text relationships extracted from 30+ million PubMed articles, followed by new or unpublished link prediction. Eleven trials comprised of 9 nodes each (3 nodes per factor x 3 factors per combination treatment) are run for each three-way factor treatment. The natural language processing link prediction model predicts new or unpublished text relationships in the subgraph to determine which drugs or substances are most likely to have the desired modulatory effect. The link prediction results include an average hits@10 for relation prediction across all factor treatment ensemble models of 0.973. Collectively, this study provides a ranked list of predicted key stabilizing pathophysiological modulating etiologies and a corresponding list of real-world drugs or substances to be tested as combination therapies in future ALS mouse model experiments.

Disclosures: **J. deng:** None. **S. Bi:** None. **A. Lee:** None. **C.S. Mitchell:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.20

Topic: C.06. Neuromuscular Diseases

Title: Electrophysiological characterisation of motoneuron properties in FUSdelta14 model of Amyotrophic Lateral Sclerosis

Authors: *M. G. OZYURT[1], F. NASCIMENTO[1], R. BROWNSTONE[1], M. BEATO[2];
[1]Queen Square Inst. of Neurol., [2]Neuroscience, Physiology, Pharmacol., Univ. Col. London, London, United Kingdom

Abstract: A proportion of familial cases of Amyotrophic Lateral Sclerosis (ALS) are caused by mutations in the Fused in Sarcoma enzyme (FUS). FUS mutations can give rise to an aggressive form of the disease, with early onset and rapid time course. In this study we used a homozygous transgenic mouse, that expresses a humanized truncated FUS protein (FUSdelta14) and compared the cellular and synaptic properties of motoneurons at an early (P1-P5) and later (P15-P25) time points in the initial stages of disease progression. Homozygous FUSdelta14 mice show early signs of motor unit loss, detected in juvenile animals (P25-30), measured through *in vivo* electromyography (EMG). These signs became more pronounced at 2 months of age in EMG along with a decline in grip strength force. *In vitro* experiments showed that, in homozygous FUSdelta14 mutants, motoneurons had a larger capacitance and an increased proportion of putative fast motoneurons. Repetitive firing characteristics were not affected, but during development we observed a marked decrease in spike size, not accompanied by a change in the threshold for firing, nor by alterations in the I_h or in the persistent inward currents. Recurrent circuits were also altered, with an increase in the strength of recurrent inhibition in juveniles. The response to afferent inputs was affected in P15-25 animals, with both mono-synaptic Ia excitation and di-synaptic Ia/Ib inhibition reduced in mutant mice. Our data show that the homozygous FUSdelta14 mouse model of ALS displays early abnormalities in both cellular properties of motoneurons and in local spinal circuits.

Disclosures: M.G. Ozyurt: None. **F. Nascimento:** None. **R. Brownstone:** None. **M. Beato:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.21

Topic: C.06. Neuromuscular Diseases

Title: Investigating the role of the galactosylceramide and sulfatide in the pathogenesis of amyotrophic lateral sclerosis

Authors: *E. JENSEN[1], L. GUO[1], T. TRELEAVEN[1], R. TAHIR[1], J. WEISSER[1], M. GONCALVES[1], T. TAKSIR[1], L. AN[1], B. RICHARDS[1], A. R. BIALAS[1], B. ZHANG[1], T.-H. NGUYEN[1], P. SARDI[1], J. C. DODGE[2];
[2]Rare and Neurolog. Dis., [1]Sanofi, Cambridge, MA

Abstract: Amyotrophic lateral sclerosis (ALS) is a progressive paralytic neuromuscular disorder characterized by the degeneration of motor neurons (MNs) in the brain and spinal cord. Mutations in TARDBP, the gene that encodes TAR DNA-binding protein 43 kDa (TDP-43), are

a known cause of ALS. Furthermore, in 97% of all familial and sporadic ALS patients, TDP-43 is depleted from the nucleus and mislocalized to cytoplasmic aggregates in MNs and glia. Previously, we have shown that glycosphingolipids are misregulated in the SOD1 mouse model and in ALS patient spinal cord tissue. Importantly, altering various glycosphingolipid levels through central nervous system (CNS) pharmacologic inhibition or lipid infusion can modify the disease progression in the SOD1 mouse model (Dodge, et al. PNAS, 2015). Here, we report on our efforts to determine if the glycosphingolipids, galactosylceramide (GalCer) and sulfatide (ST), contribute to disease pathogenesis in ALS. Using liquid chromatography mass spectrometry (LC-MS) and matrix assisted laser-desorption ionization-mass spectrometry (MALDI-MS) imaging, we found that GalCer and ST were significantly elevated in the cytoplasm and grey matter in the spinal cords of rNLS8 mice, an ALS mouse model with TDP-43-dependent neurodegeneration. Follow up transcriptomic and proteomic studies in rNLS8 mouse spinal cord revealed significant changes in the enzymatic regulators of GalCer/ST synthesis corroborating our lipidomic findings. Excitingly, MALDI-MS imaging analysis of human samples also displayed significant GalCer and ST accumulation in the ALS spinal cord grey matter. Analysis of ALS patient spinal cord transcriptomic data showed perturbations in the GalCer/ST synthetic machinery similar to that observed in the rNLS8 mouse model. Moreover, LC-MS analysis showed that GalCer and ST were significantly increased in the cerebrospinal fluid (CSF) of sporadic ALS patients. The relatively slow CNS turnover of GalCer/ST makes it difficult to assess the therapeutic benefit of lowering these lipids in the rNLS8 mice, a rapidly progressing model of ALS. Nevertheless, we did find that pharmacological inhibition of UGT8, a regulator of glycosphingolipid synthesis, reduced newly synthesized GalCer/ST levels in the CNS of rNLS8 mice. In conclusion, we show that GalCer/ST pathway is perturbed in the spinal cord in a TDP-43 mouse model of ALS and ALS patient-derived samples. Additional experiments are underway to determine the cell autonomous role of UGT8 and whether UGT8 inhibition can modulate inflammation and neuroprotection in cellular models of ALS.

Disclosures: **E. Jensen:** A. Employment/Salary (full or part-time):; Sanofi. **L. Guo:** A. Employment/Salary (full or part-time):; Sanofi. **T. Treleaven:** A. Employment/Salary (full or part-time):; Sanofi. **R. Tahir:** A. Employment/Salary (full or part-time):; Sanofi. **J. Weisser:** A. Employment/Salary (full or part-time):; Sanofi. **M. Goncalves:** A. Employment/Salary (full or part-time):; Sanofi. **T. Taksir:** A. Employment/Salary (full or part-time):; Sanofi. **L. An:** A. Employment/Salary (full or part-time):; Sanofi. **B. Richards:** A. Employment/Salary (full or part-time):; Sanofi. **A.R. Bialas:** A. Employment/Salary (full or part-time):; Sanofi. **B. Zhang:** A. Employment/Salary (full or part-time):; Sanofi. **T. Nguyen:** A. Employment/Salary (full or part-time):; Sanofi. **P. Sardi:** A. Employment/Salary (full or part-time):; Sanofi. **J.C. Dodge:** A. Employment/Salary (full or part-time):; Sanofi.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.22

Topic: C.06. Neuromuscular Diseases

Support: NIH Grant

Title: Development of Novel Models to Study Oligodendroglia Dysfunction in C9orf72 ALS

Authors: *O. SPEAD[1], J. ROTHSTEIN[2];
[1]Neurology/Brain Sci. Inst., [2]Neurol., Johns Hopkins Univ., Baltimore, MD

Abstract: Amyotrophic Lateral Sclerosis (ALS) is a fatal disease characterized by progressive motor neuron degeneration. The majority of ALS cases are sporadic, though 10-15% are caused by known inherited mutations in genes such as *C9ORF72* and *SOD1*. While much is known about how these mutations affect motor neurons, their effect on glial cells such as oligodendrocytes (OLs) or oligodendrocyte progenitor cells (OPCs) is largely unknown. Recently, we have observed in a mutant SOD1 mouse model of ALS that OPCs aberrantly proliferate while mature OLs degenerate and that removing mutant *SOD1* from oligodendrocytes prolongs the lifespan of these mice, highlighting the importance of oligodendrocytes in ALS disease pathology. To date, very few studies have investigated the role of the *C9orf72* hexanucleotide repeat expansion (HRE), the most common genetic cause of ALS, in oligodendrocytes. To investigate the contribution of OPCs and OLs to CNS health in this context, we have developed three novel model systems: 1) an OL-like immortalized cell line (Oli-Neu) expressing the C9 HRE; 2) OL differentiations from induced pluripotent stem cells (iPSCs) derived from patients diagnosed with C9-ALS; 3) an oligodendrocyte-specific AAV expressing the C9 HRE for in vivo analysis of oligodendrocyte lineage cells. All three of these models will be used to better understand the contribution of OPCs and OLs to ALS disease pathology and to further investigate the cellular mechanisms contributing to neurodegeneration.

Disclosures: O. Spead: None. **J. Rothstein:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.23

Topic: C.06. Neuromuscular Diseases

Support: Ulla-Carin Lindquist´s Foundation for ALS Research
The Swedish Research Council
Västerbottens läns landsting

Title: Investigating ALS pathogenesis caused by distinct SOD1 strains in transgenic mouse models

Authors: *I. SIGFRIDSSON, M. MARKLUND, T. BRÄNNSTRÖM;
Med. Biosci., Umeå, Sweden

Abstract: Aggregates of misfolded superoxide dismutase-1 (SOD1) are hallmarks of amyotrophic lateral sclerosis (ALS) caused by mutations in the *SOD1* gene. In our laboratory, two structurally distinct strains of human (h) SOD1 aggregates (denoted A and B) have previously been identified in transgenic (Tg) *hSOD1* mice. Intra-spinal inoculations of these strains have shown their capability to induce strain-specific aggregation through templating and cause premature fatal motor neuron disease, supporting that ALS caused by SOD1 mutations is a prion-like disease. Additionally, strain A and B affect the progression of the disease differently, suggesting a difference in pathogenesis provoked by the distinct strains. Whether this difference could be explained by unique subcellular localizations in the central nervous system (CNS) has previously not been examined. In this study, we optimized protocols to histologically visualize strain A and B aggregates and investigate their subcellular localization in ALS Tg mouse models. For visualization, antibodies targeting strain specific sequences was used to label hSOD1 aggregates in spinal cords of $hSOD1^{G85R}$ and $hSOD1^{D90A}$ Tg mice. The histological strain characterization was determined using immunofluorescence and Proximity Detection. Subsequently, the spinal cords were examined using immunogold-labeling electron microscopy to study the subcellular localization of hSOD1 aggregates. Our results show successfully developed protocols for histological visualization of strain A and B and for studying localization of hSOD1 aggregate species in the CNS of ALS Tg mouse models. These findings form the basis to further investigate the difference in disease progression provoked by strain A and B hSOD1 aggregates. This knowledge will be of great importance to better understand the pathogenesis behind ALS.

Disclosures: I. Sigfridsson: None. **M. Marklund:** None. **T. Brännström:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.24

Topic: C.06. Neuromuscular Diseases

Support: Alzheimer's Association New Investigator Research Grant: NIRG-1-241456
National Institute on Aging: 1K01AG042500
Delaware IDeA Network of Biomedical Research Excellence (INBRE) Pilot Award: NIH-NIGMS: 5P20GM103446
NIH-NIGMS Centers of Biomedical Research Excellence (COBRE): 5P20GM103653
Delaware Economic Development Office from the State of Delaware

Title: Rapid progression, pathology, and reduced lifespan in a triple mutant TDP-43 mouse model

Authors: *M. DOPLER, K. E. COX, S. AREZOUMANDAN, M. KOTEY, W. SMITH, T. PETERSEN, C. PREDDIE, M. A. GITCHO;
Biol. Sci., Delaware State Univ., Dover, DE

Abstract: Rapid progression, pathology, and reduced lifespan in a triple mutant TDP-43 mouse model

Matthew B. Dopler[1], Keyshawn E. Cox[1], Charmise Preddie[1], Tyler Petersen[1], Miranda Kotey[1], Whitnei Smith[1], Sanaz Arezoumandan[1], and Michael A. Gitcho[1,2]
[1]Delaware State University, Dover, DE, USA, [2]Delaware Center for Neuroscience Research, Dover, DE, USA

Amyotrophic Lateral Sclerosis (ALS) is a progressive neurodegenerative disease characterized by the death of upper and lower motor neurons leading to gradual weakness of skeletal muscles. The average survival time after diagnosis is 3-5 years with most common cause of death being respiratory failure. In ALS, TAR DNA binding protein 43 (TDP-43) is one of the major pathological protein. Pathological TDP-43 forms phosphorylated insoluble aggregates and fragments primarily in the cytosol leading to neuronal loss, muscle degeneration, and eventually death. Though primarily sporadic, 5-10% of ALS cases are linked to familial mutations making these mutations ideal targets for developing mammalian models. We have developed a mouse model expressing three familial ALS mutations (A315T, M337V, S379P) in TDP-43 (3XTDP-43). The tetracycline inducible cell-specific expression system was used to selectively drive 3XTDP-43 with the human neurofilament heavy polypeptide promoter driving tetracycline-controlled transactivator protein (NEFH-tTA) in the brain and spinal cord.Motor deficits emerge at approximately 35 days with a rapid progression leading to paralysis at death. The 3XTDP-43 expressing mice show reduced lifespan typically between 44-66 days. Biochemical and pathological analysis reveals aggregation of pathological TDP-43. In addition, protein levels of glial fibrillary acidic protein (GFAP) were significantly increased in the brain and spinal cord. We hope that this model can provide a better understanding of motor neuron disease and other TDP-43 proteinopathies.

Disclosures: M. Dopler: None. **K.E. Cox:** None. **S. Arezoumandan:** None. **M. Kotey:** None. **W. Smith:** None. **T. Petersen:** None. **C. Preddie:** None. **M.A. Gitcho:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.25

Topic: C.06. Neuromuscular Diseases

Support: ALS Canada
Brain Canada
CIHR
FRQS

Title: Zebrafish TDP-43 knock-in CRISPR mutants develop a robust ALS-like phenotype

Authors: *Z. HARJI, E. C. RODRIGUEZ, G. A. B. ARMSTRONG;
Montreal Neurolog. Institute, McGill Univ., Montreal Neurolog. Institute, McGill Univ., Montreal, QC, Canada

Abstract: Amyotrophic lateral sclerosis (ALS) is an adult-onset neurodegenerative disease that is characterized by loss of upper and lower motor neurons which eventually leads to paralysis and death within 2-5 years of diagnosis. A pathological hallmark of the disease is the cytoplasmic mislocalization of the nuclear RNA binding protein TAR-DNA Binding Protein of 43 kDa (TDP-43) in motor neurons in ~97% of all ALS cases. Mutations in *TARDBP*, encoding TDP-43, are associated with a small percentage of ALS cases and among these are the mutations encoding the A382T (the most commonly found variant) and G348C TDP-43 variants. By using the CRISPR/Cas9 mutagenic system, we have developed knock-in zebrafish lines carrying point mutations that encode orthologous A382T and G348C variants in Tdp-43. These mutant zebrafish were found to have a reduced lifespan compared to their wild-type siblings. As adults, they develop a degenerative motor phenotype in both a free swim assay and a swim tunnel which was coincident with the loss of large spinal motor neurons and muscle degeneration. Furthermore, while Tdp-43 expression levels in the brain are similar between wild-type and mutant zebrafish, a compensatory splice variant (*tardbpl_tv1*) that arises from a *tardbp* paralogue present in the zebrafish genome (*tardbpl*) was significantly increased, particularly in *tardbp*$^{G347C/G347C}$ brains. Ablation of *tardbpl* expression altogether led to a ~2-fold increase in Tdp-43, which we believe results in a more severe phenotype in animals that harbour *tardbp* mutations. Preliminary data also suggests that *tardbp*$^{A379T/A379T}$, but not zebrafish *tardbp*$^{G347C/G347C}$ display greater quantities of insoluble Tdp-43 that is independent of its subcellular localization. Lastly, at a pre-symptomatic stage, we performed RNA-sequencing of spinal cord tissue from homozygous mutants and found an upregulation of transcripts involved in the immune response and inflammation in both mutants. In homozygous *tardbp*$^{G347C/G347C}$ animals, we also found downregulation of transcripts involved in synaptic transmission and function, some of which have been previously identified in sporadic ALS patients. Future directions include investigation of structural and functional neuromuscular junction defects, as well as testing the use of new gene-editing technologies to repair these mutations.

Disclosures: Z. Harji: None. **E.C. Rodriguez:** None. **G.A.B. Armstrong:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.26

Topic: C.06. Neuromuscular Diseases

Support: CIHR Grant MOP110950
CIHR Grant PJT-173547
NSERC Grant RGPIN 04880

Title: Differential changes in spinal V3 interneurons pre- and post-symptom onset in a $SOD1^{G93A}$ mouse model of amyotrophic lateral sclerosis

Authors: *C. MACKAY, J. BOROWSKA-FIELDING, Y. ZHANG;
Med. Neurosci., Dalhousie Univ., Halifax, NS, Canada

Abstract: Amyotrophic lateral sclerosis (ALS) is a progressive and fatal neurodegenerative disease characterized by progressive motor neuron (MN) death leading to muscle atrophy and paralysis, with 80% of patients dying within 5 years of diagnosis. Recent studies have shown that spinal interneurons (INs) may play critical roles in disease onset and progression in different ALS animal and human IPS cell models. V3 INs are an important group of excitatory neurons, involved in establishing the robustness of the locomotor rhythm by promoting left-right synchronization in the spinal cord. In this study we aimed to investigate the changes of V3 spinal interneurons in the lumbar spinal cord at different stages of ALS disease using a SOD1G93A mouse model. We generated a $Sim1^{Cre/+};Rosa26^{floxstopTdTom}::SOD1^{G93A}$ mouse line to specifically visualize V3 INs by their expression of tdTomoto fluorescent protein. We then studied the anatomical distribution, the electrophysiological properties, and their morphologies at the earliest age of locomotor maturity (P21), as well as five distinct stages of pre- and post-symptomatic disease progression (~P85 to end-stage) (n = 5 for each). Our results showed a 30.3% ($p < 0.0001$) reduction in total V3 INs in the lumbar spinal cord of SOD1G93A mice at P21. This trend persisted into symptomatic disease stages, with a potentially further substantial decrease by the end stage. Even though we found that V3 INs in SOD1G93A mice showed similar intrinsic electrophysiological properties to their wild-type counterparts, we did observe a slight but significant increase in spike frequency of the SOD1 mutant V3 INs at P21, indicating an increased excitability. This change in excitability, however, later converted to a decrease by the time of symptom onset and persisted until later stages of the disease. Furthermore, investigation into V3 cell morphology showed a significant decrease in the complexity of neurite arborization at later stages of the disease in the SOD1G93A mutants compared to their wild-type littermates. Our results indicate clear early perturbations in V3 IN properties that change with differing trajectories as the disease progresses. These results suggest that V3 INs may play an important role in disease onset and progression of ALS, but more research will be required to fully elucidate their exact potential contributions to pathology of the disease.

Disclosures: C. MacKay: None. **J. Borowska-Fielding:** None. **Y. Zhang:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.27

Topic: C.06. Neuromuscular Diseases

Support: NIH 5R35NS097224

Title: Mitochondrial dysfunction in a Drosophila model for ALS

Authors: *M. SWARTZLANDER, J. M. SABANDAL, A. A. BUTLER, R. L. DAVIS;
UF Scripps Biomed. Res., UF Scripps Biomed. Res., Jupiter, FL

Abstract: Amyotrophic lateral sclerosis (ALS) is a neurodegenerative disorder affecting motor neurons and has been linked to mutations in TAR DNA-binding protein 43 (TDP43). ALS patients exhibit progressive muscle weakness resulting in an eventual loss in voluntary motor function, as well as other cognitive and behavioral symptoms. As treatment options are limited, patients often die within a few years after diagnosis. TDP43 has been associated with mitochondrial impairment. Additionally, a growing body of literature in GWAS studies, animal models, and cell culture implicate mitochondrial dysfunction as a central player in the onset of ALS and other neurodegenerative diseases. However, studies of ALS using mammalian models can be prohibitively costly and time consuming, often spanning hundreds of days. Here, we describe behavioral and cellular phenotypes observed in a series of assays using *Drosophila melanogaster* with induced expression of mutant TDP43(G298S) in motor neurons during adulthood. We observed decreased lifespan, motor impairment, and changes in mitochondrial morphology that are like other ALS models. The phenotypes observed are progressive as in ALS, declining with age. Induced expression of TDP43(G298s) in adult *Drosophila* provides a useful tool for *in vivo* studies of TDP43 pathology, reducing the time and cost required for similar experiments with a mammalian model by an order of magnitude.

Disclosures: M. Swartzlander: None. **J.M. Sabandal:** None. **A.A. Butler:** None. **R.L. Davis:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.28

Topic: C.06. Neuromuscular Diseases

Support: Sino Danish Center for Education and Research
Center for Stochastic Geometry and Advanced Bioimaging

Title: Structural and functional changes in a genetic model of amyotrophic lateral sclerosis

Authors: *S. HASSELHOLT[1], H. J. T. STEPHENSEN[3], D. JANSSEN[4], A. HASSELHOLT[1], A. DUBOIS[5], U. HAHN[2], Y. WANG[6], J. SPORRING[3], Z. XU[6], J. R. NYENGAARD[1];
[1]Dept. Of Clin. Med., [2]Dept. of Mathematics, Aarhus Univ., Aarhus, Denmark; [3]Dept. of Computer Sci., Univ. of Copenhagen, Copenhagen, Denmark; [4]Dept. of Biomed. Engin.,

Eindhoven Univ. of Technol., Eindhoven, Netherlands; [5]Bioelectron Microscopy Core Facility, École Polytechnique Fédérale de Lausanne, Lausanne, Switzerland; [6]Inst. of Genet. and Developmental Biol., Chinese Acad. of Sci., Beijing, China

Abstract: The progressive, neurodegenerative disorder amyotrophic lateral sclerosis (ALS) usually has a fatal outcome within two to four years after appearance of the initial symptoms. For the majority of patients, the aetiology is unknown underlining a continued need for knowledge about disease pathogenesis. Based on a screening of Chinese ALS patients, we have developed a new genetic mouse model of ALS. This mouse lacks a gene involved in regulation of myelination, and white matter changes are a consistent, and early finding in neuroimaging studies of ALS patients. Mice were examined *in vivo* using a behavioural test battery focusing on motor abilities (n= 5-8/group, separate groups for M and F, 9 and 14 months). Evoked activity of primary- and secondary motor cortices was assessed with microelectrode array (n=3/group and a total of 6-8 slabs/group, F only, 10.6 months), and an extensive structural- (n=8-12/group, M and F, 10.6 months) and ultrastructural (n=4-5/group, M only, 10.6 months) evaluation of motor cortex and white matter was performed using stereological sampling and - analysis principles, and machine-learning on 3-dimensional image stacks. Muscles of the lower hind limb were evaluated after a double immunohistochemical stain for fast- and slow muscle fibres (n=3-7, F only, 10.6 months). At 9 months of age, KO mice presented fine motoric deficits in the beam walk task. They had longer latencies to traverse the beam and more two-feet slips from the beam than WT mice. Motor learning assessed with rotarod was not affected at this time point. In motor cortex layer V, KO mice had decreased mean volume of large motor neurons and the evoked electrical activity was impaired. Node of Ranvier length in motor cortex measured in 3D was shortened in KO mice but surprisingly, no white matter changes were identified. Cross-sectional area of muscle fibres was affected more in slow- than in fast muscle fibres, however, in *m. soleus* (the only of the four examined muscles with a sufficient amount of slow fibres for adequate fibre type comparison) the effect of genotype was borderline. At 14 months of age, motor learning was affected in KO mice indicating a progressive phenotype. In summary, our mouse model with loss of gene X* presents mild changes in motor cortex and muscles compatible with alterations seen in ALS patients. This gene may therefore be a contributor to ALS pathogenesis but it cannot explain the clinical presentation on its own.* *The identity of the gene is known by all authors*

Disclosures: S. Hasselholt: None. **H.J.T. Stephensen:** None. **D. Janssen:** None. **A. Hasselholt:** None. **A. Dubois:** None. **U. Hahn:** None. **Y. Wang:** None. **J. Sporring:** None. **Z. Xu:** None. **J.R. Nyengaard:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.29

Topic: C.06. Neuromuscular Diseases

Support: NIH grant R01NS064131
VA Career Development Award Level 2 BX004341-01A1

Title: Ubiquilin modifies TDP-43 mediated neurodegeneration in C. elegans

Authors: *A. SAXTON[1], R. KOW[1,2], B. HENDERSON[1], B. KRAEMER[1,2,3,4];
[1]Geriatrics Res. and Educ. and Clin. Ctr., Veterans Affairs Puget Sound Hlth. Care Syst., Seattle, WA; [2]Div. of Gerontology and Geriatric Medicine, Dept. of Med., [3]Dept. of Psychiatry and Behavioral Sci., [4]Dept. of Lab. Med. and Pathology, Univ. of Washington, Seattle, WA

Abstract: Amyotrophic lateral sclerosis (ALS) is a debilitating, fatal neurodegenerative disease that causes rapid muscle wasting. It shares a spectrum of symptoms and pathology with frontotemporal lobar degeneration (FTLD). Mutations in the human gene encoding TDP-43 cause ALS, while mutations in human UBIQUILIN-2 (UBQLN2) cause ALS/FTLD with TDP-43 positive inclusions. UBQLN2 encodes a ubiquitin-like adaptor protein involved in the ubiquitin-proteasome protein degradation pathway. In disease, UBQLN2 and TDP-43 aggregate in skein-like inclusions with other ALS and FTLD associated proteins. To understand the molecular drivers of neurodegeneration, we have modelled ALS in *Caenorhabditis elegans,* a simple animal model advantageous for its short lifespan, ease of cultivation, thoroughly characterized nervous system, and well-established behavioral assays. We have previously generated and published a variety of transgenic *C. elegans* models of ALS-related proteinopathies by overexpressing human TDP-43 and/or UBQLN2 with or without disease causing mutations. While loss of the *C. elegans* TDP-43 homolog TDP-1 does not modify neurodegenerative phenotypes of UBQLN2 overexpression, we have recently identified a mutation in the *C. elegans* UBQLN2 homolog, *ubql-1*, which modifies TDP-43 related neurodegenerative phenotypes. We will present the ongoing characterization of the complex interaction between TDP-43 and UBQLN2 with discussion of the molecular mechanisms contributing to neurodegeneration.

Disclosures: A. Saxton: None. **R. Kow:** None. **B. Henderson:** None. **B. Kraemer:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.30

Topic: C.06. Neuromuscular Diseases

Support: Independent Research Fund Denmark

Title: Tdp-43 pathology drives a reversible hyperexcitability of spinal motoneurones in a sporadic model of amyotrophic lateral sclerosis.

Authors: Z. ZHAO[1], S. DJUKIC[1], L. M. H. JØRGENSEN[2], A. N. BAK[1], D. B. JENSEN[3], ***C. F. MEEHAN**[1];

[1]Univ. of Copenhagen, Copenhagen, Denmark; [2]Univ. of Copenhagen, Denmark, Denmark; [3]Dept. of Neurosci., Univ. of Copenhagen, Copenhagen, Denmark

Abstract: TMS, reflex and nerve excitability studies all indicate an increased motoneurone excitability in patients with Amyotrophic Lateral Sclerosis (ALS), which we have confirmed with intracellular recording in SOD1 mice models of the disease. However, SOD1 mutations account for only a minority (~2%) of ALS patients. What drives the hyperexcitability in the vast majority of sporadic cases is therefore unknown. The most common key pathological feature found in ~95% of all ALS patients are cytoplasmic aggregates of the protein TDP-43. To test whether this is sufficient to drive motoneurone hyperexcitability, we used the TDP-43(ΔNLS) mouse model which successfully recapitulates this pathology expressing a mutant human TARDBP, TAR DNA binding protein with a defective nuclear localization signal, controlled by the tetO, tetracycline operator promoter. After 4 weeks of transgene induction, adult mice (both male and female) show a severe ALS phenotype documented by a series of different behavioural tests. Re-suppression of the transgene at this point results in a subsequent recovery of motor function by 6-8 weeks. In vivo intracellular recordings after 4 weeks induction revealed significant decreases in rheobase currents for repetitive action potential firing and increases in the gain of spinal motoneurones, both of which returned to normal values after re-suppression of the transgene. Anatomical experiments revealed a significant reduction in soma size in induced mice, consistent with reductions in input resistance. Also consistent with the increased excitability, axon initial segments on both gastrocnemius and soleus motoneurones were significantly longer and thinner in the 4-week induced mice. We therefore conclude that TDP-43 pathology is sufficient to drive a severe but reversible hyper-excitability of spinal motoneurones, explaining the excitability changes observed in sporadic patients.

Disclosures: Z. Zhao: None. **S. Djukic:** None. **L.M.H. Jørgensen:** None. **A.N. Bak:** None. **D.B. Jensen:** None. **C.F. Meehan:** None.

Poster

537. ALS Mechanisms and Models I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 537.31

Topic: C.06. Neuromuscular Diseases

Support: Funding from Department of Science and Technology, Government of India (DST/CSRI/2017/258)
Senior Research fellowship to B. Dey from Department of Biotechnology (DBT), Government of India

Title: Chronic stress acts as a determinant of the onset of amyotrophic lateral sclerosis in an optineurin knock-out mouse model

Authors: B. DEY[1,2], S. KOTTAPALLI[1], D. ROY[1], N. K. SINGH[1], S. C. MOHARIR[1,3], D. T. SOWPATI[1,2], S. CHAKRAVARTY[2,4], G. SWARUP[1,2], A. B. PATEL[1,2], *A. KUMAR[1,2]; [1]CSIR-Centre For Cell. and Mol. Biol. (CCMB), Hyderabad, India; [2]Acad. of Scientific & Innovative Res. (AcSIR), Ghaziabad, India; [3]Tata Inst. for Genet. & Society (TIGS), Bengaluru, India; [4]CSIR-Indian Inst. of Chem. Technol. (IICT), Hyderabad, India

Abstract: There is a paucity of *in vivo* studies on how the onset of motor neurodegeneration in amyotrophic lateral sclerosis (ALS) is modified by the influence of environmental factors on the endophenotype produced in a genetically vulnerable background. We investigated the role of chronic stress on the progression of motor neuropathy in a mouse model of whole-body knock-out (KO) of the ALS-associated multifunctional adaptor protein optineurin (OPTN). Notably, we did not observe a clear ALS phenotype in non-stressed *Optn* KO mice when compared to wild-type (WT) mice (n=6 in each group) till 12 months of age. Hence, 9-month-old mice were exposed to the chronic variable mild stress (CVMS) paradigm for 3 weeks. At 30 days post CVMS, an impairment in neuromuscular strength and motor coordination (evaluated by grip test and rotarod test respectively) was observed in *Optn* KO stressed mice but not in age-matched KO control, WT stressed and WT control mice (n=6 in each group) of both male and female gender. Disease progression was monitored till 18 months of age. Interestingly, we noted a delay in the onset of gross motor deficits (denoted by neurological scoring) in female KO stressed mice compared to male KO stressed mice (180 days vs 90 days post CVMS). To study the neurometabolic profile, a tracer-based approach was used via infusion of [1,6-$^{13}C_2$]glucose or [2-^{13}C]acetate followed by ^{1}H-[^{13}C] nuclear magnetic resonance spectroscopy of tissue extract. This revealed neuronal glucose hypometabolism and astroglial hypermetabolism in male KO stressed mice compared to KO controls. Transcript quantification in motor cortex and lumbar spinal cord of male mice at 18 months uncovered the altered expression of markers involved in neuroinflammation, dysregulated proteostasis, reactive gliosis and chronic stress-associated epigenetic regulators in KO stressed group compared to KO controls. Additionally, upon CVMS exposure at an early age (5 months), we found that disease onset in male KO stressed mice (n=7 in each group) was delayed considerably (4 months post CVMS). Here, bulk RNA-sequencing used for studying the disease-associated transcriptome indicated an enrichment of dysregulated pathways in ALS signaling, phagosome processes and synaptogenesis signaling in lumbar spinal cord, but not in motor cortex of KO stressed mice when compared with KO controls (n=3 in each group). In summary, our results suggest that chronic stress in *Optn* KO mice determines the onset of ALS, which possibly progresses in a retrograde manner. Future integrative studies on cell type-specific transcriptomic and epigenomic data will reveal how gene-environment interaction dictates clinical heterogeneity in ALS.

Disclosures: B. Dey: None. **S. Kottapalli:** None. **D. Roy:** None. **N.K. Singh:** None. **S.C. Moharir:** None. **D.T. Sowpati:** None. **S. Chakravarty:** None. **G. Swarup:** None. **A.B. Patel:** None. **A. Kumar:** None.

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.01

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

Support: NIH Grant F31NS125966
NIH Grant R01NS093992
NIH Grant R01NS113516
NIH Grant R01NS089770
Semmes Foundation

Title: Modeling the Neurological Effects of COVID-19 Exposure Using Human Cortical Organoids

Authors: *C. L. MCMAHON[1], H. STAPLES[3], M. GAZI[4], R. CARRION[3], J. HSIEH[2];
[1]Univ. of Texas At San Antonio Dept. of Biology/Neurobiology, [2]Univ. of Texas at San Antonio, San Antonio, TX; [3]Texas Biomed. Res. Inst., San Antonio, TX; [4]Texas Biomed. Res. Inst., san anotnio, TX

Abstract: The prevalence of neuropsychiatric disorders is increasing, and a major cause of these disorders is aberrant neurogenesis. There is growing evidence implicating prenatal exposures as a driving factor, and an increasingly common exposure is that of viral infections, due in part to the novel coronavirus disease 2019 (COVID-19) pandemic. Along with SARS-CoV-2 RNA detection in brain biopsies from fatal cases, up to 80% of hospitalized and 30% of overall COVID-19 patients have manifested neurological complications, indicating a possible neurotropism of this virus. Importantly, these rates had increased during the Delta variant wave. Because the implications of SARS-CoV-2 and resulting inflammation on the brain are still poorly understood, it is imperative to gain insight into the mechanism(s) responsible for these complications. **We hypothesize that SARS-CoV-2 infection and inflammation trigger neurodevelopmental changes leading to a disruption of neural structure and circuit function in the cortex.** Due to the inaccessibility of brain tissue from COVID-19 patients, we began to address these questions by using a 3D *in vitro* model, human stem cell-derived cortical organoids. To elucidate the cellular effects of SARS-CoV-2 infection on the development of human brain tissue, we first examined the tropism of SARS-CoV-2 in cortical organoids by identifying susceptible cell types to infection. We also evaluated viral replication potential and the consequential effects on infected cells. We next determined if the pro-inflammatory cytokine TNFα influenced infection in organoids by evaluating alterations in viral detection levels, viral replication, cell susceptibility, and cell death. Our results showed that SARS-CoV-2 WT and Delta variants infected glial cells and choroid plexus cells in cortical organoids within 6 hours, but did not replicate or cause cell death (McMahon et al., 2021). TNFα exposure did not alter this observed pathology. This study aims to reveal novel insight into the cellular etiology of potential neuropsychiatric disorder development from COVID-19 exposure, potentially contributing to future therapies and interventions.

Disclosures: C.L. McMahon: None. **H. Staples:** None. **M. Gazi:** None. **R. Carrion:** None. **J. Hsieh:** None.

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.02

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

Support: NIH R01NS113516
Kleberg Foundation
Semmes Foundation

Title: Familial Alzheimer's disease associated PSEN1 mutations affect neurodevelopment through increased Notch signaling

Authors: *E. HURLEY[1], P. MOZOLEWSKI[2], R. DOBROWOLSKI[3], J. HSIEH[4];
[1]Neurosci. Developmental and Regenerative Biol., Univ. of Texas San Antonio, San Antonio, TX; [2]Glenn Biggs Inst. for Alzheimer's and Neurodegenerative Diseases; Univ. of Texas Hlth., San Antonio, TX; [3]Dept. of Biol. Sci., Rutgers Univ., Newark, NJ; [4]Neurosci. Developmental and Regenerative Biol., The Univ. of Texas At San Antonio, San Antonio, TX

Abstract: Alzheimer's disease (AD) is the most common neurodegenerative disorder, but its rootcause may lie in neurodevelopment. Familial AD (fAD) is most commonly caused byautosomal dominant genetically inherited mutations in Presenilin1 (PSEN1) and has anage of onset before 65 years of age. PSEN1, the catalytic component of the gammasecretase, also plays a large role in neurodevelopment through regulating Notch signalingand subsequently regulating cell fate decisions during neurogenesis. Genetic mutationsin PSEN1 may then be disrupting this process through altering Notch signaling andcausing cellular changes that go unnoticed until AD-related behavioral changes manifest.Previous work has shown that PSEN1 embryonic knockout in mice causes prematureneuronal differentiation, depletion in neural stem cells, and is lethal at birth, supportingPSEN1's crucial role in proper neurodevelopment. While mouse models of AD are usefulin studying later stages of AD, there are a great number of differences between the mouseand human cortex during neurodevelopment. Using human induced pluripotent stem cell(iPSC)- derived cortical spheroids (hCS) allow for modeling the human cortex in vitro andallow access to studying the development of the human cortex at the cellular level. Weperform CRISPR/Cas9 gene editing in an isogenic iPSC line to generate distinct PSEN1mutations that have not been widely studied previously. In contrast to previous reports,we observe increased Notch signaling, an increase in neural progenitor cells and reducedneuronal differentiation in PSEN1 mutant hCS. We also observe increased size andmorphological changes in the hCS harboring PSEN1 mutations, which is blocked bytreatment with a Notch1 specific inhibitor, consistent with Notch signaling defects thatdepend on gain of function PSEN1 mutations. We will be determining if any of theneurodevelopmental changes due to PSEN1 mutations have further effects on AD-relatedneuropathology. Using this human cell-based model, these findings suggest that fADmutations alter neural progenitor expansion and neurogenesis during development,possibly predisposing neurodegeneration.

Disclosures: E. Hurley: None. **P. Mozolewski:** None. **R. Dobrowolski:** None. **J. Hsieh:** None.

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.03

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

Support: NIH Grant AI097299
NIH Grant NS104351
NIH Grant NS106585

Title: Latency instability in autonomic neuronal pathways contributes to herpes simplex virus (hsv1 and hsv2) recurrences

Authors: *G. A. MOORE[1], S. LEE[1], A. R. N. ABBOTT[2], K. L. JOHNSON[2], R. WIESKE[2], A. M. IVES[1], A. S. BERTKE[3];
[1]Biomed. and Vet. Sci., [3]Population Hlth. Sci., [2]Virginia Tech., Blacksburg, VA

Abstract: Herpes simplex virus (HSV) establishes life-long infection by traveling retrograde along neuronal axons after primary infection to establish latency in sensory and autonomic neuronal cell bodies that directly innervate the genitourinary system. The latent virus can then become reactivated by a variety of stimuli, traveling back down neuronal axons to induce recurrence of painful lesions. HSV1 and HSV2 are known to reactivate at different frequencies, and we propose the autonomic pathways contribute to this occurrence. Within the autonomic nervous system, HSV1 shows a preference for the sympathetic whereas HSV2 shows a preference for the parasympathetic neurons. To assess the contribution of sympathetic pathways in clinical recurrences, female guinea pigs were treated with 6-hydroxydopamine (6-OHDA) prior to infection to ablate sympathetic neuronal axons, making them unavailable for infection and the establishment of latency. Chemical ablation of sympathetic axons significantly reduced clinical recurrences by nearly 75% for HSV1 and by 50% for HSV2 (*p* &lt 0.001) indicating sympathetic pathways are responsible for a significant portion of HSV1 and HSV2 recurrences, with a greater impact on HSV1 than HSV2. To identify differences in latency and reactivation status in autonomic and sensory neurons, we compared viral gene expression in primary adult neuronal cultures from sensory dorsal root and autonomic major pelvic and sacral sympathetic ganglia. In contrast to sensory neurons, autonomic neurons supported spontaneous viral reactivation without induction stimuli. Further, HSV2 expressed viral genes in a cyclical pattern in autonomic neurons, likely contributing to higher frequency of recurrent genital disease. Taken together, autonomic pathways contribute to HSV1 and HSV2 clinical recurrences due to an inability to establish stable latency of HSV in autonomic neurons.

Disclosures: G.A. Moore: None. **S. Lee:** None. **A.R.N. Abbott:** None. **K.L. Johnson:** None. **R. Wieske:** None. **A.M. Ives:** None. **A.S. Bertke:** None.

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.04

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

Support: NIH Grant NS104351
NIH Grant NS106585
NIH Grant AI097299

Title: Herpes simplex viruses HSV1 and HSV2 suppress the Akt-GSK3β-β-catenin signaling pathway to maintain latency in sensory and sympathetic neurons

Authors: *A. S. BERTKE[1], P. GOSWAMI[2], T. HARRELL[3], L. CLAY[4];
[1]Population Hlth. Sci., [2]Translational Biol. Med. and Hlth., [3]Biomed. and Vet. Sci., [4]Col. of Sci., Virginia Tech., Blacksburg, VA

Abstract: Herpes simplex viruses (HSV1 and HSV2) establish latency in peripheral sensory and autonomic neurons, from which they can reactivate to cause recurrent disease episodes throughout a person's life. Various stimuli, such as stress or nerve injury, can cause viral reactivation, leading to painful skin lesions, ocular disease, benign recurrent meningitis or in some cases, encephalitis. However, molecular mechanisms regulating the reactivation process in response to different stimuli are unclear. Cortisol (CORT) reactivates HSV1 in sympathetic neurons and HSV2 in both sensory and sympathetic neurons. The canonical Wnt-β-catenin pathway maintains cell homeostasis and survival and CORT can impair these functions. Neurotrophic factors (NTFs) are target-derived ligands that promote neuronal survival, but also contribute to the maintenance of HSV latency, as deprivation of NTFs causes latent HSV to reactivate. NTFs maintain HSV latency through the PI3K-Akt pathway with GSK3β as one of its downstream targets. Our objective was to determine if signals from different reactivation stimuli potentially feed into the β-catenin signalosome via effector protein GSK3β to regulate the reactivation process in neurons. Using primary sensory and autonomic neurons cultured from trigeminal and superior cervical ganglia of adult mice, we established HSV1 and HSV2 latency for 7 days, followed by either CORT treatment or NTF deprivation to induce reactivation. Western blot analyses demonstrated that total Akt, GSK3β, and β-catenin were significantly reduced in HSV-infected neurons (reduced 67-98%, $p<0.05$), compared to uninfected neurons, with significant differences in β-catenin between sensory and sympathetic neurons (83/93% in sensory and 88/98% in sympathetic neurons infected with HSV1/HSV2, $p<0.05$). In sensory neurons, both CORT and NTF deprivation increased phosphorylation of GSK3β 3-4-fold, but only HSV1-infected neurons, correlated with an increase in β-catenin 4 hrs post treatment, suggesting these two stimuli converge on the β-catenin pathway in sensory neurons. In sympathetic neurons, β-catenin increased in response to NTF deprivation and decreased in response to CORT in uninfected neurons, with no significant changes in Akt or GSK3β. However, β-catenin was undetectable in HSV-infected sympathetic neurons. During establishment of latency, HSV caused a time-dependent reduction in β-catenin in primary cultured neurons, suggesting the virus may suppress this host protein to establish and maintain

latency in neurons. These findings have broader implications for neuron-specific effects caused by viral infections in different types of neurons.

Disclosures: **A.S. Bertke:** None. **P. Goswami:** None. **T. Harrell:** None. **L. Clay:** None.

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.05

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

Support: Fralin Rapid Response Grant

Title: SARS-CoV-2 rapidly infects the peripheral and central nervous systems of mice via neuronal entry facilitated by neuropilin-1 before detectable viremia

Authors: *J. JOYCE[1], G. MOORE[2], P. GOSWAMI[1], E. LESLIE[1], C. K. THOMPSON[3], A. S. BERTKE[4];
[1]Translational Biology, Medicine, and Hlth., [2]Biomed. and Vet. Sci., [3]Neurosci., [4]Population Hlth. Sci., Virginia Tech., Blacksburg, VA

Abstract: Up to 80% of patients with acute COVID-19 experience neurological symptoms. These symptoms can last long into recovery with 85% of those with post-acute sequelae of SARS-CoV-2 reporting them. We previously showed that sensory and autonomic ganglia of the peripheral nervous system, discrete brain regions, and the spinal cord are permissive to infection with SARS-CoV-2 in K18-hACE2 (hACE2) and wild-type mice. Invasion of these tissues may contribute to neurological symptoms of COVID-19, however the role of direct neural entry vs that of hematogenous entry, viral replication dynamics in neurons, and potential viral receptors beyond angiotensin-converting enzyme 2 (ACE2) have yet to be assessed. To differentiate between hematogenous and neural entry into the nervous system, we intranasally infected male and female hACE2 and WT mice (n=10 each) with SARS-CoV-2 isolate USA-WA1/2020 and collected tissues 18 and 48 hours post infection (n= 5 per group, daily). Peripheral nervous system sensory dorsal root (DRG) and trigeminal (TG) ganglia, autonomic superior cervical ganglia(SCG), brain regions (olfactory bulb, cortex, hippocampus, brainstem, cerebellum), whole brain hemispheres, and spinal cord samples were collected. We detected viral RNA in the PNS and CNS of both hACE2 and WT mice as early as 18 hpi, prior to detection in blood of only one hACE2 mouse at 42 hpi. Viral antigen was not detected via IF indicating the virus was transiting through the nervous system but had yet to replicate. Additionally, to determine viral growth curves in neurons, we infected primary neuronal cultures (SCG, TG, DRG) from hACE2 and WT mice and assessed viral replication via RT-qPCR, IF, and plaque assay up to five days post infection (dpi). Neuronal culture studies showed that viral genome replication and release of infectious virus occurs in hACE2 and WT neurons, with neuron specific kinetics. Since WT neurons with no hACE2 expression became infected, we pretreated primary neuronal cultures

(SCG, TG, DRG) from hACE2 and WT mice with the neuropilin-1 (NRP-1) antagonist EG00229 before infection to assess alternative viral receptors. Inhibition of NRP-1 reduced viral RNA by 99.8% and 86.7% in hACE2 and WT DRGs, respectively. Similar reductions of 71% and 89% were observed in TGs, and 95% in WT SCGs. Our results show that neuroinvasion occurs rapidly via direct neuronal entry prior to viremia, NRP-1 partially mediates neuronal entry, and neurons can be productively infected. Our data further underscores the importance of assessing the role of nervous system infection in the pathology of COVID-19 as it transitions from a pandemic disease to an endemic disease.

Disclosures: J. Joyce: None. **G. Moore:** None. **P. Goswami:** None. **E. Leslie:** None. **C.K. Thompson:** None. **A.S. Bertke:** None.

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.06

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

Support: AIIMS-Patna Intramural

Title: Short-term memory loss as a long covid symptom: indications from a follow-up study

Authors: *A. KUMAR[1], R. K. JHA[1], *G. KUMAR[1], P. DWIVEDY[1], A. ASGHAR[1], R. K. NARAYAN[7], S. K. GHOSH[1], R. KIRTI[2], D. RAI[3], L. TIWARI[4], A. BANERJEE[5], A. SARFARAZ[6];
[1]Dept. of Anat., [2]Dept. of Med., [3]Dept. of Pulmonary Med., [4]Dept. of Pediatrics, [5]Dept. of Biochem., [6]Dept. of Microbiology, All India Inst. of Med. Sciences-Patna, Patna, India; [7]Dept. of Anat., Dr. B.C. Roy Multispeciality Med. Res. Center, IIT Kharagpur, Kharagpur, India

Abstract: Background: The abruption of ongoing adult hippocampal neurogenesis and subsequent loss of neural functions in COVID-19 has been postulated by many authors, however, any direct clinical or laboratory evidence is still limited in this regard. We assessed the prevalence and pathogenesis of loss of short-term memory, a key hippocampal function, in discharged patients of COVID-19. **Methods:** A total of 209 hospitalized patients with COVID-19 were followed up post-discharge. The patients were verbally interviewed through a telephonic call and were asked about the presence of the symptoms confirming the loss of short-term memory, such as increased forgetfulness or a problem with retaining a piece of recent information. The time of occurrence and period of persistence of memory loss and presence of any other neurological/psychiatric symptoms, or that related to other systems, were noted for each case. Further, a retrospective analysis of the hospital data containing clinical, laboratory, and therapeutic details for each patient was made and statistically compared with a matched cohort having no long covid symptoms. **Results:** The loss of short-term memory was noted to be present in ~ 12% (25/209) of the followed-up patients. In the majority of the patients (22/25), the

problem persisted at the time of follow-up (up to one year since the date of discharge). The presence of no other neurological or psychiatric symptoms was noted in these patients. However, the dysfunction of the other systems such as gastritis, joint pain, and tiredness, have been found frequently associated. The retrospective analysis of the hospital data showed no significant association with the duration of hospital stay or severity of illness. However, systemic inflammation (↑ IL-6, ferritin, ESR, and CRP), vascular thrombosis (↑ PT/INR, ↑ D-dimer, ↓ platelets), metabolic derangements (↑ blood sugar and LDH levels, and ↓ uric acid), and electrolyte imbalance (↓ Na and Ca, ↑ K) were found as the most frequent associations. However, in comparison to the control, no significant differences were noted ($p>0.05$). **Conclusion:** The loss of short-term memory is a frequent long covid symptom in the survivors indicating that abruption of ongoing adult hippocampal neurogenesis may be an actual phenomenon in COVID-19. There is no distinct evidence that the systemic presence of inflammation, vascular thrombosis, or metabolic derangements are the causal factors leading to hippocampal dysfunction. A direct viral invasion of the hippocampal neurogenic niche remains an open possibility.

Disclosures: **A. Kumar:** None. **R.K. Jha:** None. **G. Kumar:** None. **P. Dwivedy:** None. **A. Asghar:** None. **R.K. Narayan:** None. **S.K. Ghosh:** None. **R. Kirti:** None. **D. Rai:** None. **L. Tiwari:** None. **A. Banerjee:** None. **A. Sarfaraz:** None.

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.07

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

Support: NS04817
DA044552
DA044552-03S1
DA033966
NS060632
MH122241

Title: Sars-cov-2 enhances hyperactivity of medial prefrontal cortex pyramidal neurons in hiv-1 tg rats and cocaine self-administered rats

Authors: *S. CASSODAY[1], L. CHEN[2], A. DONNER[1], L. AL-HARTHI[2], X.-T. HU[2]; [1]Rush Univ. Grad. Col., [2]Rush Univ. Med. Ctr., Chicago, IL

Abstract: Neurological manifestations including headache, sensory disturbances, and impaired cognition are commonly reported in SARS-CoV-2 infected patients. While the mechanism behind these neurological and neuropsychiatric symptoms is not fully understood, many can be attributed to hyperexcitability and neurotoxicity in cognitive regulating brain regions including

the medial prefrontal cortex (mPFC). Similar hyperexcitability/neurotoxicity is also found in people living with HIV (PLWH), especially in those diagnosed with HIV-associated neurocognitive disorder (HAND, a.k.a. neuroAIDS), which are worsened by chronic cocaine (Coc) abuse. Furthermore, substance use disorders (SUD; including cocaine use disorders, CUD) are often associated with HAND; while both patient populations suffer worse clinical outcomes upon infection with SARS-CoV-2. Collectively, these findings suggest a synergistic consequence of these pandemics on functional activity of neurons in the brain. However, it is unknown how and to what extent SARS-CoV-2 disturbs the activity of living brain neurons in the context of neuroHIV and CUD that may lead to these neuropsychiatric and neurocognitive deficits. To fill this knowledge gap, we used two combined animal models: a transgenic (Tg) rat model of neuroHIV (HIV-1 Tg rats) and a rat model of cocaine self-administration (Coc-SA). HIV-1 Tg rats self-administered Coc for 2 weeks, followed by a forced withdrawal period for 3 weeks during which, they underwent drug-seeking behavior assessments at days 3 and 21. Saline-yoked HIV-1 Tg rats, as well as non-Tg rats, were used as controls. Immediately following the withdrawal, rats were transcardially perfused, their brains were removed and sliced for electrophysiology evaluation. Firing of mPFC pyramidal neurons was assessed using whole-cell patch-clamping, with or without bath application of SARS-CoV-2 spike protein in a dose-dependent manner (1, 2.5 and 5nM). We found that there was no significant difference in Coc-taking behavior (number or volume) regardless of genotype. We also found that firing was significantly increased in mPFC neurons from HIV-1 Tg rats and Coc-SA rats; while SARS-CoV-2 spike protein further facilitated such neuronal hyperactivity. Thus, our data suggests SARS-CoV-2 likely enhances the deleterious effects of neuroHIV and/or Coc-SA by enhancing hyperexcitability/neurotoxicity in mPFC pyramidal neurons, which may jointly contribute to the mechanism underlying the syndemic of HAND/CUD/COVID-19.

Disclosures: **S. Cassoday:** None. **L. Chen:** None. **A. Donner:** None. **L. Al-Harthi:** None. **X. Hu:** None.

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.08

Title: WITHDRAWN

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.09

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

Support: NIGMS
NIEHS

Title: Covid-19 and the risk of dementia: insights from a novel transgenic mouse model of post-acute neurocognitive and neurologic sequelae of sars-cov-2 infection

Authors: K. M. KEYS[1], H. ZHANG[2], X. TIAN[4], Y. WANG[2], J. LONG[2], A. YUROCHKO[3], ***X.-H. LU**[2];
[1]Louisiana Univ. Shreveport Hlth. Sci. Cen, Shreveport, LA; [3]Microbiology, [2]LSU Hlth. Shreveport, Shreveport, LA; [4]Dept. of Pharmacology, Toxicology & Neurosci., Hlth. Sci. Ctr., Shreveport, LA

Abstract: With the emergence of SARS-CoV-2 infection across the globe and the rising record of reinfections and relapses, concerns arise regarding multiple symptoms and multi-tissue/organ injury that persist long past the time that patients have recovered from the initial stages of COVID-19 (Post-Acute Sequelae of SARS-CoV-2 infection, PASC, or long COVID), e.g., prominent neurocognitive and neurologic symptoms: "brain fog," mood changes, sleep problems, and anosmia/ ageusia. Virus-host interaction provokes the evolutionarily conserved response to DNA damage, known as the DNA damage response (DDR). orchestrated by Ataxia-telangiectasia mutated (ATM), dictating the fate of cells into apoptosis or senescence to prevent the replication of a damaged genome. To understand the causal pathogenetic role systemic inflammation driven genotoxic stress in long COVID, a vital early step is to develop animal models with high construct, face, and predictive validities that can be easily deployed by the broad research community. We have successfully developed a human ACE2 Bacterial artificial Chromosome (BAC) transgenic mouse model with full-length human ACE2 regulatory regions that faithfully recapitulated the structure, tissue distribution, and gene regulation of the human gene. To facilitate the analysis of PASC in an ABSL-2 facility, we developed a strategy to model the acute or senescence induction phase of SARS-CoV-2 infection that recapitulates the signature cytokine storm and acute respiratory distress syndrome (ARDS). Known genotoxic stress-inducers: Doxorubicin or Bleomycin, and environmental genotoxic agents (e.g., Parkinson's disease (PD)-associated herbicide, Paraquat) exerted strong genotoxic stress and senescence phenotypes to facilitate the virus infection, which can be blocked by an ATM inhibitor, KU60019. Robust genotoxic stress was also observed in the brain in cortical neurons and microglia, with no obvious viral neuroinvasion. Two months after infection, the mice manifested long-lasting cognitive deficits and anosmia. Thus, we generated a novel small animal model of to study long lasting effects of environmental exposure and SARS-CoV2 interaction that can be easily deployed by a broad research community. Exploiting knowledge on environmental exposure-host-virus interaction in disease susceptibility, severity and manifestation of neurological and neurocognitive PASC will enhance our ability to address the long-term clinical outcome of viral infection, as it will shed light on the underlying mechanisms of pathobiology at molecular virus-host interfaces.

Disclosures: K.M. Keys: None. **H. Zhang:** None. **X. Tian:** None. **Y. Wang:** None. **J. Long:** None. **A. Yurochko:** None. **X. Lu:** None.

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.10

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

Title: Hsv-1 intranasal infection contributes to olfactory behavioral deficits in 5xfad mice

Authors: *C. S. NIEMEYER[1], J. E. HASSELL, Jr.[2], L. MERLE[3], B. BAXTER-BOLT[4], A. BUBAK[5], D. RESTREPO[6], M. A. NAGEL[4];
[1]12700 E. 19th Ave, RC2 Room 5450C, Univ. of Colorado Denver Sch. of Med., Aurora, CO; [2]Univ. of Colorado Anschutz, Univ. of Colorado Anschutz, Aurora, CO; [3]Univ. of Colorado, Univ. of Colorado, Aurora, CO; [4]Univ. of Colorado, Sch. of Med., Aurora, CO; [5]CU Procurement, Univ. of Colorado, Denver - Dept. of Neurol., Aurora, CO; [6]Universtiy of Colorado Anschutz Med. Campus, Aurora, CO

Abstract: Multiple studies show that herpes simplex virus type-1 (HSV-1) contributes to Alzheimer's disease (AD) progression. HSV-1 increases the risk of dementia and infection shares similar pathological characteristics as those seen in AD, including amyloid accumulation, neuroinflammation, neurodegeneration, and cognitive impairment. In a parallel body of literature, early AD is characterized by smell loss that has been associated with amyloid deposition in the olfactory epithelium (OE) and olfactory bulb (OB), neurodegeneration, and cognitive decline. These studies raise the possibility that HSV-1 disruption of olfactory pathways can accelerate AD. Thus, we hypothesize that HSV-1 infection of the olfactory system triggers smell loss and pathological processes characteristic of early AD. To test this hypothesis, we determined if HSV-1 can infect the olfactory system from the periphery. Mice (5xFAD, AD murine model; C57BL/6) were intranasally inoculated with HSV-1 (10e6 PFU/animal, McKrae strain) or PBS. Five days post-infection (DPI), immunohistochemical analysis revealed more HSV-1 antigen in the OB of the 5xFAD compared to C57BL/6. Surprisingly, C57BL/6 had more HSV-1 antigen in brainstem nuclei compared to 5xFAD, particularly in the trigeminal nuclei. We also found amyloid deposition was increased in the OE of HSV-1 infected mice compared to mock-infected mice. Next, we determined the effects of viral infection on olfactory behavior using a food foraging task. Prior to infection, performance on a food foraging task was assessed in 5xFAD mice and littermate controls. Mice were then intranasally inoculated as above and olfactory deficits (latency to find food compared to pre-infection) were assessed by the food foraging task at 10 and 30 DPI. We found the 5xFAD mice had significantly increased latency to find food at 10 DPI following infection compared to littermate controls; further, these olfactory deficits persisted through 30 DPI. These findings suggest that HSV-1 can accelerate olfactory deficits in vulnerable mice with progressive AD pathology and, by extension, may accelerate progression to clinical dementia in individuals with familial AD.

Disclosures: C.S. Niemeyer: None. **J.E. Hassell:** None. **L. Merle:** None. **B. Baxter-Bolt:** None. **A. Bubak:** None. **D. Restrepo:** None. **M.A. Nagel:** None.

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.11

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

Support: NIH grant R01 DA050540

Title: SARS-CoV-2 N-protein promotes neuroinflammation via microglial NLRP3 inflammasome activation

Authors: *D. DUTTA, J. LIU, H. XIONG;
Dept. of Pharmacol. and Exptl. Neurosci., Univ. of Nebraska Med. Ctr., Omaha, NE

Abstract: In addition to excessive pulmonary inflammatory responses, SARS-CoV-2 infection also causes neurological syndrome in COVID-19 patients. Mechanisms for SARS-CoV-2-associated neurological syndromes are multifactorial. Increasing evidence indicates that SARS-CoV-2 proteins play a crucial role in SARS-CoV-2-associated neuroinflammation. It is our hypothesis that the SARS-CoV-2 nucleocapsid (N) protein, a highly immunogenic viral protein, promotes microglia activation and resultant neuroinflammation. To test this hypothesis, we studied SARS-CoV-2 N-protein on NLRP3 inflammasome activation in primary rat microglial cultures using immunocytochemical staining, RT-qPCR, immunoblotting and ELISA. Our results showed that treatment of microglia with N-protein (0.1-10 μg/ml) produced a dose-dependent microglia activation as detected by Iba-1 expression. The N-protein-treated microglial cells also exhibited a dose-response in the processing of NLRP3, caspase-1 and IL-1β. Immunocytochemical staining and enzyme activity assay revealed that N-protein induced NLRP3 and caspase-1 colocalization and increased caspase-1 activity. ELISA analyses demonstrated that N-protein significantly enhanced secretion of proinflammatory cytokines IL-1β, TNF-α, and IL-6. The involvement of N-protein in neuroinflammatory processes was further confirmed by the results showing an increased iNOS-mediated NO production. The aforementioned effects associated with N-protein were significantly attenuated by either a specific NLRP3 inhibitor MCC950 or a caspase-1 inhibitor Ac-YVAD. Taken together, these results demonstrated that the N-protein of SARS-CoV-2 promoted microglial NLRP3 inflammasome activation and resultant neuroinflammatory responses, which may underlie the pathogenesis of neurological syndromes seen in patients with COVID-19.

Disclosures: D. Dutta: None. **J. Liu:** None. **H. Xiong:** None.

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.12

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

Title: Sars-cov-2 s1 spike protein induces neuroinflammation in mice: a temporal profile of systemic and central cytokine expression

Authors: *E. B. DEFENSOR, E. CHU, C. E. WOODS, S. E. BLUMENFELD, S. FALSAFI, R. K. LAM, P. CIARI, W. TAN, A. E. BARRON, M. SHAMLOO;
Stanford Univ., Palo Alto, CA

Abstract: Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has been associated with neurological symptoms, including headache, confusion, and loss of taste or smell. Reduction in global brain size and other changes in brain structure have also been reported with SARS-CoV-2. While there are multiple mechanisms by which SARS-CoV-2 may impact the brain, it is suggested that the neurological manifestations are a result of a widespread immune response. The objective of the current study was to determine the effects of peripherally administered SARS-CoV-2 S1 spike protein (S1) in mice. Male C57BL/6J mice, aged 8-9 weeks, were dosed intraperitoneally with 20 µg of S1. To determine the impact on working memory, independent groups were tested in a Y-Maze on day 1 or day 3 after S1 delivery. To determine the impact on cytokine profile, mice were terminated, and tissue was collected on day 2 or day 4 after S1 delivery. Plasma and brain tissue were subsequently analyzed using a Luminex multiplex assay. Results from the Y-maze showed trends for decreased locomotor activity but did not show working memory impairment with S1 administration. Analysis of brain and plasma showed pro-inflammatory changes in cytokine profile with S1 administration. Taken together, these data provide utility in refining in-vivo models for SARS-CoV-2 and also identify potential therapeutic targets for future investigations.

Disclosures: E.B. Defensor: None. **E. Chu:** None. **C.E. Woods:** None. **S.E. Blumenfeld:** None. **S. Falsafi:** None. **R.K. Lam:** None. **P. Ciari:** None. **W. Tan:** None. **A.E. Barron:** None. **M. Shamloo:** None.

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.13

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

Support: Morrison Trust
San Antonio Medical Foundation

Title: Sars-cov-2 protein disruption of the blood-brain-barrier, distinct from cellular viral infection, leading to a maladaptive inflammatory response playing a role in long haul covid-19 symptoms

Authors: *A. NI[1], N. XU[1], I. BAZALDUA[2], F. VIGIL[2], M. SHINN[3], E. SUN[1], S. VYAS[1], I. A. MUZZIO[4], M. S. SHAPIRO[2];
[1]Long Sch. of Med., [2]Cell. and Integrative Physiol., Univ. of Texas Hlth. Sci. Ctr., San Antonio, TX; [4]Neuroscience, Developmental, and Regenerative Biol., [3]Univ. of Texas at San Antonio, San Antonio, TX

Abstract: Over 500 million people worldwide have contracted Covid-19 disease from SARS-Cov-2 infection. Between 22-32% of those display persistent neurological symptoms of "Long-Haul Covid" (LHC), whose etiology are still largely unknown, such as brain fog, mood disorders, cognitive impairments, and delirium after initial recovery. Even more of concern is that a large fraction of those sickened, but not hospitalized, also display LHC neurological dysfunction that presents long past the initial recovery. We are testing the hypothesis that certain SARS-Cov-2 proteins themselves, distinct from cellular viral infection, cross or disrupt the Blood-Brain-Barrier (BBB), leading to a profound and deleterious mal-adaptive inflammatory response. In our *in vitro* experiments, we compared the resulting microgliosis, astrogliosis, elevations of various inflammatory biomarkers, and neuronal death after tail-vein injection (*i.v.*, 5 ug) of the spike (S) protein S1 subunit and the nucleocapsid (N) protein in wild-type (C57/BI6) mice. We also are performing *in vivo* 2-photon microcopy through a glass window over the cortex to assay BBB permeability of a large-molecular weight dye-conjugated dextran into the parenchyma. Additionally, we are conducting behavioral assays and *in vivo* electrophysiological recordings to determine the effect of these proteins on cognition and brain spatial representations. We administer the proteins or vehicle only as above and evaluate memory and spatial orientation using an object/place recognition task and a reorientation paradigm. Our data indicate that animals injected with the S1 subunit show profound memory and spatial deficits 24 hr following training. *In vivo* recordings from retrosplenial cortex, a region important for memory and navigation, indicate that animals injected with the S1 subunit lack spatial tuning, which parallels the pattern of behavioral disorientation exhibited by this group. The spatial confusion persists at least 4 days following the protein injection, suggesting that "Covid-fog" could in part be due to the effects of the S subunits themselves. Our results may demonstrate that profound inflammation generated by certain SARS-Cov-2 proteins underlie long-term brain damage and dysfunction that likely persist long after the resolution of the acute infectious phase of the disease. In addition, we will show, *in vivo*, whether behavioral deficits, such as impaired memory and spatial recognition, are due to the same mechanism. We hope our findings will lead to novel modes of therapeutic intervention to prevent long-lasting neurological damage in those with Covid-19, or to help those who have already been affected.

Disclosures: A. Ni: None. **N. Xu:** None. **I. Bazaldua:** None. **F. Vigil:** None. **M. Shinn:** None. **E. Sun:** None. **S. Vyas:** None. **I.A. Muzzio:** None. **M.S. Shapiro:** None.

Poster

538. Neuroinflammation and Neurodegeneration: HSV and SARS-CoV2

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 538.14

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

Support: NS121741

Title: Coronavirus Infection leads to changes in Cholesterol Biosynthesis Genes in Hippocampus of Susceptible mice.

Authors: *J. TINGLING[1], A. LOUIE[2], A. STEELMAN[3];
[1]Div. of Nutritional Sciences, Univ. of Illinois at Urbana-Champaign, Urbana, IL; [2]Neurosci. Program, Univ. of Illinois at Urbana-Champaign, Urbana, IL; [3]Animal Sci. Department, Univ. of Illinois at Urbana-Champaign, Urbana, IL

Abstract: Members of the viral family *Coronaviridae* contain enveloped large positive-sense single stranded RNA genomes (~25-32 kb). While these viruses have been known to infect the respiratory, hepatic, digestive and central nervous systems of animals and humans, the current pandemic, caused by the betacoronavirus SARS-CoV-2, has presented ongoing challenges to the health community. Intranasal inoculation of Murine hepatitis virus (MHV-1), a betacoronavirus, and naturally occurring pathogen of mice, into the susceptible A/J mouse strain reproduces many clinical features of SARS including: high mortality, lung pathology, and uncharacteristically high levels of the pro-inflammatory cytokines (a cytokine storm). Thus, this mouse-specific virus provides a clinically relevant animal model with which to study coronavirus pathogenesis. Previous studies have demonstrated influenza virus infection causes changes in cholesterol biosynthesis and myelin genes within various regions of the brain. In this study we sought to establish the effect of MHV-1 infection on the expression of proinflammatory gene expression in the lung as well as cholesterol biosynthesis and myelin genes in the hippocampus. The study was performed by using 6-week-old male and female A/J mice that were inoculated intranasally with 5000 P.F.U. of MHV-1 and were sacrificed on day 7 p.i. Daily total body weights were recorded, mRNA levels of inflammatory genes, as well as viral RNA levels, were assessed in the lungs of mice as well as the hippocampus.Mice inoculated with MHV-1 had significantly greater weight loss and had increased expression levels of genes involved in inflammation including *Ifng, Tnf, Il6, Cxcl2, Ccl5, Cxcr2* and *Ccr5* as well as viral mRNA in the lungs compared to saline inoculated controls. Viral mRNA was detectable in the hippocampus of infected mice. We also found that three cholesterol biosynthesis genes, (*Dhcr7, Hmgcs1*, and *Mvd*) were significantly decreased in the hippocampus of infected mice. Other cholesterol biosynthesis genes were also noted to be relatively low as compared to controls. These findings indicate that viral coronavirus mRNA can be detected 7 days p.i. in the brains of infected mice, specifically the hippocampus which is responsible for learning and memory in mammals. We also have demonstrated that our model of coronavirus infection produces the characteristic cytokine profile that has been previously reported. Further research into the impact of coronavirus infection on learning and memory in a susceptible strain of mice can be critical in elucidating the long-term effects of coronavirus respiratory infection.

Disclosures: J. Tingling: None. **A. Louie:** None. **A. Steelman:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.01

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH-R21AG057974
NIH-R25NS080687
HRD-1906130

Title: Establishing effective parameters for electroporation of echinoderm nerve cord explant

Authors: *Y. MIRANDA-NEGRÓN, J. E. GARCÍA-ARRARÁS;
Univ. de Puerto Rico, Rio Piedras, San Juan, PR

Abstract: For decades, scientist have employed extensive resources to elucidate important gene regulators involved in the process of nervous tissue regeneration. Regeneration-competent organisms have been studied to learn how to overcome the limitations present in mammalian central nervous system (CNS) regeneration. Our laboratory has used the sea cucumber *Holothuria glaberrima*, a deuterostome with amazing regeneration capacities, as a model system to study CNS regeneration. Nevertheless, the available techniques to study *H. glaberrima* nervous system at a molecular level are still limited. Hence, we have focused on developing new tools, among these a gene silencing method via siRNA transfection for radial nerve cord explants. This technique enables us to study the role of specific genes in the regeneration process. As a first step in the development of the siRNA we needed to develop an effective transfection method. Isolated radial nerve cord explants were electroporated using different parameters. Electroporation parameters were explored by altering either electric field strength (Volts) or electric field exposure time (msec). Electroporation under the presence of a fluorescent reporter dye (Tetramethyl Rhodamine) served as a first quality control filter to assess explant morphology, survival, and cell dye permeability. Radial nerve cord explants were electroporated with siRNA for *Myc* transcription factor to determine the possible effect on mRNA levels. Parameters tested ranged from 7V to 150V and exposure times of 5 to 65ms. Our results demonstrated that the radial nerve cords tolerated a broad electric field with little to no change in radial nerve cord morphology. Conversely, exposure times above 55msec or 150V led to substantial cell death. Electroporation trials indicated that increasing field strength (up to 80V) while lowering the electric field exposure time to 5ms improved the explant dye uptake response with no indication of cellular damage. Ongoing experiments are targeted toward optimizing the inhibition of Myc expression in order to establish a viable method to transfect *H. glaberrima* CNS in vitro. Successful radial nerve cord explant gene knockout has never been achieved in echinoderms, therefore setting up this protocol would help advance the characterization of gene function in this important group of animals.

Disclosures: Y. Miranda-Negrón: None. **J.E. García-Arrarás:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.02

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Grant NS108189

Title: Consequences of AAV-retro mediated deletion of PTEN following cervical spinal cord injury in mice

Authors: *M. METCALFE, O. STEWARD;
Anat. and Neurobio., Univ. California Irvine, Irvine, CA

Abstract: Cre-driven deletion of phosphatase and tensin homolog (PTEN) in neurons in $PTEN^{f/f}$ mice reliably enables axon regeneration after spinal cord injury (SCI). Here, we test efficacy of an approach using AAV/Cre vectors that are transported retrogradely from an injection site in the spinal cord to the majority of neurons that give rise to spinal tracts. We report results of 3 separate studies that use $PTEN^{f/f}$;$Rosa^{tdTomato}$ mice in which Cre induces expression of tdT and simultaneously deletes PTEN. In two studies, controls were $Rosa^{tdTomato}$ mice that received AAV-retro/Cre; in the other, controls were $PTEN^{f/f}$;$Rosa^{tdTomato}$ mice that received AAV-retro/GFP. 8-week old female and male mice received different genome copies (GC) of AAV-retro at the same time as a dorsal hemisection injury at C5 (12E9 GC: $Rosa^{tdTomato}$ n=13, $PTEN^{f/f}$;$Rosa^{tdTomato}$ n=12; 6E9 GC: AAV-retro/Cre n=9; AAV-retro/GFP n=7; 3E9 GC: $Rosa^{tdTomato}$ n=15, $PTEN^{f/f}$;$Rosa^{tdTomato}$ n=25). Forelimb motor function was assessed over time using a grip strength meter by individuals who were blind to treatment group. Following SCI, average grip strength decreased to <15% of pre-operative control immediately after injury and then gradually recovered. In studies involving higher (12E9 GC) and mid (6E9 GC) doses of AAV-retro/Cre, $PTEN^{f/f}$;$Rosa^{tdTomato}$ mice exhibited greater recovery than respective controls. Mice that received 12E9, 6E9, and 3E9 GC, recovered to a peak of 74.2%, 21.6%, and 5.2% of pre-operative control values respectively. Despite initial recovery, grip strength began to decline at around 1 month post injury in PTEN-deleted mice and at 2-3 months post-injury, 35-50% of the mice began to exhibit incessant scratching and hindlimb dystonia. These late developing pathophysiologies were not observed in un-injured $PTEN^{f/f}$;$Rosa^{tdTomato}$ mice that received intra-spinal cord injections of AAV-retro/Cre, indicating that the pathophysiologies are not due to PTEN deletion alone, or AAV itself. In addition to retrograde transduction of cells of origin of spinal pathways in the brain, some DRG neurons in ganglia near the injection site are also transduced. However, co staining for tdT and calcitonin gene-related peptide (CGRP) revealed no transduction of CGRP-positive neurons that give rise to C-afferents that mediate nociception. We conclude that, although intra-spinal injections of AAV-retro/Cre in $PTEN^{f/f}$;$Rosa^{tdTomato}$ mice can lead to initial improvements in forelimb motor recovery after SCI, there are late-developing functional abnormalities with the experimental conditions used here. The mechanisms underlying late-developing pathophysiologies remain to be defined.

Disclosures: M. Metcalfe: None. **O. Steward:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); OS is a co-founder and has economic interests in the company Axonis Inc, which holds a license on patents relating to PTEN deletion and axon regeneration.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.03

Topic: C.11. Spinal Cord Injury and Plasticity

Support: Supported by Operational Programme Research, Development and Education in the framework of the project "Center of Reconstructive Neuroscience" .02.1.01/0.0./0.0/15_003/0000419
The Charles University Grant Agency (GA UK), project number 320421

Title: Aav-mediated gene therapy for sensory regeneration after spinal cord injury

Authors: B. SMEJKALOVA[1,2], K. STEPANKOVA[1,2], P. JENDELOVA[1,2], ***L. MACHOVA URDZIKOVA**[1], J. C. F. KWOK[3,1], J. W. FAWCETT[4,1];
[1]Inst. of Exptl. Med., Prague, Czech Republic; [2]Dept. of Neuroscience, Charles University, Second Fac. of Med., Prague, Czech Republic; [3]Fac. of Biol. Sciences, Univ. of Leeds, Leeds, Czech Republic; [4]Cambridge Univ., Cambridge Univ., Cambridge, United Kingdom

Abstract: After spinal cord injury (SCI), axons cannot regenerate mostly due to an inhibitory environment consisting of the highly upregulated tenascin-C and chondroitin sulphate proteoglycans (CSPGs). This suggests that expression of an appropriate integrin isoform, which binds and uses tenascin-C as substrate, might enhance axon regeneration after SCI. During development, tenascin-C promotes growth of axons in presence of a tenascin-binding integrin, particularly α9β1. After SCI, adult neurons do not upregulate an integrin α9 that interacts with tenascin-C. In addition, integrins can be inactivated by CSPGs, however, this inhibition can be overcome by the expression of an integrin activator, kindlin-1. The aim of this project was to achieve sensory axon regeneration in rats with dorsal column crush lesion using viral vector delivery of the relevant genes (combination of integrin α9 and kindlin 1 in 3:1 ratio, kindlin 1 alone or GFP) to the DRG. We addressed two different levels of SCI, C4 lesion with DRG C6 and C7 injections for forelimb sensory restoration and T10 lesion with DRG L4 and L5 injections for hindlimb sensory restoration. The animals (Lister Hooded females, n=12/per group) underwent dorsal column crush injury with concurrent DRG injections followed by 12 weeks of behavioural testing. Significant improvement was observed in Von Frey test for mechanical perception and Hargreaves test for thermal sensation in treated animals with both, cervical and thoracic lesions when compared to controls. Tape Removal Test was improved only in treated animals with T10 lesion. Positive behavioural outcome was confirmed by staining for cFos below and above lesion and TRPV1 and Kv7.2 channels in DRGs. Histology of regenerating axons shows GFP and V5 positive axons from the integrin α9 and kindlin 1 group growing above the lesion on tenascin C rich surface. GFP and V5 staining confirmed that transport of integrin and kindlin occurred over the full regeneration distance in the regrowing axons in the spinal cord, from L4, L5 up to cervical cord. About 40% of axons below the lesion regenerated their axons to at least 4cm above the T10 lesion. Axons regenerating above C4

lesion were partly growing through meninges. In conclusion, the AAV-mediated gene therapy leads to sensory regeneration after SCI at C4 and T10 level, as proved by behavioural tests and immunohistochemical staining.

Disclosures: **B. Smejkalova:** None. **K. Stepankova:** None. **P. Jendelova:** None. **L. Machova Urdzikova:** None. **J.C.F. Kwok:** None. **J.W. Fawcett:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.04

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NEURORECON CZ.02.1.01/0.0/0.0/15_003/0000419
IRP grant P186

Title: Pi3kδ Gene therapy in spinal cord injury leads to axon regrowth and function restoration

Authors: *K. KAROVA[1], Z. POLCANOVA[1], S. SUCHANKOVA[2], R. HOLOTA[3], L. MACHOVA URDZIKOVA[1], P. JENDELOVA[1], J. C. F. KWOK[4,1], R. TURECEK[2], J. W. FAWCETT[5,1];
[1]Ctr. for Reconstructive Neurosci., [2]Dept. of Auditory Neurosci., Inst. of Exptl. Med., Prague, Czech Republic; [3]Inst. of Biol. and Ecology, Kosice, Slovakia; [4]Sch. of Biomed. Sci., Univ. of Leeds, Leeds, United Kingdom; [5]Cambridge Univ., Cambridge Univ., Cambridge, United Kingdom

Abstract: Development and maturation of the central nervous system naturally lead to severe loss of its regeneration potential. After injury of adult spinal cord axons, we don´t observe much regrowth or formation of new functional connections, which are necessary for correct gait and fine movement. Here, we use PI3Kδ gene therapy to show that we can stimulate cortical neurons to regenerate their axons and improve behavioural outcomes. PI3Kδ produces PIP3, a key signalling lipid regulating motility, protein translation, transport/trafficking and epigenetic controls. In mature neurons, much of the growth machinery is excluded from axons, and PI3Kδ enables anterograde transport of at least some of such restricted molecules, like the integrins, which have been shown to promote growth. We used a model of C4 dorsal column lesion in male/female rats and injected the right motor cortex at 4 sites concurrently with a total of 2 µl of viral vector mixture of AAV1-hSYN-eGFP + AAV1-hSYN-PIK3CD or with titre matched AAV1-hSYN-eGFP only. We allowed rats to survive for 12 or 16 weeks before transcardial perfusion. Using fluorescent immunohistochemistry, we evaluated levels of PI3Kδ and GFP in 40 µm frozen floating brain sections and determined over 80% co-expression as soon as 6 weeks after transduction which remained stable at both 12 (n=4) and 16 weeks (n=5). Next, we counted GFP labelled axons in 20 µm spinal cord sagittal sections and found hundreds of axons extending at least 1.3 cm below lesion after both 12 weeks (n=4) and 16 weeks (n=5) with more

axons at the later time point, reflected with corresponding regeneration indexes. Rigorous weekly behavioural testing for 16 weeks revealed functional improvements in skilled paw reaching, grip strength and ladder rung walking in rats treated with PIK3CD (n=15) compared to GFP only controls (n=14). Functional recovery of PIK3CD treated rats (n=7) was also confirmed with electrophysiological recordings of responses from areas below lesion and in forelimb muscles after stimulation of the right pyramid using a tungsten electrode (5 square pulses at 300 Hz) when compared to controls. Increasing current amplitudes between 30 and 300 μA were used to elicit cord dorsum potentials (CDPs), measured with silver ball electrodes above and below lesion (0.5 and 1 cm below), as well as EMG of the left forepaw distal flexor muscles. Subsequent re-lesion led to loss of responses. From our data, we conclude that gene therapy driven expression of PI3Kδ in cortical neurons induces robust axon regeneration and results in significant function restoration.

Disclosures: K. Karova: None. **Z. Polcanova:** None. **S. Suchankova:** None. **R. Holota:** None. **L. Machova Urdzikova:** None. **P. Jendelova:** None. **J.C.F. Kwok:** None. **R. Turecek:** None. **J.W. Fawcett:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.05

Topic: C.11. Spinal Cord Injury and Plasticity

Support: Department of Defense SC170295

Title: Evaluation of novel conopeptide with CB1 receptor activity in a model of chronic central pain

Authors: *S. JERGOVA[1], K. LIEBMANN[1], J. DING[1], J. IMPERIAL[2], B. M. OLIVERA[2], J. SAGEN[1];
[1]Univ. of Miami, Miami, FL; [2]Univ. of Utah, Salt Lake City, UT

Abstract: Cannabinoids are a promising class of analgesic agents and preclinical studies in rodent models suggest they may be particularly potent in relieving spinal cord injury (SCI)-induced neuropathic pain. However, clinical acceptance of cannabinoids has been limited due to CNS side effects. The goal of the proposed studies is to overcome clinical barriers by identifying novel cannabinoid peptides. Marine cone snails produce a wealth of selective peptides (conopeptides) with analgesic activities. We have previously screened and identified possible Conus venom fractions possessing cannabinoid 1 (CB1) receptor activities. The current project aimed to obtain a purified venom fraction with CB1 activity and to identify its gene therapy potential. Previously identified HPLC fractions from C. Textile, CTex-185 and CTex-195, underwent immunoprecipitation to obtain CB1 active components, were analyzed by Mass Spectrometry and their sequences were identified. AAV2/8 viral particles encoding CTex-185 or

CTex-195 sequences were designed and engineered by Vector Builder. Male Sprague Dawley rats (220-250g) underwent spinal cord clip compression injury. AAV2/8_CTex-185/ CTex-195/ or control GFP were then injected at 4 weeks post SCI using intraspinal, intrathecal, or intra-DRG routes. Changes in tactile, cold and heat hypersensitivity were monitored weekly up to 10 weeks. CB1 antagonist AM251 was injected in some rats to evaluate CB mechanisms. CSF levels of inflammatory cytokines were evaluated at the end of experiment. Results showed analgesic effects of the CTex AAVs via all three injection routes, with DRGs injections appearing most potent and prolonged for both transgenes. The CTex-195 produced more robust antinociceptive effects overall, particularly in reducing tactile hypersensitivity via the intrathecal and intraspinal routes. AM251 partially reduced the analgesic effects of CTex-195. Spinal CSF samples taken from the CB1 AAV-treated animals retained CB1 internalization capacity, with CTex-195 significantly more potent than CTex-185, and this activity was trypsin sensitive, supporting CB1 peptidergic activity. The level of TNFα in CSF and spinal cord homogenates was reduced in animals treated with CTex-195, in contrast to elevated levels in SCI animals with the control AAV treatment. IL-1β levels were also reduced in both groups of CB1 conopeptide treated animals. These finding indicate that the identified C. Tex fractions have the capacity to be used in gene therapy to manage chronic SCI pain. This study provides a first step in the identification of novel cannabinoid receptor-active substances suitable for gene therapy of chronic pain.

Disclosures: S. Jergova: None. **K. Liebmann:** None. **J. Ding:** None. **J. Imperial:** None. **B.M. Olivera:** None. **J. Sagen:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.06

Topic: C.11. Spinal Cord Injury and Plasticity

Title: The therapeutic effects of mesenchymal stem cell transplantation in ischemic spinal cord injury

Authors: *J. HWANG[1], J. YOO[1], J. CHOI[1], M. RAMALINGAM[1], E. JANG[1,2], C. KIM[3], H.-S. JEONG[1], S. JANG[1];
[1]Dept. of Physiol., Chonnam Natl. Univ. Med. Sch., Hwasun-gun,, Korea, Republic of;
[2]Biopharmaceutical Res. Ctr., Jeonnam Bioindustry Fndn., Hwasun-gun,, Korea, Republic of;
[3]Neurosurg., Kangwon national university, Chucheon, Korea, Republic of

Abstract: The therapeutic effects of mesenchymal stem cell transplantation in ischemic spinal cord injury
Jinsu Hwang[1], Jin Yoo[1], Jiyun Choi[1], Mahesh Ramalingam[1], Eunjae Jang[1,2], Choonghyo Kim[3], Han-Seong Jeong[1], Sujeong Jang[1,*1]Department of Physiology, Chonnam National University Medical School, Hwasun, Jeollanam-do 58128, Korea[2]Biopharmaceutical Research Center,

Jeonnam Bioindustry Foundation, Hwasun, Jeollanam-do 58141, Korea[3]Department of Neurosurgery, Kangwon National University, College of Medicine, Chuncheon, KoreaThere are many paraplegic patients worldwide due to spinal cord injury, and viable options for regenerative repair are desperately needed. The therapeutic effects of human adipose-derived mesenchymal stem cells (hADSCs) transplantation have been demonstrated in several studies, however, the exact underlying molecular mechanism is poorly understood. The aim of this study is to investigate the therapeutic potential of hADSCs in experimentally induced ischemic spinal cord injury. To evaluate the ADSC treatment and the inhibitory effects of ADSCs on the molecular mechanism, mice were subjected to ischemic spinal cord injury following aortic clamping surgery for 8 min, and neural-induced ADSCs (NI-hADSCs) were injected into the lesion site for 1 week after the injury. Motor function was evaluated using the Basso Mouse Scale (BMS) for 8 weeks. Spinal cord neuropathies and neuron apoptosis were observed by HE and TUNEL staining. To validate the hypothesis of the therapeutic effects of hADSCs, neuronal staining and analyses of microglia/macrophage activation and molecular signaling pathways were performed. We demonstrated that the number of dead cells and microglia/macrophage activation was reduced after treatment of hADSCs. Also, Wnt/Notch signaling molecules were observed and the expression was increased via the injured model. However, we could not find axonal regeneration in the treatment. The result of this study demonstrated that the therapeutic effects of hADSCs in ischemic spinal cord injury were partly due to Wnt/Notch signaling pathway. In addition, hADSCs could recover the nerve cells and preserve the supporting cells in the spinal cord.

Disclosures: J. Hwang: None. **J. Yoo:** None. **J. Choi:** None. **M. Ramalingam:** None. **E. Jang:** None. **C. Kim:** None. **H. Jeong:** None. **S. Jang:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.07

Topic: C.11. Spinal Cord Injury and Plasticity

Support: PA Department of Health 4100089346
NIH/NINDS R01NS121336

Title: Transplanting embryonic neural progenitor cells to rebuild supraspinal regulation for micturition recovery after spinal cord injury

Authors: K. PATEL, E. OATMAN, *S. HOU;
Marion Murray Spinal Cord Res. Center, Dept. of Neurobio. & Anat., Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: Traumatic spinal cord injury (SCI) often interrupts spinobulbospinal micturition reflex pathways. The loss of descending regulation results in urinary dysfunction. Although a partial voiding capability may recover due to intraspinal plasticity of segmental reflex circuitry, the

emergence of bladder hyperreflexia and detrusor-sphincter dyssynergia (DSD) causes incontinence and inefficient voiding, leading to severe consequences such as repeated lower urinary tract (LUT) infections and kidney damage. To reestablish supraspinal control of the LUT, we transplanted embryonic day 14 (E14) brainstem-derived neural progenitor cells (BS-NPCs) into the injured spinal cord and evaluated urination. Adult female F344 rats underwent a clinically-relevant contusive injury at the 10th thoracic spinal cord (T10). Subsequently, disassociated E14 BS-NPCs ubiquitously expressing green fluorescent protein were implanted into the lesion site five days post-injury. Naive and injury only rats served as two controls. Twelve weeks after SCI and transplantation, anterograde or transsynaptic neuronal tracings were implemented with histological analysis to examine the reconnection of supraspinal micturition pathways. Bladder cystometrogram (CMG) and external urethral sphincter (EUS) electromyography (EMG) recordings were used to evaluate urinary function in urethane-anesthetized rats. As a result, grafted BS-NPCs survived and integrated with the host spinal cord tissue. Differentiated excitatory or inhibitory neurons projected long axons to the caudal somatic and autonomic regions. The injection of biotin dextran amine (BDA) into the pontine micturition center (PMC) anterogradely labeled axon terminals regenerated into the graft. Transsynaptic tracing with pseudorabies virus (PRV) inoculation into the bladder detrusor revealed infected neurons in both cellular grafts and the host brainstem. Micturition function assessments suggest that the graft mitigates the severity of bladder hyperreflexia to achieve better coordination between the detrusor and sphincter. Ongoing studies are testing spontaneous micturition using metabolic cages. The results indicate that transplanting BS-NPCs may rebuild supraspinal regulation to improve micturition function after SCI.

Disclosures: K. Patel: None. **E. Oatman:** None. **S. Hou:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.08

Topic: C.11. Spinal Cord Injury and Plasticity

Title: Rebuilding supraspinal regulation of sympathetic input to improve cardio-electric disorders after spinal cord injury

Authors: *M. CUSIMANO[1], S. FERNANDES[2];
[1]Drexel Univ., philadelphia, PA; [2]Drexel Univ., Philadelphia, PA

Abstract: Rebuilding supraspinal regulation of sympathetic input to improve cardio-electric disorders after spinal cord injury
Silvia Fernandes, Emily Oatman, Marissa Cusimano, Cameron Trueblood, Shaoping HouMarion Murray Spinal Cord Research Center, Department of Neurobiology and Anatomy, Drexel University College of Medicine, Philadelphia, PA 19129, United States
High-level spinal cord injury (SCI) often interrupts supraspinal regulation of sympathetic activity

to the heart. The loss of balance between autonomic components renders cardiac disorders such as arrhythmias. It was previously reported that disruption of serotonergic (5-HT) control of sympathetic flow mainly contributes to cardiovascular dysfunction after SCI. To determine if transplanting developing 5-HT^+ neurons restore this compromised neuronal regulation to improve cardiac electrical conduction, embryonic day 14 (E14) raphe nuclei-derived neural progenitor cells (RN-NPCs) were transplanted into the lesion of a crushed rat spinal cord at the $2^{nd}/3^{rd}$ thoracic (T2/3) level. Animals receiving spinal cord-derived NPCs (SC-NPCs), injury alone or naïve served as controls. Ten weeks after cellular grafting, a radio-telemetric system was used to record electrocardiogram (ECG) and blood pressure, including 24-h recording for heart rate variability (HRV), cardiac arrhythmias during colorectal distention (CRD)-induced autonomic dysreflexia, and dobutamine stress tests to mimic exercise. Consequently, transplanting either RN- or SC-NPCs significantly increased HRV which were illustrated by parameters in both time and frequency domains. Although the grafts did not alter the occurrence of various arrhythmias during CRD, both lessened the exacerbation of rhythmic symptoms when dobutamine was delivered to excite cardiac sympathetic limb. Histological analysis revealed that grafted NPCs survived, well-integrated with the host tissue, and projected numerous axons onto the caudal autonomic regions. Furthermore, ongoing study is to examine if inhibition of spinal 5-HT_{2A} receptors blocks ECG improvements in RN-NPC grafted rats. Ultimately, transplantation of NPC may re-establish supraspinal regulation of sympathetic input to enhance cardio-electric disorders following SCI. The cellular grafts may exert general neuronal effects rather than a specific role in the recovery.

Disclosures: M. Cusimano: None. **S. Fernandes:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.09

Topic: C.11. Spinal Cord Injury and Plasticity

Support: VA Merit Review Grant

Title: Transplanted neural progenitor cells modify chronic glial scar and enable host corticospinal tract regeneration and transplant derived axonal growthafter chronic spinal cord injury

Authors: *P. P. LU[1,2], A. TRAN[1], A. VILLARTA[1], L. GRAHAM[1], M. TUSZYNSKI[1,2]; [1]UCSD Dept. of Neurosciences, UCSD Dept. of Neurosciences, La Jolla, CA; [2]VA-San Diego Healthcare Syst., San Diego, CA

Abstract: The chronic border of astrocyte reactivity around sites of spinal cord injury is often considered a barrier to axon growth and regeneration, and particularly after chronic SCI. We examined the association of chronically injured corticospinal axons with reactive astrocyte

borders 6 months after SCI. Further, we explored whether a graft of spinal cord neural progenitor cells placed 5 months after SCI altered chronic astrocyte reactivity. 18 rats underwent C6 bilateral contusive SCI; 5 months later, half of the animals received E13-derived spinal cord neural progenitor cell grafts and the corticospinal tract was anterogradely traced. **Results:**. In control lesioned animals, we found that even 6 months after injury, injured corticospinal axons were closely apposed to the lesion borders (*not* retracted from the lesion), are were deeply embedded within zones of intense GFAP labeling. Animals that received grafts of neural progenitor cells after 5 months exhibited a marked reduction of GFAP reactivity when examined one month later. Host corticospinal axons regenerated into neural progenitor cell grafts, and graft-derived axons extended into the distal host spinal cord. These results indicate that: 1) chronically injured host axons do not necessarily undergo retraction from sites of injury, 2) that neural progenitor cell grafts attenuate chronic astrocyte reactivity, and 3) chronically injured axons regenerate into neural progenitor cell grafts. These findings suggest possible avenues for treating chronic SCI. Supported by the Veterans Administration.

Disclosures: P.P. Lu: None. **A. Tran:** None. **A. Villarta:** None. **L. Graham:** None. **M. Tuszynski:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.10

Topic: C.11. Spinal Cord Injury and Plasticity

Title: Therapeutic effects of combined hiPS-NS/PCs transplantation and rehabilitative training in chronic spinal cord injury

Authors: *T. SHIBATA[1], S. TASHIRO[2], N. NAGOSHI[1], M. SHINOZAKI[3], M. KAWAI[1], S. HASHIMOTO[1], H. OKANO[3], M. NAKAMURA[1];
[1]Keio Univ. Sch. of Med., Dept. of Orthopaedic Surgery, Keio Univ. Sch. of Med., Tokyo, Japan; [2]Keio Univ. Sch. of Med., Dept. of Rehabil. Medicine, Keio Univ. Sch. of Med., Tokyo, Japan; [3]Keio Univ. Sch. of Med., Dept. of Physiology, Keio Univ. Sch. of Med., Tokyo, Japan

Abstract: Introduction
Cell transplantation therapy using human-induced pluripotent stem cell-derived neural stem/progenitor cells (hiPSC-NS/PCs) is a new therapeutic strategy for spinal cord injury (SCI). The efficacy of hiPSC-NS/PCs transplantation was reported by a number of preclinical studies which targeted sub-acute phase as a timing of therapeutic intervention, but no functional recovery was observed when transplanted at chronic phase. Rehabilitative training should be considered as an important strategy for recovering motor function after SCI. However, rehabilitative training was known to be less effective when applied in the chronic phase. A combined therapy with hiPSC-NS/PCs transplantation and rehabilitative training is attracting attention as a therapeutic option for chronic SCI because they could produce synergistic effects.

In this study, we investigated the therapeutic effect of the combined therapy of hiPSC-NS/PCs transplantation and rehabilitative training.
Method
Contusive SCI was induced in NOD-SCID mice, and hiPSC-NS/PCs were transplanted into the injured spinal cord of mice at 49 days post injury. The animals were divided into the treadmill training (TP+TMT) group and the non-treadmill training (TP) group. After transplantation, the TP+TMT group was subjected to treadmill training based on the overload principles for 8 weeks. Hindlimb locomotor function of each animal was evaluated weekly using the Basso Mouse Scale (BMS) scores up to 105 days post injury. Quadrupedal gait analysis were also performed before sacrificing the animals. Their spinal cords were removed and used for histological analyses or protein quantifications by capillary electrophoresis.
Results
The survival rate of grafted hiPSC-NS/PCs was significantly larger in the TP+TMT group than in the TP group. Moreover, the cell differentiation assay of grafted hiPSC-NS/PCs revealed that the proportion of NeuN positive neurons were significantly higher in the TP+TMT group than in the TP group, and the axons of the engrafted neural cells extended widely from epicenter to rostral and caudal. At the lumber spinal cord, Syn1 positive area and 5HT positive fibers were increased in the TP+TMT group. Capillary electrophoresis revealed that expressions of BDNF and NT3 proteins in spinal cord tissue were significantly enhanced in the TP+TMT group.The BMS scores and quadrupedal gait analysis indicated significantly better recovery in the TP+TMT group than in the TP group.
Conclusion
A combined therapy of hiPSC-NS/PCs transplantation and rehabilitative training has the potential to promote functional recovery even when starting this intervention at chronic spinal cord injury.

Disclosures: **T. Shibata:** None. **S. Tashiro:** None. **N. Nagoshi:** None. **M. Shinozaki:** None. **M. Kawai:** None. **S. Hashimoto:** None. **H. Okano:** None. **M. Nakamura:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.11

Topic: C.11. Spinal Cord Injury and Plasticity

Support: Novo Nordisk Foundation Laureate grant
The Independent Research Fund Denmark
The Innovation Fund Denmark

Title: Circuit-targeted biological repair strategy to restore function after spinal cord injury

Authors: *I. LISA VARGAS[1,2], J. NICLIS[2], C. BELLARDITA[1], O. KIEHN[1,3];
[1]Dept. of Neurosci., Univ. of Copenhagen, Copenhagen, Denmark; [2]Novo Nordisk A/S, Bagsværd, Denmark; [3]Dept. of Neurosci., Karolinska Inst., Stockholm, Sweden

Abstract: Spinal cord injury (SCI) often leads to reduced movement or paralysis and abnormal activity of the spinal circuits, referred to as spasticity. Recovery of movement after SCI is limited due to neuronal damage that results in neuronal signaling conduction failure across the lesion site. Cell replacement therapies are a promising approach to restore lost connectivity and re-establish motor control. On this line, we investigated the role of transplanted spinal neurons, including excitatory V2a neurons, to act as neuronal circuit-bridges across the lesion to relay signals from supraspinal areas to the spinal cord.
We grafted mouse embryonic day 11.5 neural stem cells (NSCs) into the spinal cord of mice with sacral injuries. To characterize graft size, we transplanted NSCs obtained from transgenic HoxB8^{Cre}::R26YFP embryos that fluorescently label all spinal cells below C4. Fluorescent images of the graft 4 and 16 weeks post-transplantation showed there is a time-dependent increase in the integration of the graft within host tissue. To evaluate the potential role of V2a neurons in the recovery of movement, we also transplanted NSCs from transgenic Chx10^{Cre}::R26ChR2YFP embryos that specifically mark V2a interneurons. 16 weeks post-transplantation, grafted V2a neurons extended long axons into the host tissue caudal to the injury site where they formed functional synapses. Unilateral optogenetic activation of grafted V2a neurons 16 weeks post-transplantation resulted in tail movement with ipsilateral excitatory to the stimulation and contralateral inhibition in the inhibition of spontaneous muscle activity on the contralateral site.
In conclusion, this study provides evidence at the anatomical, functional and behavioral levels of a functional integration of the grafted cells in the injured spinal cord. Specifically, excitatory V2a neurons form microcircuits with physiological features, including excitatory and inhibitory functional connections to host motor neurons that results in restoration of movements. Mechanisms-based repair strategies hold promise to restore spinal microcircuit function to enable recovery of movement.

Disclosures: I. Lisa Vargas: None. **J. Niclis:** None. **C. Bellardita:** None. **O. Kiehn:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.12

Topic: C.11. Spinal Cord Injury and Plasticity

Support: VA Gordon Mansfield Spinal Cord Injury Collaborative Consortium
IP50RX001045 RR&D B7332R
NIH R01-NS042291
NIH R01-NS104442
NIH NCRR P51 OD011107-56

DOD CDMRP SC170233
DARPA
Dr. Miriam and Sheldon G. Adelson Medical Research Foundation

Title: Grafts of human spinal cord neural stem cells into primate hemisection and contusion spinal cord injury improve functional outcomes after 3 months

Authors: *E. S. ROSENZWEIG[1], E. SINOPOULOU[1], J. BROCK[1,2], P. LU[1,2], H. KUMAMARU[1], J. WEBER[1], S. HAWBECKER[3], C. MANNION[3], J. HYDE[3], R. WURR[3], R. MACON[3], R. HUIE[4], A. TORRES-ESPIN[4], N. KYRITSIS[4], C. SPARREY[5], Y. NOUT-LOMAS[6], L. HAVTON[7], A. FERGUSON[4,8], M. BEATTIE[4,8], J. BRESNAHAN[4,8], M. TUSZYNSKI[1,2];
[1]Dept. of Neurosciences, Univ. of California San Diego, La Jolla, CA; [2]Veteran's Affairs Med. Ctr., La Jolla, CA; [3]California Natl. Primate Res. Ctr., Davis, CA; [4]Dept. of Neurosurg., Univ. of California San Francisco, San Francisco, CA; [5]Dept. of Mechatronic Systems Engin., Simon Fraser Univ., Surrey, BC, Canada; [6]Col. of Vet. Med. and Biomed. Sci., Colorado State Univ., Fort Collins, CO; [7]Depts. of Neurol. and Neurosci., Icahn Sch. of Med. at Mount Sinai, New York, NY; [8]Veteran's Affairs Med. Center, San Francisco, San Francisco, CA

Abstract: We previously demonstrated that human neural stem cells (hNSCs) or multipotent neural progenitor cells (hNPCs) grafted into sites of rodent or rhesus primate spinal cord injury (SCI) survive, extend axons, form synapses, support host axon regeneration, and improve functional recovery (Lu et al., *Cell* 2012; Lu et al., *Neuron* 2014; Kadoya et al., *Nat Med* 2016; Rosenzweig et al., *Nat Med* 2018). To enable translation to human clinical trials, we continue to develop this approach, specifically addressing cell source issues and the injury model's clinical relevance.
First, we have developed a human-embryonic-stem-cell-derived Neural Stem Cell line driven to a *spinal cord* identity (H9-scNSC; Kumamaru et al., *Nat Methods* 2018) as a candidate cell type for human translation. Second, we have grafted this candidate cell type into rhesus monkeys two weeks after a C7 unilateral spinal cord hemisection (lesion model described in Rosenzweig et al., *Nat Neurosci* 2010). Third, we have grafted this candidate cell type into rhesus monkeys four weeks after a more clinically-relevant C6 unilateral spinal cord contusion (lesion model described in Salegio et al., *J Neurotrauma* 2016).
We find that: 1) H9-scNSC grafts placed into sites of C7 hemisection or C6 unilateral contusion SCI survive, extend axons, form synapses, and support host axon regeneration into the stem cell graft. 2) Successful H9-scNSC grafts are associated with significant functional improvement in both hemisection and hemicontusion models 3 months after injury. 3) Principal Components Analysis showed strong correlations between the extent of functional recovery and the extent of H9-scNSC graft survival, integration, and growth.

Disclosures: E.S. Rosenzweig: None. **E. sinopoulou:** None. **J. brock:** None. **P. lu:** None. **H. kumamaru:** None. **J. weber:** None. **S. hawbecker:** None. **C. mannion:** None. **J. hyde:** None. **R. wurr:** None. **R. macon:** None. **R. Huie:** None. **A. torres-espin:** None. **N. Kyritsis:** None. **C. sparrey:** None. **Y. Nout-Lomas:** None. **L. havton:** None. **A. ferguson:** None. **M. beattie:** None. **J. bresnahan:** None. **M. tuszynski:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.13

Topic: C.11. Spinal Cord Injury and Plasticity

Support: CIRM TRAN1-11579 TUSZYNSKI 183C4A

Title: Progress in production of large scale GMP neural stem cell banks for clinical trials in spinal cord injury

Authors: *T. VOKES[1], J. BROCK[1,2], A. BLESCH[1,2], T. LAMOUR[3], H. YOUNG[3], M. TUSZNYSKI[1,2];
[1]Dept. of Neurosciences, Univ. of California – San Diego, La Jolla, CA; [2]Veterans Admin. Med. Ctr. – San Diego, La Jolla, CA; [3]Advanced Cell Therapy Laboratory, Univ. of California – San Diego, La Jolla, CA

Abstract: We have developed a lead candidate cell line for translation to human clinical trials in spinal cord injury (SCI): H9 embryonic stem cells (ESCs) driven to a spinal cord fate ("H9-scNSCs"). These H9-scNSCs partially restore function in rodent and non-human primate models of SCI by providing cellular relays across the lesion site. We previously optimized derivation of spinal cord NSCs from H9 ESC lines using small molecule-based cellular reprogramming (Kumamaru et al, Nature Methods 2018). To support clinical translation, research-grade cells and processes must be scaled up in a GMP-compliant format to satisfy FDA and scientific standards, including reproducibility, efficacy, and safety. We have now successfully transferred our cell reprogramming process to a GMP-compliant clinical laboratory at UCSD, the Advanced Cell Therapy Laboratory (ACTL). Four different production runs of cell banks at ACTL using GMP-compliant methods have consistently met our genotypic and phenotypic criteria from cell production. These GMP-compliant H9-scNSC cell banks mirror our previously generated cell lines, exhibiting increased expression of SOX1+ and PAX6+ cells, reduced expression of pluripotency markers OCT4 and NANOG and mesodermal markers Brachyury and TBX6. We also show increased expression of HOXC6, indicating a cell fate below the C2-3 level. The phenotype has been confirmed with flow cytometry indicating increased expression of neural lineage surface markers, and absence of contaminating cell types expressing pluripotent and mesodermal surface markers. The surface marker expression indicates an NSC population that is heterogeneous for both early stage NSCs and later stage neural progenitor cells (NPCs). These cells have the capacity to differentiate into mature neurons, shown by the presence of MAP2 and Tau+ cells. With this heterogeneous population of scNSC, we hypothesize that these cells can differentiate into multiple cell types of a spinalized neural lineage after grafting into a spinal cord lesion cavity to form a neural-supportive environment. We demonstrate a highly reproducible cell manufacturing strategy that is appropriate for scale up to provide a consistent and abundant source of cells to be used in a first-in-human clinical trial for spinal cord injury.

Disclosures: T. Vokes: None. **J. Brock:** None. **A. Blesch:** None. **T. Lamour:** None. **H. Young:** None. **M. Tusznyski:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.14

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH-5R01EB005678-10
Dr. Lorna Carlin Scholar Award

Title: Combination of biomaterial bridge implantation and interleukin-10 expression to modulate the inflammatory response and promote neural regeneration and connectivity after spinal cord injury

Authors: *U. NEKANTI[1], A. ZAHEDI[1], P. SAKTHIVEL[1], R. NISHI[1], Z. GUO[1], X. LIN[1], N. HERNANDEZ[1], X. CHEN[2], C. M. DUMONT[3], J. MARTINEZ[1], B. J. CUMMINGS[1], L. SHEA[4], X. XU[1], A. J. ANDERSON[1];
[1]Univ. Of California, Irvine - Interdepartmental Neurosci. Program, IRVINE, CA; [2]Univ. of California, Riverside, Riverside, CA; [3]Univ. of Miami, Coral Gables, FL; [4]Univ. of Michigan, Ann Arbor, MI

Abstract: Spinal cord injury (SCI) is caused by an initial trauma that leads to immune cell infiltration and the initiation of an inflammatory cascade that mediates axon degeneration, demyelination, neural cell loss including apoptosis of oligodendrocytes, and the formation of inhibitory barriers for regeneration. Macrophages are the key players in the neuroimmune niche following SCI, playing divergent roles based on their surrounding stimuli. Anti-inflammatory cytokines such as interleukin-10 (IL-10) can induce neuroprotective and pro-regenerative activation states in macrophages. In this study, we investigated the effect of localized lentiviral expression of IL-10 with or without multi-channel poly (lactide-co-glycolide) (PLG) bridge implantation on the outcome after spinal cord injury. Combinatorial IL-10 delivery and PLG bridge implantation resulted in a synergistic improvement in ipsilateral paw function. Transsynaptic pseudorabies virus (PRV) retrograde tracing demonstrated that axons regenerate through the PLG bridge and form a synaptic relay between corticospinal neurons as well as the paraventricular nucleus of the thalamus and the neuromuscular junction, and that the number of connected neurons in the PVN was increased by combined treatment with IL-10 and Bridge implant. Our findings support the combination of PLG bridge implantation and IL-10 expression as a novel therapeutic strategy for modulating neuroinflammation and promoting motor recovery after SCI by reestablishing the damaged circuitry.

Disclosures: U. Nekanti: None. **A. Zahedi:** None. **P. Sakthivel:** None. **R. Nishi:** None. **Z. Guo:** None. **X. Lin:** None. **N. Hernandez:** None. **X. Chen:** None. **C.M. Dumont:** None. **J. Martinez:** None. **B.J. Cummings:** None. **L. Shea:** None. **X. Xu:** None. **A.J. Anderson:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.15

Topic: C.11. Spinal Cord Injury and Plasticity

Support: 2021R1A2C2006110
2021M3E5D9021364
2019R1A5A2026045

Title: Mechanical stiffness of hydrogel determines cellular adhesion in vitro and survival of neural stem cell grafts in the injured spinal cord

Authors: *H. PARK[1], Y.-K. KIM[2], S. GENISCAN[1], D. HWANG[1], H. KIM[1], S. SONG[2], B. KIM[1,3];
[1]Dept. of Brain Science, Dept. of Biomed. Sciences, Ajou Univ., Suwon, Korea, Republic of; [2]Ctr. for Biomaterials, Korea Inst. of Sci. and Technology, Seoul, 02792, Republic of Korea., Seoul, Korea, Republic of; [3]Dept. of Neurology, Ajou Univ. Sch. of Med., Suwon, Korea, Republic of

Abstract: The therapeutic potential of cell transplantation for spinal cord injury is frequently compromised by poor survival of grafted cells. Biomaterial-based scaffolds may support cellular engraftment in the inhospitable microenvironment of the injured spinal cord. We have previously demonstrated that injection of a thermosensitive injectable hydrogel can create an extracellular matrix (ECM) in the lesion epicenter that would otherwise become cystic cavities. We expected that space-filling effects of the hydrogel would be highly conducive to graft survival. However, neural stem cells (NSCs) delivered as a complex with the hydrogel barely survived the transplantation with frequent graft failures. Accumulating evidence suggests that mechanical stimuli transmitted to cell membranes can significantly affect cellular behaviors. Therefore, we hypothesized that hydrogel mechanical stiffness may be a critical factor to regulate the survival of NSCs transplanted within the hydrogel complex. We first established an *in vitro* culture system where E14 spinal cord derived NSCs were grown on hydrogel substrates with different stiffness ranging from 0.2 to 25 kPa. NSCs grown on rigid substrates showed an increase in the spreading area and cellular perimeter, indicating an improvement in cellular adhesion. This was accompanied by a significant increase and decrease in the percentage of living and dying cells, respectively. We found that NSCs express several mechanosensitive ion channels, which allow calcium currents in response to mechanical stress. The extent of calcium oscillations was markedly increased in NSCs grown on 25kPa hydrogel compared to those on 0.2 kPa. Pharmacological inhibition of mechanosensitive channels using GsMTx4 significantly attenuated the stiffness-dependent improvement in NSC adhesion and survival *in vitro*. Finally, transplantation of NSCs with a varying percentage of hydrogel showed a concentration-dependent increase in the areas of NSC grafts, demonstrating that modulation of hydrogel mechanical stiffness can improve the survival of transplanted NSCs.

Disclosures: **H. Park:** None. **Y. Kim:** None. **S. Geniscan:** None. **D. Hwang:** None. **H. Kim:** None. **S. Song:** None. **B. Kim:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.16

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH-R01 NS119337 (AGR & SPP)
DoD W81XWH2010347 (AGR)
SCoBIRC Chair Endowments (AGR & PGS)

Title: Delivering mitochondria to the spinal cord via engineered erodible hydrogels

Authors: *S. P. PATEL[1], F. M. MICHAEL[1], J. THARAPPEL[1], H. M. VAUGHT[1], M. KHAN[2], P. G. SULLIVAN[3], J. E. DEROUCHEY[4], T. D. DZIUBLA[2], A. G. RABCHEVSKY[1];
[1]Spinal cord & Brain Injury research Center, Dept. of Physiol., [2]Dept. of Chem. and Materials Engin., [3]Spinal cord & Brain Injury research Center, Dept. of Neurosci., [4]Dept. of Chem., Univ. of Kentucky, Lexington, KY

Abstract: Mitochondrial dysfunction following spinal cord injury (SCI) is attributed to increased oxidative stress and inflammation that lead to irreparable functional deficits. Acute mitochondrial transplantation via intraspinal injections improves mitochondrial bioenergetics of injured tissues 24 hr post-injury, but not behavioral measures. Since technical limitations of invasive delivery lead to accumulation at injection sites that rapidly reduces mitochondrial integrity ex vivo, we pursued a subdural delivery route utilizing thermogelling erodible hydrogels that can be fabricated to degrade at a controlled rate in vivo to deliver healthy mitochondria at and around the injury site. We first established that hydrogel composed of methylcellulose (1%) and hyaluronic acid (1%) releases ~80% of mitochondria by 1 hr and preserves their integrity at 37°C. To track mitochondrial uptake into host cells, we transplanted either Mitotracker Red (MTR)-labeled mitochondria or genetically modified mitochondria tagged with red fluorescence protein (RFP) derived from SH-SY5Y human neuroblastoma cells. Results showed that following transplantation, MTR dye leaks from transplanted mitochondria and labels host mitochondria non-specifically. Alternatively, RFP-tagged transgenic mitochondria were taken up into both rat PC-12 and human SH-SY5Y cells by 2 hr, with time-dependent increases in uptake up to 24 hr, which was confirmed by presence of dose-dependent human mitochondrial DNA (mtDNA) in PC-12 cells. Compared to intraspinal injections, intrathecal delivery of RFP-tagged mitochondrial within hydrogels showed widespread cellular incorporation with a lack of host parenchyma glial cell or brain macrophage activation (OX-42+) at the delivery sites. Notably, human mtDNA was detected 24 hr after intraspinal or subdural mitochondrial transplantation in both naïve and injured spinal cords. In summary, subdural delivery of mitochondria within erodible hydrogels is effective in localized rostro-caudal

dispersion and host cellular uptake. Ongoing experiments are assessing the dose- and time-dependent effects of intraspinal vs subdural mitochondrial delivery on cellular bioenergetics and cell-specific internalization.

Disclosures: S.P. Patel: None. **F.M. Michael:** None. **J. Tharappel:** None. **H.M. Vaught:** None. **M. Khan:** None. **P.G. Sullivan:** None. **J.E. DeRouchey:** None. **T.D. Dziubla:** None. **A.G. Rabchevsky:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.17

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Grant 1R01NS115025-01A1
Craig H. Neilson 476694
Morton Cure Paralysis Fund
Craig H. Neilsen 296453
Center for Neurotechnology, University of Washington

Title: Optogenetic stimulation of the rat cervical spinal cord promotes enhanced functional recovery, axonal growth, and angiogenesis

Authors: *S. E. MONDELLO[1], V. DANG[1], N. M. TOLLEY[1], L. YOUNG[1], A. E. FISCHEDICK[1], T. WANG[1], M. A. BRAVO[1], D. LEE[1], B. TUCKER[1], B. D. PEDIGO[1], P. J. HORNER[6], C. T. MORITZ[1,2,3,4,5];
[1]Dept. of Rehabil. Med., [2]Inst. for Neuroengineering, [3]Dept. of Physiol. and Biophysics, [4]Grad. Program in Neurosci., [5]Ctr. for Neurotechnology, Univ. of Washington, Seattle, WA; [6]Dept. of Neurolog. Surgery, Ctr. for Neuroregeneration, Houston, TX

Abstract: Spinal cord injury (SCI) leads to debilitating sensory and motor deficits that greatly limit patients' quality of life. This work aims to develop a mechanistic understanding of how to best promote recovery following SCI to create future treatments. Electrical spinal stimulation is one promising approach to promoting recovery in both animal models and humans with SCI. Optogenetic stimulation is an alternative method of stimulating the spinal cord that allows for targeted stimulation of specific cell types of interest. The present work investigates the effects of neuron-specific optogenetic spinal stimulation on forelimb recovery, axonal growth, and vasculature in rats after a cervical spinal cord injury. Adult rats received a moderate cervical hemicontusion (C4) followed by injection with an optogenetic viral vector (*AAV2-hSyn-ChR2-YFP*) ipsilateral and caudal to the lesion site at C6. Afterwards, rats began rehabilitation 5x/week on the skilled forelimb reaching task. At 4 weeks post-injury, rats received a microLED implant to illuminate the C6 spinal cord to deliver neuron-specific optogenetic stimulation that excludes direct glial activation. Stimulation began at 6 weeks post-injury and occurred in conjunction with

activities to promote forelimb use. Following 6 weeks of stimulation, rats were perfused and tissue stained for GAP-43 (axonal growth), laminin (vasculature), and Cresyl violet and myelin (lesion magnitude). The location of viral transduction and transduced cell types was also assessed. Neuron-specific optogenetic spinal stimulation significantly enhanced recovery of skilled forelimb reaching after SCI. As expected, the original lesion magnitude greatly affected the recovery level achieved by optogenetic stimulation. We also found significantly greater GAP-43 labeling at the stimulation site and at the lesioned segments following optogenetic stimulation, indicating enhanced axonal growth in those regions. Laminin staining indicated that vasculature was significantly enhanced throughout the cervical spinal cord following stimulation, suggesting that optogenetic stimulation promotes angiogenesis. Viral transduction of opsins that enable optogenetic stimulation occurred within an evenly mixed population of glutamatergic and GABAergic synapses. The findings from this study indicate optogenetic spinal stimulation improves forelimb reaching behavior, axonal growth, and angiogenesis following SCI. Future studies will utilize these mechanisms to develop the next generation of spinal cord injury therapies to promote recovery of function after injury.

Disclosures: S.E. Mondello: None. **V. Dang:** None. **N.M. Tolley:** None. **L. Young:** None. **A.E. Fischedick:** None. **T. Wang:** None. **M.A. Bravo:** None. **D. Lee:** None. **B. Tucker:** None. **B.D. Pedigo:** None. **P.J. Horner:** None. **C.T. Moritz:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.18

Topic: C.11. Spinal Cord Injury and Plasticity

Support: R01NS107807

Title: Chronic chemogenetic activation of corticospinal tract neurons improves neither axon sprouting nor behavioral recovery after pyramidotomy injury

Authors: *Z. WANG, M. BRANNIGAN, L. FRIEDRICH, N. CHHIKARA, N. SAMBERG, I. ANTOLAK, M. BLACKMORE;
Marquette Univ., Marquette Univ., Milwaukee, WI

Abstract: Chemogenetic tools like Designer Receptors Exclusively Activated by Designer Drugs (DREADDs) offer a means to continually modulate the excitability of specific sets of neurons. Interestingly, prior work in sensory neurons showed that both chemogenetic activation and silencing resulted in increased growth from injured axons, indicating a potentially complex relationship between neural activity and axon growth. Here, we tested the effect of chronic chemogenetic stimulation on corticospinal tract (CST) sprouting and on forelimb function. To specifically target CST neurons, AAV2-retro- DIO-hM3Dq-mCherry or matched mCherry control was injected to the cervical spinal cord of adult Emx-Cre transgenic mice. Pilot studies

verified selective expression in CST neurons and found that both IP injections of CNO and administration of clozapine resulted in long term elevation of neural activity in CST neuron as assessed by cFos immunohistochemistry. In subsequent experiments, mice received viral injections, were pre-trained on a pellet retrieval task, and then received unilateral pyramidotomy injury to selectively ablate the right CST. Mice then received continual clozapine via drinking water and weekly testing on the pellet retrieval task, followed by cortical injection of EGFP tracer to assess cross-midline sprouting by the spared CST. Immunohistochemistry for cFos verified elevated CST activity in hM3Dq-treated animals at the time of sacrifice, eight weeks post-injury, and PKCy immunohistochemistry verified unilateral ablation of the CST in all animals. Despite the chronic elevation in CST activity in hM3Dq-treated animals, however, both groups showed similar levels of cross-midline CST sprouting and similar success in the pellet retrieval task. These data indicate that long term elevation of activity in CST neurons does not affect compensatory sprouting or directed forelimb movements. Ongoing work is testing the impact of Gi-DREADD activation on axon regeneration and functional recovery after pyramidotomy.

Disclosures: Z. Wang: None. **M. Brannigan:** None. **L. Friedrich:** None. **N. Chhikara:** None. **N. Samberg:** None. **I. Antolak:** None. **M. Blackmore:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.19

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH NINDS 5R01NS064004
Craig H Neilsen Foundation 547040

Title: Activity-dependent neuromodulation ameliorates transneuronal premotor interneuron degeneration after a bilateral corticospinal tract lesion

Authors: *J. L. PATHAN[1,2], T. BETHEA[1,2], J. H. MARTIN[1,2];
[1]Dept. of Molecular, Cellular, and Biomed. Sci., City Univ. of New York Sch. of Med., New York, NY; [2]Neurosci. Program, Grad. Ctr. of the City Univ. of New York, New York, NY

Abstract: Producing skilled movements in humans and many animals requires the corticospinal tract (CST), the direct motor pathway connecting the cerebral cortex with the spinal cord. Spinal cord injury results in CST damage and leads to spinal interneuron degeneration, a phenomenon termed transneuronal degeneration (Jiang et al 2018; J Neurosci 2018). Complement protein C1q triggers this process and induces microglia, the innate immune cells of the CNS, to phagocytose non-apoptotic cholinergic (Pitx2) and glutaminergic (Chx10) spinal interneurons. These interneuron classes are essential to the spinal motor circuit as they receive direct CST inputs and synapse onto motor neurons to produce muscle contraction. Yet, it is unknown if CST neural

activity is necessary, or if descending brainstem pathways can provide sufficient neural activity to prevent transneuronal interneuron degeneration after CST injury. We hypothesize that indirect cortical-brainstem pathways can provide neural activity to prevent transneuronal degeneration, and that general neural excitability of the local spinal circuit can also maintain spinal interneuron survival. Using Designer Receptor Exclusively Activated by Designer Drugs (DREADDs) in C57Bl/6J mice to enhance neural activity, we examined the role of indirect cortical-brainstem pathways and local spinal motor circuitry in preventing transneuronal degeneration of 2 interneuron classes after a bilateral CST lesion (biPTX). Mice either underwent bilateral injections of excitatory DREADDs in motor cortex or cervical enlargement. At appropriate timings of transfection (motor cortex, 3 weeks; cervical enlargement, 2 weeks), all mice received biPTX. Clozapine nitric-oxide (CNO) was administered for 10 consecutive days beginning 24 hours post-surgery to activate DREADD^{+} neurons. We observed biPTX induces significant degeneration of Pitx2 and Chx10 interneurons ($p=0.0001$), while significantly increasing phagocytic microglia density ($p<0.0001$). For both motor cortex and spinal cord DREADD transfection, we observed combined biPTX and CNO treatment significantly reduced Pitx2 and Chx10 interneuron degeneration and decreased phagocytic microglia density ($p<0.0001$) compared to that of the biPTX-only group. On-going experiments assess grip strength for motor recovery and serum cytokines for markers of inflammation among experimental groups. Pitx2 and Chx10 mouse Cre lines will evaluate cell-specific DREADD activation. Together, our results support our hypothesis and imply that excitatory DREADD neuromodulation ameliorates transneuronal degeneration and modulates the innate immune system after CST injury.

Disclosures: **J.L. Pathan:** None. **T. Bethea:** None. **J.H. Martin:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.20

Topic: C.11. Spinal Cord Injury and Plasticity

Support: Mission Connect, TIRR Foundation
NIH Grant R01NS116404
Craig H. Neilsen Foundation

Title: Acute chemogenetic silencing of nociceptive signaling to promote functional recovery following spinal cord injury

Authors: *P. AMAR KUMAR[1], J. STALLMAN[1], E. KERIM[1], J. HOPPE[1], Y. KHARBAT[1], A. LEONARDS[1], B. NGUYEN[1], S. LETCHUMAN[1], J. DULIN[1,2];
[1]Biol., [2]Texas A&M Inst. for Neurosci., Texas A&M Univ., College Station, TX

Abstract: Spinal cord injury (SCI) typically results in an immediate loss of neurological function due to massive disruption of endogenous neural circuitry. One of the debilitating

consequences of SCI that affects more than two-thirds of individuals living with SCI is the development of neuropathic pain. Previous work in rodent SCI pain models has established that maladaptive hyperactivity within primary nociceptors of the dorsal root ganglion (DRG) occurs as early as 24 hours after SCI, contributing to the onset of neuropathic pain. This not only results in sensory dysfunction but also undermines locomotor recovery. We hypothesized that silencing the activity of nociceptors early after injury will improve long-term functional outcomes. To test this hypothesis, we utilized inhibitory Gi-DREADDs (Designer Receptors Exclusively Activated by Designer Drugs) to selectively silence nociceptors during the acute phase of SCI in adult female Sprague Dawley rats. We delivered AAV6-Gi-DREADD to lumbar DRG nociceptors through bilateral intrasciatic injections, then performed thoracic contusion SCI 4 weeks later. Immediately following SCI and continuing for 14 days post-injury, Gi-DREADDs were activated through oral delivery of agonist clozapine-N-oxide (CNO). We performed sensory and motor behavioral assessments weekly up to 10 weeks post SCI. Gi-DREADD expression was restricted to small-diameter nociceptors including $CGRP^+$(Calcitonin Gene Related Peptide), substance P^+, and IB4-binding (Isolectin-B4) neurons. Through analysis of behavioral outcomes, we observed significantly higher thermal withdrawal thresholds, and greater hindlimb locomotor recovery in subjects that received acute nociceptor silencing, compared to controls. Histological assessments of spinal cord tissue suggest a trend showing reduced lesion volume and increased $CGRP^+$ axon sprouting in Gi-DREADD treated subjects compared to control animals. Together, these findings suggest that nociceptor silencing early after SCI may promote beneficial plasticity in the acute phase of injury that can impact long-term functional outcomes.

Disclosures: P. Amar Kumar: None. **J. Stallman:** None. **E. Kerim:** None. **J. Hoppe:** None. **Y. Kharbat:** None. **A. Leonards:** None. **B. Nguyen:** None. **S. Letchuman:** None. **J. Dulin:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.21

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH NIBIB DP 2-EB029757
NIH BRAIN Initiative R01NS123655-01
UG3NS123723-01

Title: A High-Channel Count, Wrap-Around Microelectrode Array for Focal Stimulation of the Rat Spinal Cord

Authors: *S. M. RUSSMAN[1], R. VATSYAYAN[2], H. U[3], S. A. DAYEH[2];
[1]Bioengineering, [2]Electrical and Computer Engin., [3]Neurosurg., UC San Diego, La Jolla, CA

Abstract: Spinal cord injury (SCI) can have dramatic, life-changing impact due to irreversible loss of motor, sensory, bowel, bladder, and sexual functions. The impact is so debilitating that even very small improvements in function lead to huge gains in quality of life. In the past decade, epidural spinal cord stimulation (SCS) with flexible microelectrode arrays has garnered interest as a possible route to functional restoration for SCI. Currently, epidural SCS employs a bulky paddle array that limits its utility. The stimulation is non-focal because the fatty dura dissipates part of the stimulation voltage and the cerebrospinal fluid in the subarachnoid space shunts current to other areas. These limitations are further amplified by the inability of such systems to operate near the ventral side of the spinal cord due their bulky size. Ventral access is important because it permits closer proximity to motor efferent fibers, thereby increasing efficiency of muscle stimulation. Given these limitations of epidural SCS, we aimed to develop a novel stimulation approach allowing for easy access to the ventral spinal cord. We hypothesized that a thin, high-channel count cuff-like microelectrode array would provide high-density coverage to all aspects of the spinal cord, including the ventral surface, which has not been accomplished with prior state of the art. First, we developed an electrode design and implantation technique that allows for consistent wrapping of the electrode around the spine. Next, we built a computational model of the rat spinal cord with the wrap-around electrode in place and identified optimal bipolar stimulation parameters. We stimulated the rat spinal cord circumferentially, varying the stimulation contacts in a semi-random order, and recorded EMG signals from four muscle groups in the lower limbs. We showed that stimulating the ventrolateral side of the spinal cord required lower activation thresholds to generate a muscle response and that our stimulation approach was able to activate muscle groups with high selectivity. We quantified our measurements by calculating muscle recruitment curves and muscle selectivity indices, relying on methods previously described in literature.In summary, we developed a novel, wrap-around microelectrode array to deliver focal stimulation to the rat spinal cord. In the long term, this stimulation approach holds the potential to allow patients to regain motor function. As for immediate impact, the wrap-around electrode will serve as a tool to understand spinal cord function by probing motor pathways of the spine in ways not previously attainable.

Disclosures: **S.M. Russman:** None. **R. Vatsyayan:** None. **H. U:** None. **S.A. Dayeh:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.22

Topic: C.11. Spinal Cord Injury and Plasticity

Support: OT2 OD025340
CH Neilsen Foundation
Paralyzed Veterans of America
KURe NIH K12

Title: Electroanatomical mapping of lumbosacral spinal cord stimulation responses in the external urethral sphincter and peripheral nerves of the lower urinary tract

Authors: *C. J. STEADMAN, A. J. TENNISON, C. L. LANGDALE, N. A. PELOT, W. M. GRILL;
Dept. of Biomed. Engin., Duke Univ., Durham, NC

Abstract: Spinal cord stimulation (SCS) can treat lower urinary tract (LUT) dysfunction in spinal cord injury (SCI) and other etiologies of neurogenic bladder dysfunction. Animal and clinical studies showed success in ameliorating LUT dysfunction, but significant gaps remain in our understanding of the mechanisms through which SCS modulates the LUT. The objective of this study was to use electrical stimulation to map the spatial distribution of locations over the lumbosacral spinal cord that evoked activity in the external urethral sphincter (EUS), as well as in the pudendal, pelvic, and sciatic nerves. Male (n=4) and female (n=6) spinally intact Sprague Dawley rats were implanted with cuff electrodes on the pudendal, pelvic, and sciatic nerves to record evoked neural activity (ENG), and a bipolar electrode was placed over the EUS to measure EMG. A high-density 15-channel electrode array with five rows of contacts was placed over the L4-S2 spinal cord for epidural SCS. We stimulated the spinal cord at amplitudes of 10-400 μA to measure EUS EMG responses. After gallamine administration, SCS was delivered at amplitudes of 10-1000 μA. We calculated stimulus-triggered averages for each SCS location and amplitude. Based on conduction velocities and distance from the stimulating electrode to the recording electrodes, we estimated the latencies for neural fiber types and used a mixed-effects model to determine relationships between stimulation location, stimulation amplitude, and activation of the EUS and peripheral nerves. Evoked EUS EMG and peripheral nerve ENG activity was observed between 2-60 ms after stimulation, increased in an amplitude-dependent manner, and was similar in both male and female rats. SCS over L4 evoked short latency EUS EMG activity (2-12 ms), while more caudal SCS (L6-S2) evoked both short latency (2-12 ms) and longer latency (12-30 ms) EUS responses. Rostral (L4) SCS with amplitudes between 10-200 μA activated Aβ fibers in the sciatic nerve. SCS over L6-S1 at amplitudes between 10-200 μA evoked robust responses of Aβ and Aδ fibers in the pudendal nerve, and, at amplitudes above 600 μA, weak responses of Aδ fibers in the pelvic nerve. The early EUS response to SCS over L4 suggests activation of descending efferent projections to the EUS, while SCS of L6-S2 evoked both direct and indirect EUS responses. These results provide insights into the mechanism of action of SCS effects on LUT function and enhance our understanding of SCS location and stimulation parameters when considering therapeutic design to improve LUT function.

Disclosures: C.J. Steadman: None. **A.J. Tennison:** None. **C.L. Langdale:** None. **N.A. Pelot:** None. **W.M. Grill:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.23

Topic: C.11. Spinal Cord Injury and Plasticity

Title: Low intensity theta burst ultrasound stimulation over the spinal cord attenuates neuropathic pain

Authors: *T. PHAN, K.-H. LEE, H. LEE, Y. KIM, J. PARK;
Inst. of Basic Sci., Inst. of Basic Sci., Daejeon, Korea, Republic of

Abstract: Therapeutic ultrasound has been indicated as a promising non-invasive treatment for pain management, yet various potential aspects and underlying mechanisms for neuropathic pain treatment still remain unclear. To investigate the therapeutic role of ultrasound stimulation targeting the spinal cord for neuropathic pain treatment, in this study, we developed a low-intensity continuous theta-burst ultrasound (cTBUS-200Hz)stimulation and accessed how it systematically modulates the behavioral responses as well as alters the spinal molecular level in partial crush injury (PCI) model. As a result, following 10 daily sessions of cTBUS-200Hztreatment, a significant increase in mechanical sensory thresholds was observed during and post-cTBUS-200Hztreatment, indicating a reduction of pain sensitivity. Consistent with previous studies demonstrating spinalBDNF as a key modulator in central sensitization and inflammatory pain, we also found an increment of BDNFprotein accumulation at the spinal L4-L5 lumbar sections in the peripheral injured groups, while it was significantly reduced in ultrasound-treated groups. Intriguingly, glial cells had been shown to contribute to the development and maintenance of neuropathic pain by controlling the spinal disinhibition and inducing hypersensitivity. We showed that low-intensity cTBUS-200Hz stimulation suppressed reactive astrocyte activity during chronic pain by reducing ambient GABA and increasing proBDNF levels [3], further electrophysiology studies are required to confirm whether ultrasonic stimulation can convert the switch ofGABAergic responses from being inhibitory to excitatory. As dorsal horn parvalbumin neurons activation after nerve injury alleviates mechanical pain, we additionally found the upregulation of activated dorsal horn parvalbumin neurons under low-intensity cTBUS-200Hz stimulation in the neuropathic pain model. Taken together, our results suggest that low-intensity continuous theta-burst ultrasound stimulation can efficiently attenuate the mechanical allodynia in neuropathic pain, thus contributing to the clinical application of ultrasonic technology in pain management. **Keywords**: Neuropathic pain, theta-burst stimulation, BDNF, reactive astrocytes, parvalbumin.

Disclosures: **T. Phan:** None. **K. Lee:** None. **H. Lee:** None. **Y. Kim:** None. **J. Park:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.24

Topic: C.11. Spinal Cord Injury and Plasticity

Support: DoD CDMRP W81XWH-18-1-0753
Craig H. Neilsen Foundation 546901
NIH R01 NS12119
F32 HD107806

Title: Ultrasound Imaging characteristics of tissue at risk for secondary damage in rodent spinal cord injury

Authors: *Z. Z. KHAING[1], M. F. BRUCE[2], J. S. HARMON[2], J. E. HYDE[1], L. N. CATES[1], C. P. HOFSTETTER[1];
[1]Neurolog. Surgery, [2]Applied Physics Lab., Univ. of Washington, Seattle, WA

Abstract: Traumatic spinal cord injury (tSCI) is characterized by a near complete loss of blood flow at the injury epicenter and substantial disturbance of blood flow in the penumbral zone, resulting in additional ischemia and cell death. The penumbral region is thought to expand with time after injury, resulting in a larger area of damaged tissue during the secondary phase of SCI. Indeed, the tissue at risk of damage and death during the secondary phase of the injury is the target for neuroprotective therapies. Here we examined the blood flow characteristics of this important penumbral region using innovative ultrafast contrast-enhanced ultrasound imaging (CEUS). Following a T7-T9 laminectomy to produce an acoustic window, CEUS imaging was acquired at baseline, and following a contusion SCI (150 kDyn) at acute (1 hour), 3, and 14 days post injury (dpi) timepoints. Intravital CEUS imaging showed a 3-fold increase in the area of perfusion deficit (1.62 ± 0.08 mm2 to 4.05 ± 0.28 mm2) of the contusion at 3 dpi, along with marked alterations in surrounding macrovascular (>75 um) morphology. Specifically, ventral central sulcal arteries (CSA) were displaced 19°± 1° degrees rostral to and 44°± 5° degrees caudal to the contusion, combined with an increase in tortuosity of ventral CSAs extending caudally from the lesion. Marked distortion of the dorsal macrovasculature (i.e. lengthening and bowing) was also observed at 3 dpi. We also performed standard histological analysis of the tissue collected to characterize the development of the lesion with time after the injury. Group analysis of the histological studies revealed that the lesion volume expanded from 0.47 ± 0.10 mm3 at the acute time-point to 4.41 ± 0.60 mm3 at 3 dpi, and increased further to 4.85 ± 0.56 mm3 by 14 dpi. Importantly, both intravital CEUS imaging and histology demonstrates that the majority of injury expansion occurs within 3 dpi. Additional histological and ultrasound imaging studies are currently underway to further characterize and evaluate the cellular and molecular changes within the tissue regions at risk of progressive damage identified by CEUS after SCI. Results obtained from this project have the potential to identify tissue at risk for secondary damage after an acute ischemic injury and to serve as an imaging biomarker for injury stratification for neuroprotective therapies.

Disclosures: Z.Z. Khaing: None. **M.F. Bruce:** None. **J.S. Harmon:** None. **J.E. Hyde:** None. **L.N. Cates:** None. **C.P. Hofstetter:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.25

Topic: C.11. Spinal Cord Injury and Plasticity

Support: New Frontiers Grant NFRFE-2021-00385
Horizons Grant DH-2022-00922

Title: Three-dimensional mapping of motor responses to spinal cord stimulation

Authors: *O. EDDAOUI, J. HARNIE, S. MARI, P. JEHANNIN, S. YASSINE, J. AUDET, C. LECOMPTE, F. SOUCY, R. ALARAB, A. FRIGON, C. IORIO-MORIN;
Sherbrooke Univ., Sherbrooke, QC, Canada

Abstract: Spinal cord stimulation (SCS) has recently emerged as a promising solution for functional rehabilitation following traumatic spinal cord injury (SCI). In fact, by adjusting electric pulse parameters and electrode position in relation to the spinal cord, epidural SCS has shown potential in restoring locomotion following complete SCI. However, to date, the biggest challenge remains to determine the optimal location of electrodes on the spinal cord to generate specific and selective muscle contractions, which considerably limits this technology. We hypothesize that ventral stimulation will directly activate lumbosacral motoneurons and achieve higher selectivity in muscle activations compared to the typical dorsal approach. While ventral and dorsal epidural SCS have been studied, there is limited evidence directly comparing the two approaches. The aim of this study is to rigorously map motor responses to both dorsal and ventral epidural SCS onto an accurate 3-dimensional representation of the spinal cord. We studied 4 cats with a spinal transection at T12-T13 and 5 spinal-intact cats. We obtained high-resolution axial 3T-MRI images of the lumbosacral spinal cord for each cat to build a personalized 3D model of their spinal cord. Then, under anesthesia, we installed a 120-electrode grid dorsally and multiple flexible electrodes ventrally to cover spinal segments from L2 to S1. Bipolar stimulations were sent to pairs of electrodes while gradually increasing the stimulation amplitude. For each stimulation, we recorded electromyographic (EMG) responses from 32 hindlimb muscles as well as leg movements from video cameras. Through a finite element analysis using personalized 3D spinal cord models and documented position of the electrodes, we calculated the current density distribution on the entire lumbosacral spinal cord following every stimulation to create maps of SCS motor responses. We could trigger selective muscle activations by varying stimulation electrode location and amplitude. These maps will help determine optimal electrode locations for restoring locomotion following traumatic SCI.

Disclosures: O. Eddaoui: None. **J. Harnie:** None. **S. Mari:** None. **P. Jehannin:** None. **S. Yassine:** None. **J. Audet:** None. **C. Lecompte:** None. **F. Soucy:** None. **R. Alarab:** None. **A. Frigon:** None. **C. Iorio-Morin:** None.

Poster

539. Promoting Spinal Cord Repair: Genes, Cells, Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 539.26

Topic: C.11. Spinal Cord Injury and Plasticity

Support: TCMF-EP 110-02

Title: Preliminary outcome of EES for incomplete SCI patients

Authors: *Y.-C. CHEN[1,2], M.-Y. WU[1,3], C.-T. DAI[1], S.-T. TSAI[1,4,5];
[1]Dept. of Neurosurg., Hualien Tzu Chi Hospital, Buddhist Tzu Chi Med. Fndn., Hualien, Taiwan; [2]Dept. of Med. Informatics, Tzu-Chi Univ., Hualien, Taiwan; [3]Dept. of Physical Therapy, [4]Dept. of Med., [5]Inst. of Med. Sci., Tzu Chi Univ., Hualien, Taiwan

Abstract: Motivation Spinal cord injury (SCI) causes devastating sensory and motor dysfunction and life-long sensorimotor impairment. Epidural electrical stimulation (EES) has been shown recovery of voluntary control of lower limb motor function for patients with American Spinal Injury Association (AIS) A or B. This study aimed to investigate whether EES improves lower limbs function for the cervical level of chronic SCI patients with AIS C or D as well. **Methods** Four cervical level chronic SCI patients with AIS C or D were enrolled (AIS C = 2, AIS D = 2), and the range of post-injury years was from 1.5 to 10 years, and the average age was 56.25 ±7 years at the time of implant. Three individuals revealed gait instability and walking with assistance and one could not stand or walk independently with minimal motor activity. They received an individualized rehabilitation program plus multi-mode electrical stimulation treatment for 36 weeks. This study used surface electromyography (EMG) to measure lower limb muscles and applied GAITRite walkway systems to evaluate gait. **Results** The improvement percentage of walking gait was more than 30% for patients with 1.5 to 3 injury years (35% - 92%). In contrast, the patient with 10 years SCI showed trivial improvement. Nevertheless, the analysis results of muscular activity indicated a significant increase ($P < 0.05$) under EES treatment. Moreover, one patient with AIS C improved to AIS D. **Conclusion** We observed that EES enables faster recovery of lower limb function for patients with C or D, but there were still some factors lead to variable improvement. We found that longer duration injury (injury year≧10 years) might cause EES to improve neuroplasticity obscurely in corticospinal motor circuitry. However, EES for activating spinal cord circuitry under EMG analysis all showed significant enhancement. We suggested that SCI patients with long-term injury might need combinational therapeutic approaches to facilitate effectiveness of EES and enhance corticospinal circuit motor plasticity.

Disclosures: Y. Chen: None. **M. Wu:** None. **C. Dai:** None. **S. Tsai:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.01

Topic: D.02. Somatosensation – Pain

Support: F32-DA053099
T32-MH014654
DP2-GM140923
T32-DA028874

Title: Nociceptive opioidergic circuits in the cingulate cortex

Authors: *N. M. MCCALL[1], B. C. REINER[1], R. CRIST[1], J. WOJICK[1], G. J. SALIMANDO[1], M. KINDEL[1], J. STUCYNSKI[1], M. KIM[1], C. RAMAKRISHNAN[2], K. DEISSEROTH[2], **G. F. CORDER**[1];
[1]Univ. of Pennsylvania, Univ. of Pennsylvania, Philadelphia, PA; [2]Stanford Univ., Stanford Univ., Palo Alto, CA

Abstract: Pain is an unpleasant emotional perception driven by the transformation of sensory neural signals into affective-cognitive information in cortical regions, including the anterior cingulate cortex (ACC). Opioid action in the ACC lessens aspects of the aversive quality of pain through mu opioid receptors (MOR). We hypothesized that MOR-expressing and functionally nociceptive ACC neurons represent a crucial neural circuit cell-type to pain affect and pathological attention to chronic pain. Using single nuclei RNA sequencing of murine ACC (n=20 adult male mice), we found three cell-subtypes of *Slc17a7* glutamatergic neurons were the most transcriptionally active to noxious stimuli. Transcriptional activity was identified by weighted expression of 139 immediate early genes (IEGs) and differential gene expression (DGE) analysis between nuclei from uninjured and chronic neuropathic pain mice. These cell-subtypes all co-expressed *Oprm1* along with single genetic identifiers of layer-specific ensembles in L2/3 IT, L5 ET, and L5 PT neurons, respectively. Gene ontology analysis revealed DGEs from neuropathic pain ACC nuclei were almost entirely related to synaptogenesis and plasticity mechanisms. To gain genetic access to ACC nociceptive-*Oprm1* ensembles, we developed an intersectional approach combining genetic elements of IEGs and *Oprm1* with retrograde viral recombinases. Thus, this approach drove expression of circuit mapping tools and optogenetic actuators with activity-, molecular-, and projection-dependency. To mimic opioid analgesia, we are starting studies in which ACC MOR+ nociceptive cell-types are inhibited with an iC++ opsin (on-going study, n=~25 adult male and female mice). Concurrently with this inhibition, pain-related behaviors are captured using a novel deep-learning system for unbiased pose-estimation of nocifensive behaviors. Our unpublished work will provide insight into the output projections of nociceptive ACC cell-types and subcortical input to this region. Notably, there is a possible reciprocal circuit between a subpopulation of medial basolateral amygdalar nociceptive cells and ACC L2/3 cell-types. Identifying the local cortical and brain-wide structure of the specific cell-type networks underlying opioid analgesia can aid the development of circuit-targeted treatments for pain with improved selectivity and reduced addiction liability.

Disclosures: N.M. McCall: None. **B.C. Reiner:** None. **R. Crist:** None. **J. Wojick:** None. **G.J. Salimando:** None. **M. Kindel:** None. **J. Stucynski:** None. **M. Kim:** None. **C. Ramakrishnan:** None. **K. Deisseroth:** None. **G.F. Corder:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.02

Topic: D.02. Somatosensation – Pain

Support: NIH/NIGMS DP2 1DP2GM140 923-01 (Corder)
5T32DA028874-11 (Salimando)

Title: The contribution of a parabrachial nucleus-to-central amygdala circuit to fentanyl withdrawal and hyperalgesia

Authors: *L. WOOLDRIDGE[1], G. J. SALIMANDO[2], C. RAMAKRISHNAN[3], K. T. BEIER[4], K. DEISSEROTH[3], G. F. CORDER[2];
[1]Univ. of Pennsylvania Neurosci. Grad. Group, Univ. of Pennsylvania Neurosci. Grad. Group, Philadelphia, PA; [2]Dept. of Psychiatry, Univ. of Pennsylvania, Philadelphia, PA; [3]Departments of Psychiatry and Behavioral Sci. and Bioengineering, Howard Hughes Med. Institute, Stanford Univ., Stanford, CA; [4]Univ. of California, Irvine, Irvine, CA

Abstract: Agonists for the mu opioid receptor (MOR), such as fentanyl, remain a frontline option for moderate to severe pain management. However, a range of highly aversive side effects limit their long-term clinical use, including dependence and withdrawal, as well as a paradoxical potentiation of pain known as opioid-induced hyperalgesia (OIH). Preclinical studies have associated neural hyperactivity in the capsular central amygdala (CeC) in opioid withdrawal and OIH. The molecular and functional cell-types in the CeC and the connected circuitries contributing to opioid withdrawal and OIH are not fully understood. Here, using a mouse model of fentanyl dependence, we characterized an opioid withdrawal-activated CeC population and tested its necessity to produce the behavioral correlates of withdrawal and OIH. To produce dependence, mice were given fentanyl (0.02 mg/mL) or untreated water (control) in their homecage drinking water supply for 8 days. By day 6, fentanyl-drinking mice increased mechanical and thermal hypersensitivity compared to both their pre-fentanyl baselines and to control mice, indicating the development of fentanyl-induced hyperalgesia. Furthermore, naloxone administration (1 mg/kg) on day 8 induced classic murine signs of withdrawal in fentanyl-drinking but not control mice. Fentanyl withdrawal also induced robust cFOS expression in the central amygdala, particularly in the anterior portion of the CeC, indicating that this region is highly active during opioid withdrawal. Fluorescent *in situ* hybridization (RNAScope) revealed substantial overlap between withdrawal-induced *Fos* and *Prkcd* mRNA, suggesting that the withdrawal-activated population overlaps with the pronociceptive CeC population expressing Protein Kinase C-delta (CeC^{PKCd}). The MOR-rich parabrachial nucleus (PBN) sends a strong excitatory projection to the CeC, to transmit ascending threat-related and nociceptive signals from the spinal cord. Monosynaptic input tracing with a modified rabies virus in PKCd-Cre mice suggested that MOR-expressing PBN neurons (PBN^{MOR}) send monosynaptic inputs to CeC^{PKCd} neurons. Collectively, these data indicate a PBN^{MOR}-CeC^{PKCd} circuit and suggest a role for the CeC^{PKCd} pronociceptive subpopulation in driving CeC hyperactivity during opioid withdrawal. Future studies will examine the the ability of PBN^{MOR} inputs to the CeC^{PKCd} population to drive opioid dependence- and OIH-related behaviors.

Disclosures: **L. Wooldridge:** None. **G.J. Salimando:** None. **C. Ramakrishnan:** None. **K.T. Beier:** None. **K. Deisseroth:** None. **G.F. Corder:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.03

Topic: D.02. Somatosensation – Pain

Support: T31DT1729

Title: Insula to basolateral amygdala circuit drives pain response

Authors: *M. HUI[1], G. J. SALIMANDO[2], G. F. CORDER[2], K. T. BEIER[1];
[1]Univ. of California, Irvine, Irvine, CA; [2]Psychiatry, Univ. of Pennsylvania, Philadelphia, PA

Abstract: Most pain experiences are perceived as aversive and are highly modifiable by attention, context, and experience. This shared and dynamic affective feature among pain types suggests the existence of a common and distributed set of neural circuits in the brain for transforming nociception into negative affective information. We recently identified a key neural ensemble in the basolateral amygdala (BLA) required for pain affective behaviors. However, it is not resolved how and to what function this BLA circuit integrates with cortical structures for pain perception, or how these integrated circuits are modulated during chronic pain states. Taking a whole-brain view, using immediate early gene mapping in combination with an activity-based rabies viral tracing screen, in iDisco++ cleared tissue, we identified several brain regions that directly synapse onto the BLA nociceptive ensemble with increased labeling during the chronic pain state, which is indicative of persistently elevated levels of activity that correspond to elevated intrinsic excitability. Among these sensitized input regions was the anterior insular cortex (AIC).To determine the functional role of this AIC->BLA circuit in pain we employed an intersectional genetic approach to silence these projection neurons using a viral-transduced inward rectifying potassium channel. We found that inhibition of AIC->BLA cell-types prevented sensitization of nocifensive behaviors following repetitive noxious thermal stimulation, and decreased sensitivity to noxious thermal stimuli in a temperature place-preference assay, without changes to overall locomotion. Our results raise the possibility that the AIC might shape the excitability of subcortical circuits such as the BLA to drive top-down facilitation of pain chronification. We are now exploring inhibition of this circuit prior to a chronic pain-inducing injury for its contribution to neuropathic pain development, and further investigating the molecular, genetic, physiological underpinnings of the larger connected pathways with the AIC and BLA.

Disclosures: **M. Hui:** None. **G.J. Salimando:** None. **G.F. Corder:** None. **K.T. Beier:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.04

Topic: D.02. Somatosensation – Pain

Support: NIH Grant F32DA055458
NIH Grant F32DA053099
NIH Grant DP2GM140923
Rita Allen Foundation Award in Pain

Title: Mu opioid receptor expression defines a nociceptive neural ensemble in the ventrolateral periaqueductal gray

Authors: B. A. KIMMEY[1], N. M. MCCALL[1], C. RAMAKRISHNAN[2], K. DEISSEROTH[2], G. CORDER[1];
[1]Univ. of Pennsylvania, Philadelphia, PA; [2]Stanford Univ., Stanford, CA

Abstract: Opioid analgesics engage mu opioid receptor (MOR) signaling across multiple brain regions to alleviate pain. Notably, the ventrolateral periaqueductal gray (vlPAG) plays a dual functional role for both nociceptive processing and robust antinociception. However, the molecular identity, signaling dynamics, and plasticity of MOR+ neurons in vlPAG, as well as their role in acute and chronic pain, remains unclear. Here, we characterized the nociceptive MOR+ neural populations in the vlPAG to gain insight into the molecular markers and temporal dynamics that define this functional ensemble. To this end, we employed mouse and viral genetic approaches to capture, monitor, and manipulate vlPAG cell-types at the intersection of nociception and molecular MOR expression across acute and inflammatory pain states. Using the targeted recombination in active populations (TRAP) approach, we found a gradient in pain-active vlPAG neurons that increased posteriorly, suggesting spatial heterogeneity in vlPAG with respect to pain processing. Next, we distinguished molecular markers of pain-active vlPAG MOR+ neurons, such as *Vglut2* and *Vgat*, while further defining the projection targets of MOR+ cells using TRAP. Capitalizing on a MOR promoter-driven viral vector, we used *in vivo* fiber photometry imaging to record calcium transient activity reported by fluorescence of the genetically encoded calcium indicator GCaMP6f in MOR+ vlPAG neurons. With this approach, we discovered that vlPAG MOR+ neurons broadly demonstrate increased calcium activity in response to acutely noxious stimuli that was suppressed by morphine. Additionally, calcium activity in this population was enhanced following induction of Complete Freund's Adjuvant inflammatory pain. In contrast to the MOR+ population, optogenetic activation of enkephalinergic neurons in vlPAG produced antinociception during hotplate exposure indicating a potential local microcircuit that blunts the activity of MOR+ neurons and associated pain behavior. Experiments employing chemogenetic manipulation of vlPAG MOR+ neurons are ongoing, which test our hypothesis that inhibition of vlPAG MOR+ neurons will be antinociceptive while activation of the same neurons will be pronociceptive. In total, we have uncovered important properties of MOR+ neuronal populations and pathways in the vlPAG, a critical nociceptive brain region. These data will inform targeted interventions to selectively

modulate pain-related neurocircuitry and thereby mitigate the harmful effects of prescription opiates.

Disclosures: B.A. Kimmey: None. **N.M. McCall:** None. **C. Ramakrishnan:** None. **K. Deisseroth:** None. **G. Corder:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.05

Topic: D.02. Somatosensation – Pain

Support: NIH NIDA R00 DA043609-05
NIH DP2 GM140923-01

Title: Characterization of a Nociceptive Amygdala to Accumbens Neural Circuit

Authors: *J. A. WOJICK[1], N. M. MCCALL[1], J. G. JAMES[1], S. TONEGAWA[2], G. F. CORDER[1];
[1]Univ. of Pennsylvania, Philadelphia, PA; [2]MIT, MIT, Cambridge, MA

Abstract: Why does the experience of pain hurt, feel unpleasant, and lead to protective behaviors? A key first step to understand this aversive aspect of pain is to functionally and genetically identify nociceptive neurons connected across affective-motivational neural circuits, and how injury and chronic pain alter the dynamics of these nociceptive cell-types. We recently identified a nociceptive subpopulation of negative valence basolateral amygdala (BLA) neurons essential for pain aversion. Here, we used targeted recombination in active populations (TRAP) mice to genetically capture nociceptive BLA neurons (*noci*TRAP) and to test their necessity and sufficiency to produce nociceptive hypersensitivity and negative affective behaviors. Next, we imaged bulk fluorescent calcium activity of *noci*TRAP and negative valence *Rspo2*+ BLA somas and axon terminals in a downstream target of *noci*TRAP BLA neurons—the nucleus accumbens Shell (NAcSh)—across the transition from acute to chronic neuropathic pain. Broadly, aberrant activity of the NAcSh has been linked to motivational deficits in chronic pain, yet much remains unknown regarding specific NAcSh nociceptive cell-types or their modulation by BLA nociceptive neurons. Surprisingly, we found *Rspo2*+ BLA axon terminals, which spread across the anterior-posterior axis of the NAcSh, are largely inhibited in response to highly-salient noxious and non-noxious stimuli. Using histological methods, we found a previously unreported posterior medial NAcSh subnuclei—anatomically encompassed by the Islands of Cajella granule cell clusters—that contains numerous *noci*TRAP neurons. This region, which we termed the "*NAcre*" (named after the inner shell layer of mollusks), receives projections from nociceptive BLA neurons. Importantly, the majority of acute *noci*TRAP NAcre neurons also display increased immediate early gene FOS expression to light touch following a peripheral nerve injury, revealing consistent activation across pain states independent of stimulus sensory

modality. Finally, we found *noci*TRAP NAcre neurons, while molecularly heterogeneous, were primarily medium spiny neurons expressing dopamine receptor 2 and kappa opioid receptor mRNA. In total, the NAcre is a nociceptive subregion of the posterior medial NAcSh that receives transmissions from the BLA and may be involved in pain-related kappa opioid aversion processes. Further work will determine the necessity and sufficiency of the NAcre and the BLA axon projections it receives for affective-motivational behaviors in acute and chronic pain states.

Disclosures: **J.A. Wojick:** None. **N.M. McCall:** None. **J.G. James:** None. **S. Tonegawa:** None. **G.F. Corder:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.06

Topic: D.01. Somatosensation

Title: Proprioceptive sensory attenuation in area 3a during voluntary movement and action observation in macaque

Authors: *K. SEKI[1], **S. KIKUTA**[1], S. KUBOTA[1], J. CONFAIS[1], A. YARON[1,2], T. OYA[1,3,4]; [1]Natl.inst.Neurosci., Tokyo, Japan; [2]Intl. Res. Ctr. for Neurointelligence, The Univ. of Tokyo, Tokyo, Japan; [3]The Brain and Mind Inst., [4]Dept. of Physiol. and Pharmacol., Univ. of Western Ontario, London, ON, Canada

Abstract: Our sensory systems respond not only to external sensory information but also to self-generated information caused by our movement. For example, we feel less when we move. This phenomenon is known as sensory attenuation. Although the sensory attenuation of cutaneous afferent input to the CNS is well documented, that of a proprioceptive afferent signal is not. Furthermore, it is unclear if the comparable modulation of afferent sensory input could also be triggered by the non-motor-related events, e.g., action observation. To address these questions, we recorded somatosensory-evoked potentials (SEPs) from Brodman's area 3a (BA3a) by applying electrical stimuli to the cuff electrode implanted in the deep radial nerve (DR: muscle afferent of wrist extensor) (DR-SEPs) and that from BA3b from the superficial radial nerve(SR: cutaneous afferent from wrist dorsum)(SR-SEPs) while monkeys performing reaching and grasping (self-movement) (n=2), and observing person sitting in front of monkeys performing the same task (action observation)(n=1). During self-movement task, we found that the size of DR-SEPs ($p < 0.01$), not only the SR-SEP ($p < 0.01$), was significantly suppressed in the movement phase compared to the pre-movement control period (inter-trial-interval; t-test). Therefore, we conclude that the cortical SEPs evoked by muscle and cutaneous afferents show sensory attenuation during movement execution. Interestingly, we also found two contrasting results between DR- and SR-SEP's modulation during the self-movement and action observation tasks. First, while the onset of DR-SEP attenuation was after the movement initiation, that for SR-SEP attenuation is before the movement onset in the self-movement task ($p < 0.01$). Second,

while we found no difference in the size of DR-SEPs during the task, the size of SR-SEPs was consistently larger throughout the action observation task; and that was also a more significant pre-movement control period during the self-movement task, both compared with the inter-task interval period. These differences between DR-SEPs and SR-SEPs suggest the differential source of sensory gain modulation to the primary sensory cortex's proprioceptive and cutaneous sensory input. While the sensory attenuation of the proprioceptive input could be driven robustly by the efference copy of volitional motor command, the cutaneous signal could be more flexibly modulated depending on its value for current and future movement.

Disclosures: K. Seki: None. **S. Kikuta:** None. **S. Kubota:** None. **J. Confais:** None. **A. Yaron:** None. **T. Oya:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.07

Topic: D.02. Somatosensation – Pain

Support: NRF-2019R1I1A1A01059697
NRF-2020R1A2C3008481

Title: Insular cortex stimulation modulates synaptic plasticity for the relief of neuropathic pain in rats

Authors: *K. KIM[1], M. CHA[1], G. NAN[1,2], L. KIM[1,2], B. H. LEE[1,2];
[1]Dept. of Physiol., Yonsei Univ. Col. of Med., Seoul, Korea, Republic of; [2]Grad. Sch. of Med. Science, Brain Korea 21 Project, Yonsei Univ. Col. of Med., Seoul, Korea, Republic of

Abstract: Neuropathic pain evokes typical symptoms such as allodynia or hyperalgesia that persists for days or years, and the enhancement of synaptic plasticity deteriorates the symptoms. The insular cortex (IC) is involved in pain perception, and the neural plasticity in the IC can be induced by nerve injury. Although many treatments have been developed to attenuate the neuropathic pain, the side effects and tolerance remained. Therefore, many prospective cases of brain stimulation in neuropathic pain have reported the pain-relieving effect. However, the fundamental mechanisms of brain stimulation, especially the IC stimulation (ICS), are still elucidated. This study aimed to investigate the pain-relieving effect induced by ICS in neuropathic rats and elucidate the mechanisms of the synaptic plasticity modulation through ICS. Behavioral tests were conducted to observe the pain-relieving effects in neuropathic rats. Western blot was performed to identify the changes in the expression level of synaptic plasticity-related receptors such as the subunit of N-methyl-D-aspartate receptor (NMDAR), and α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptor (AMPAR) after ICS. Proteomics was conducted to detect specific proteins which contributed to pain attenuation by ICS among the groups. Consequently, neuropathic pain was alleviated by ICS at 50 Hz-120 μA. Although ICS

was stopped, the alleviation effect was maintained for 4 days. The expression level of NR2A which is a subunit of NMDAR was not changed between groups. However, the expression levels of AMPAR and NR2B was significantly decreased in ICS group than sham stimulation group. Bioinformatics analysis of proteomics suggested that collapsin response mediator protein 2 (CRMP2) was markedly altered between the groups. These results inferred that the ICS reduced the long-term potentiation (LTP) and CRMP2 could attenuate LTP accompanied by neuropathic pain following neural injury.

Disclosures: **K. Kim:** None. **M. Cha:** None. **G. Nan:** None. **L. Kim:** None. **B.H. Lee:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.08

Topic: D.02. Somatosensation – Pain

Support: CIHR grant FDN-159906
ANR-18-CE37-0004

Title: Chronic pain-induced depression changes the excitation-inhibition balance of anterior cingulate cortex

Authors: *J. ALONSO[1], I. YALCIN-CHRISTMANN[2], Y. DE KONINCK[1];
[1]CERVO Brain Res. Ctr., Laval Univ. / IUSMQ, Quebec, QC, Canada; [2]CNRS INCI, Strasbourg Cedex, France

Abstract: Chronic pain and major depressive disorder are two pathologies with a strong comorbidity. The anterior cingulate cortex (ACC) is a brain structure involved in both pathologies. Its activity appears important in the development of anxio-depressive like behaviors, but the underlying mechanisms remain unclear. While most studies try to explain modification in activity with plasticity in glutamatergic (GLU) cells, we characterized the activity of GABAergic (GABA) cells and the effect of the inhibition on GLU cells in the ACC of a mouse model of chronic pain-induced depression (CPID). We used adult male mice that underwent a sciatic nerve constriction surgery as an experimental model of neuropathic pain. Control animals had the nerve exposed but not constricted. Eight weeks after surgery, animals presented mechanical hypersensitivity and anxio-depressive like behavior. All animals received an injection in the ACC of an adeno-associated virus encoding the light-sensitive channel Channelrhodopsin 2 (ChR2) under the mDlx enhancer for selective expression in GABA neurons. The ACC activity was studied by recording single cell activity *in vivo* in animals anaesthetised with urethane and isoflurane. Recordings were performed with an all-glass micro-optrode allowing simultaneously extracellular electrophysiological recordings and optogenetic stimulations. Locally delivered light stimulations allowed identification of the cell types and to characterize their electrophysiological properties. Recordings were obtained from 28 animals with CIPD and 30

control animals. We found that GABA cells of CPID animals were less able to sustain firing to repetitive light stimulation than those of control animals ($P < 0.05$; 2-ways ANOVA). Consistently, putative GLU cells of CPID animals were less inhibited by increasing frequency stimulation than those of control animals ($P < 0.05$; 2-ways ANOVA). Finally, a subset of GLU cells displayed a rebound of activity after being inhibited and that rebound was larger in CPID animals than in controls ($P < 0.001$; non-linear regression test). The results indicate that, in the CPID model, GABAergic inhibition in the ACC is impaired and this appears linked to changes in intrinsic excitability of GABA neurons. The inhibition of GLU cells appear to be also reduced because of rebound excitation following optogenetic inhibitory control. These findings could lead to the implementation of a new treatment for depression induced by chronic pain allowing re-establishing a physiological activity level.

Disclosures: J. Alonso: None. **I. Yalcin-Christmann:** None. **Y. De Koninck:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.09

Topic: D.02. Somatosensation – Pain

Support: NIH MH116003
NIH NS118731

Title: Analysis of analgesic effect of recombinant Cbln1 in the central amygdala

Authors: *S. SABNIS, P. GANDHI, D. Y. GAWANDE, S. DRAVID;
Creighton Univ., Omaha, NE

Abstract: Analysis of analgesic effect of recombinant Cbln1 in the central amygdalaAuthorsSiddhesh Sabnis[1], Pauravi J. Gandhi[1], Dinesh Y. Gawande[1], Shashank M. Dravid[11] Department of Pharmacology and Neuroscience, Creighton University School of Medicine, Omaha, NE 68178, USA.
DisclosureSiddhesh Sabnis: None, **Pauravi J Gandhi:** None, **Dinesh Y. Gawande:** None, **Shashank M. Dravid:** None
AbstractThe amygdala, being a central hub for processing and tuning pain signals, has been implicated in many debilitating chronic pain conditions. We have recently shown that dysregulation of GluD1-Cbln1 signaling in the central amygdala contributes to aversive and affective behaviors in inflammatory and neuropathic pain. Specifically, downregulation of GluD1 and Cbln1 was found in the central amygdala in pain models. Administration of Cbln1 through intra-cerebroventricular (icv; 1.5ug/1.5ul) and intra-central amygdalar (iCeA; 250ng/0.5ul) route in the inflammatory pain models were found to be highly effective in relieving pain. In contrast, recombinant Cbln1 was ineffective in reducing pain-related behaviors in GluD1 KO mice demonstrating GluD1-dependent action of Cbln1. In the present study, we

explored the physiological role of Cbln1 in normal animals and the structural-functional aspect of GluD1-Cbln1 signaling relevant to the pain pathway. Administration of recombinant Cbln1 in normal wild type animals increased the hypersensitivity and decreased the paw withdrawal threshold. This effect was completely abolished by iCeA co-administration of D-serine. Moreover, D-serine by itself did not produce any significant effects when administered into CeA in control wild type animals. These results imply that D-serine potentially reduces the hypersensitivity caused by Cbln1 in wild type animals. The effect of D-serine was in agreement with our previous results which showed that the D-serine blocked the analgesic effect of the Cbln1 in the CFA injected WT animals. In the ongoing studies we are exploring the dependency of the D-serine's effect through GluD1.

Disclosures: S. Sabnis: None. **P. Gandhi:** None. **D.Y. Gawande:** None. **S. Dravid:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.10

Title: WITHDRAWN

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.11

Topic: D.02. Somatosensation – Pain

Support: Heidelberg Pain Consortium (SFB 1158, Project B08)

Title: Multiscale correlation of structural plasticity of cortical grey matter volume in chronic pain

Authors: *A. DAS GUPTA[1], J. JOHN[1], L. ASAN[1], C. FALFAN-MELGOZA[2], C. BERETTA[1], W. WEBER-FAHR[2], T. KUNER[1], J. KNABBE[1,3];
[1]Functional Neuroanatomy, Inst. of Anat. and Cell Biology, Heidelberg Univ., Heidelberg, Germany; [2]Dept. of Neuroimaging, Central institute for mental health, Mannheim, Germany; [3]Dept. of Gen. Psychiatry, Ctr. for Psychosocial Medicine, Heidelberg Univ., Heidelberg, Germany

Abstract: Grey matter volume (GMV) changes due to chronic pain have been extensively studied in humans, showing the cingulate and insular cortices to be the main areas of the brain

undergoing a change in GMV within two months after onset of chronic pain. However, the underlying neurobiological mechanisms behind the development of chronic pain and changes in GMV are poorly understood. The goal of the project was to investigate the cellular underpinnings of GMV alterations in chronic pain and provide a basis for novel strategies for prevention and therapy of this debilitating disease.
In a longitudinal study design, a MRI-compatible chronic cranial window was implanted on top of the prefrontal cortex of male mice expressing eGFP in all cell nuclei (HIST1H2BB/EGFP, 7-9 weeks old), allowing in vivo two photon volumetric imaging and analysis of all cell nuclei as well as the reidentification of the same imaging location over time. MRI imaging (voxel-based morphometry, diffusion tensor imaging) was performed in parallel to in vivo two-photon imaging of the anterior cingulate cortex (ACC) at different timepoints up to 12 weeks after the induction of chronic neuropathic pain with the spared nerve injury model (SNI). Additionally, behavioral paradigms were employed during acute pain and progression towards chronic pain to capture emotional effects of pain. Using a combination of distinct nuclear features (intensity, morphological and texture) of the eGFP signal, a deep learning neural network was trained with ground truth data to reliably identify different cell types (neuronal, astroglial, oligodendroglial, microglial and endothelial) based on their respective nuclear features. Preliminary analysis of behavioral data show depressive and anxious behavior phenotypes in SNI mice. Analysis of imaging data reveals differences between sham-operated and SNI mice regarding nuclei properties and tissue volume, which correlate significantly with GMV change and behavior. Furthermore, employing the newly developed nuclei classification algorithm, changes caused by chronic pain affecting a few specific cell types could be identified.
In summary, this study established a novel multi-modal imaging approach suitable to provide a more comprehensive understanding of the cellular mechanisms underlying changes in GMV caused by chronic pain, to answer major open questions in the field of pain research.

Disclosures: **A. Das Gupta:** None. **J. John:** None. **L. Asan:** None. **C. Falfan-Melgoza:** None. **C. Beretta:** None. **W. Weber-Fahr:** None. **T. Kuner:** None. **J. Knabbe:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.12

Title: WITHDRAWN

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.13

Topic: D.02. Somatosensation – Pain

Title: Functional alterations of intrinsic networks at various stages of neuropathic pain and its comorbidity

Authors: *S. CAZZANELLI[1,2], S. DIEBOLT[1,2], A. BERTOLO[1,2], J. FERRIER[1], M. VERT[1,2], L. DELAY[2], T. DEFFIEUX[2], B.-F. OSMANSKI[1], M. TANTER[2], S. PEZET[2];
[1]Iconeus, Paris, France; [2]Physics for Med. - Inserm, ESPCI Paris-PSL, CNRS, Paris, France

Abstract: Chronic pain is an abnormal pain sensation that persists longer than the temporal course of natural healing. It interferes with the emotional well-being of the patient and leads to several comorbidities, such as anxiety and depression. It has been hypothesized that chronic pain is due to abnormal, maladaptive neuronal plasticity in the structures known to be involved in pain perception (Bliss et al. 2016). This means that nerve injury would trigger long-term potentiation of synaptic transmission in pain-related areas (Zhuo et al. 2013). Our hypothesis is that the aforementioned maladaptive plasticity in these brain areas could be key mechanisms for the development of comorbidities, such as anxiety and depression. This study aimed at identifying, using functional ultrasound (fUS) imaging, how the functionality of brain networks is altered in link with neuropathic pain and/or the associated comorbidities. We measured the functional connectivity (FC) at rest in awake, head fixed animals: I) naïve, II) subjected to neuropathic pain (2W cuffing of the sciatic nerve), or III) during the emergence of either anxiety (8W) or IV) depression (12W).The experiments were performed in awake, head-restrained animals. We measured functional connectivity at rest, transcranially, in 3 different sagittal planes overtime. N=20 mice (n=9 Sham and n=11 neuropathic) were included in the study and each animal was re-imaged several times and overall, n=98 acquisitions were generated (n=47 Sham and n=51 neuropathic). Our results show alterations of the intrinsic connectivity between control (Sham) and neuropathic (NP) mice (treatment effect), in specific regions of the 'pain network' at 2W. More precisely: in the neuropathic group, the connectivity between the different subregions of the insula (AID, AIP, AIV) and as well between somatomotor regions is increased compared to the sham group.On the other hand, at the following time points (8W and 12W) the FC changed but similarly changing in both groups, suggesting a time effect that could be interpretated as linked to the aging of the animal, more than to a treatment effect. In conclusion, the results at 2W supports our hypothesis that the 'pain network' undergoes a maladaptive plasticity during the development of neuropathic pain. However, the changes associated with comorbidities are more complex and suggest that an ageing affect might be involved as well.

Disclosures: S. Cazzanelli: None. **S. Diebolt:** None. **A. Bertolo:** None. **J. Ferrier:** None. **M. Vert:** None. **L. Delay:** None. **T. Deffieux:** None. **B. Osmanski:** None. **M. Tanter:** None. **S. Pezet:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.14

Topic: D.02. Somatosensation – Pain

Support: NRF-2017R1A2B3005753
NRF-2019R1I1A1A01059697
NRF-2020R1A2C3008481

Title: Glial cells in the insular cortex modulate neuropathic pain

Authors: *L. KIM[1,2], K. KIM[1], G. NAN[1,2], M. CHA[1], B. H. LEE[1,2];
[1]Dept. of Physiol., Yonsei Univ. Col. of Med., Seoul, Korea, Republic of; [2]Grad. Sch. of Med. Science, Brain Korea 21 Project, Yonsei Univ. Col. of Med., Seoul, Korea, Republic of

Abstract: The insular cortex (IC) is essential for regulating nociception and is associated with pain behavior. However, analgesic effects of glial inhibition in the IC have not yet been explored. The aim of this study was to investigate pain-relieving effects after glial inhibition in the IC during the early or late phase of pain development. The effects of glial inhibitors in early or late phase of neuropathic pain were characterized in astrocytes and microglia expressions in the IC of an animal model of neuropathic pain. Changes in withdrawal responses during different stages of inhibition were compared and morphological changes in glial cells with purinergic receptor expressions were analyzed. Inhibition of glial cells had an analgesic effect that persisted even after drug withdrawal. Both GFAP and CD11b/c expressions were decreased after injection of glial inhibitors. Morphological alterations of astrocytes and microglia were observed with expression changes of purinergic receptors. These findings indicate that inhibition of glial activity in the IC alleviates chronic pain, and that purinergic receptors in glial cells are closely related to chronic pain development.

Disclosures: L. Kim: None. **K. Kim:** None. **G. Nan:** None. **M. Cha:** None. **B.H. Lee:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.15

Topic: D.02. Somatosensation – Pain

Support: National Medical Research Council
National University of Singapore, University Strategic Research (DPRT/944/09/14)
NUS Yong Loo Lin School of Medicine Aspiration Fund (R-185-000-271-720)
MOE AcRF Tier 3 Award MOE2017-T3-1-002
Ms Karen Chung for her assistance in animal husbandry

Title: Septal Neurokinin-1 Receptor - Cholinergic Neuron Axis Mediates Experimental Neuropathic Pain.

Authors: M. ARIFFIN, S. NG, S. LO, D. KOH, N. LOH, N. MICHIKO, H. NADIA, *S. KHANNA;
Natl. Univ. Singapore, Natl. Univ. Singapore, Singapore, Singapore

Abstract: While the expression and/or availability of forebrain neurokinin-1 receptors (NK1Rs) are altered in chronic pain in both human and rodent, their function in chronic pain, especially neuropathic pain remains unclear. The NK1R are canonical receptor for substance P (SP) and have been extensively studied for their role in nociception, especially in the spinal cord and brainstem. Interestingly, the NK1R in the forebrain medial septum (MS) are localized exclusively on cholinergic neurons. The septal cholinergic neurons project both to the medial prefrontal cortex (mPFC) and the hippocampus. All three regions, namely MS, the mPFC and the hippocampus are implicated in nociception. In the present study we have explored the hypothesis that a NK1R-cholinergic neurons axis in the septo-hippocampus mediate experimental neuropathic pain. In this context, our investigations show that optogenetic stimulation (20Hz, 25mW) of septal cholinergic neurons in mice, or intraseptal microinjection of SP (2ug/ul, 0.5ul) in rat evoked hyperreflexia in uninjured animals. The hyperreflexia was marked by a decrease in paw withdrawal threshold (PWT) to mechanical stimuli and the paw withdrawal latency (PWL) to thermal stimuli. The hyperreflexia was attenuated by systemic atropine sulphate (5mg/kg, i.p.), an antagonist at muscarinic-cholinergic receptors. This suggests that SP excites septal cholinergic neurons to evoke aversive behaviour. Consistently, intraseptal SP also evoked a conditioned place avoidance. Furthermore, intraseptal microinjection of L-733,060 (0.0176ug/ul, 0.5ul), an antagonist at NK1Rs, or bilateral microinjection of atropine (0.007ug/ul, 0.5ul) or mecamylamine (2ug/ul, 0.5ul), an antagonist of nicotinic receptor, into dorsal hippocampus (DH) attenuated peripheral hypersensitivity (PH) in chronic constriction injury model (CCI) of neuropathic pain. While pre-emptive destruction of septal cholinergic neurons prevented the development of PH. Collectively, the preceding suggests that an axis of septal NK1R-cholinergic neurons with DH mediate the development and maintenance of experimental neuropathic pain. Interestingly, the antinociceptive effect of intraseptal L-733,060 was submaximal, partially reducing PH and the cellular responses in forebrain mPFC but not spinal cord. While microinjection of cholinergic antagonists into DH reversed PH and attenuated cellular changes in both forebrain and spinal cord. Thus, while nociception-induced activation of septal NK1R evoke a submaximal nociception, the nociception-induced cholinergic activation evokes a maximal nociceptive drive through downstream DH.

Disclosures: M. Ariffin: None. **S. Ng:** None. **S. Lo:** None. **D. Koh:** None. **N. Loh:** None. **N. Michiko:** None. **H. Nadia:** None. **S. Khanna:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.16

Topic: D.02. Somatosensation – Pain

Support: NIH Grant R34NS121875 (AO)
NIH Grant R01DA023281 (LT)
NIH Grant R01DA040882 (LT, AO)
FAU Brain Institute Pilot Grant Award (AO)

Title: Neural circuits regulating pain behaviors in the anterior cingulate cortex

Authors: D. PETERS[1], M. MINKOFF[1], K. M. TARGOWSKA-DUDA[1,2], L. TOLL[1], *A. OZAWA[1];
[1]Dept. of Biomed. Science, Charles E. Schmidt Col. of Med., Florida Atlantic Univ., Boca Raton, FL; [2]Dept. of Biopharmacy, Med. Univ. of Lublin, Lublin, Poland

Abstract: Pain is an unpleasant sensory experience triggered by noxious stimuli, which provokes essential protective behaviors against life threatening events. Changes in neural activity within distinct regions of the anterior cingulate cortex (ACC) are known to be linked to many psychological and motor functions and have also been shown to take part in regulating pain-related behaviors. Yet, there is very limited understanding on the mechanism by which the ACC regions assign the emotional condition to nociceptive information to generate pain aversion at neural circuit and cellular levels. Here we investigate the functionality of the neural circuits potentially regulating pain behaviors by focusing on pain-activated neurons in the ACC using c-Fos-2A-iCre (TRAP2) mice combined with neuroanatomy and chemogenetics under a chronic pain condition. We observed that the application of light touch, generally a non-painful stimulus for naïve animals on injured hindpaw prominently increases c-Fos expression in the ACC compared to that in uninjured mice. Selectively inhibiting these pain-activated ACC neurons via hM4Di receptors (an inhibitory DREADD, Designer Receptors Exclusively Activated by Designer Drugs) reduced affective pain, as demonstrated by the development of conditioned place preference. Interestingly, these chronic pain-dependently activated ACC neurons represented are responsible for regulating sensory pain. In neuroanatomical analysis using TRAP2 mice crossed with Ai9 (Cre-dependent tdTomato reporter) mice combined with a retrograde tracer, we also found a greater percentage of specific activated ACC circuits in chronic pain animals compared to non-injured animals. These findings provide us with information about the ACC circuitries potentially regulating distinct pain behaviors under chronic pain conditions.

Disclosures: D. Peters: None. **M. Minkoff:** None. **K.M. Targowska-Duda:** None. **L. Toll:** None. **A. Ozawa:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.17

Topic: D.02. Somatosensation – Pain

Support: NIH BRAIN Grant R01AT011447

Title: Functions of Tacr1- and Gpr83-expressing spinoparabrachial neurons and their neuropeptide signaling in acute and neuropathic pain

Authors: *S. CHOI[1], J. TURECEK[1], A. R. MAGEE[1], D. A. YARMOLINSKY[2], C. J. WOOLF[2], D. D. GINTY[1];
[1]Neurobio., Harvard Med. Sch., Boston, MA; [2]F.M. Kirby Neurobio. Ctr., Boston Children's Hosp., Boston, MA

Abstract: Pain is initiated by the activation of nociceptors that innervate the skin and internal organs. Nociceptive signals are propagated into the spinal cord and then transmitted to the brain by spinal cord projection neurons (PNs). These spinal PNs are attractive therapeutic targets for pain treatment because nociceptive signals emanating from the periphery are channeled through these spinal cord output neurons *en route* to the brain to produce pain sensations. Spinoparabrachial (SPB) neurons, a major population of spinal PNs that innervate the lateral parabrachial nucleus of the pons, represent an ideal neuronal population for developing new approaches to treat pain because they convey touch and pain information to higher brain centers that control the affective aspects (i.e., emotional “feelings”) of touch and pain. Previously, we showed that *Tacr1*$^+$ and *Gpr83*$^+$ SPB neurons form two largely-nonoverlapping subdivisions of the SPB pathway that cooperate to convey tactile, thermal and noxious signals from the spinal cord to the brain. To further define the contribution of each SPB subdivision to pain sensation and associated behavioral responses to noxious stimuli, we have begun to examine the effects of acute silencing of *Tacr1*$^+$ and *Gpr83*$^+$ SPB neurons, individually or simultaneously, on nocifensive behaviors using a newly generated dual recombinase-dependent SYP1-miniSOG mouse line that enables selective, light-dependent silencing of synaptic transmission. In addition, to determine if neuropeptide signaling mediated by either TACR1, GPR83 or both in the spinal cord is required for pain transmission, we also have begun to examine acute and neuropathic pain behaviors following spinal cord-specific deletions of the *Tacr1* and *Gpr83* genes using mouse lines that harbor conditional alleles of *Tacr1* and *Gpr83* in conjunction with spinal cord specific Cre lines. Collectively, these behavioral analyses will provide insights into the functions of *Tacr1*$^+$ and *Gpr83*$^+$ SPB neurons and neuropeptide signaling mediated by the TACR1 and GPR83 GPCRs in acute and neuropathic pain and may reveal novel therapeutic targets for treating pain.

Disclosures: S. Choi: None. **J. Turecek:** None. **A.R. Magee:** None. **D.A. Yarmolinsky:** None. **C.J. Woolf:** None. **D.D. Ginty:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.18

Title: WITHDRAWN

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.19

Topic: D.02. Somatosensation – Pain

Support: NIH Contract No. 75N95019D00026

Title: Elucidating pain-related activity in the EEG from the rat spinal nerve ligation model (SNL) of neuropathic pain

Authors: *L. DEVI[1], D. BRUNNER[1], A. YELISYEYEV[1], A. ASCHENBRENNER[1], S. A. WOLLER[2], S. IYENGAR[3], S. R. MORAIRTY[1], S. LEISER[1];
[1]PsychoGenics, Inc., Paramus, NJ; [2]DTR, [3]NIH/NINDS, Rockville, MD

Abstract: In both clinical settings and in preclinical models, specific electroencephalographic (EEG) frequencies increase during high pain states. In collaboration with the NIH HEAL Initiative Preclinical Screening Platform for Pain (PSPP) we conducted a longitudinal evaluation of the EEG for pain signatures in the spinal nerve ligation (SNL) model of pain in male and female Sprague Dawley rats for a period of 120 days following SNL surgery. Naïve, Sham and SNL rats were implanted for EEG recordings. Ligation of the L5 and L6 spinal nerves was performed as described in Kim and Chung (1992). Gait analyses and paw withhold threshold were also analyzed and reported elsewhere. EEG recording was performed in freely behaving rats for ~22 hours and analyzed to identify quiet wake epochs using an automated wake classifier. Quiet wake epochs were detected using an in-house developed algorithm based on Recurrent Neural Networks. The algorithm was trained with rat EEG data scored by humans for 10-s epochs corresponding to resting wake periods (defined as alertness with no locomotor behavior) and removed epochs with artifact. Absolute Theta (4-8 Hz), Low gamma (30-50 Hz), and High gamma (65-100 Hz) power was extracted from wake periods using a MATLAB program written by Carl Saab and colleagues (Saab et. al, 2012, 2013). For normalization, we selected a period with stable quiet wake behavior (the first 6 h of the dark phase) and generated mean pre-SNL EEG power. To minimize inter-subject variability, each subject's post-SNL frequency power data was normalized by their corresponding pre-SNL baseline. All statistical analysis were conducted using the Linear Mixed Model with random intercepts and random slopes on *Time* nested within *Session* (day). A significant and sustained S1 EEG theta power was observed in the female SNL group as compared to the Naïve and Sham groups. There was a transient elevation of S1 theta in female Sham rats following surgeries (D7-D21) as compared to the naïve controls. A trend for an elevated theta power in PFC in the males failed to reach significance. Low and high Gamma did not show any significant changes in the SNL model. These data and any other relevant ongoing analyses will also be discussed.

Disclosures: L. Devi: A. Employment/Salary (full or part-time):; Full time. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and

pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **D. Brunner:** A. Employment/Salary (full or part-time):; Full time. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **A. Yelisyeyev:** A. Employment/Salary (full or part-time):; Full time. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **A. Aschenbrenner:** A. Employment/Salary (full or part-time):; Full time. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **S.A. Woller:** A. Employment/Salary (full or part-time):; Full time. **S. Iyengar:** A. Employment/Salary (full or part-time):; Full time. **S.R. Morairty:** A. Employment/Salary (full or part-time):; Full time. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **S. Leiser:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.20

Topic: D.02. Somatosensation – Pain

Title: Parabrachial NPY Y1 receptor-expressing neurons allow for the gating of inflammatory pain by hypothalamic AgRP neurons

Authors: *N. GOLDSTEIN, J. R. E. CARTY, J. N. BETLEY;
Univ. of Pennsylvania, Philadelphia, PA

Abstract: During food deprivation, food intake must be prioritized over non-essential behaviors to ensure survival. For example, protecting an injury from further damage is important, but cannot come at the expense of starvation. We have recently demonstrated that hunger suppresses persistent pain through a hypothalamic-to-parabrachial circuit that is activated during caloric deficit. Agouti-related protein (AgRP)-expressing neurons in the arcuate nucleus projecting to the lateral parabrachial nucleus (lPBN) suppress behavioral pain responses in several models of persistent and chronic pain. AgRP neurons projecting to the lPBN suppress persistent pain through the release of neuropeptide Y (NPY) onto the Y1 NPY receptor. We have determined that the site of action is on Y1 receptor (Y1R)-expressing neurons in the lPBN. lPBN Y1R neurons are activated during pain and are anatomically positioned to integrate pain and hunger

information. In addition, Y1R neurons condition a strong place avoidance and are required for proper expression of pain behavior. Hunger or AgRP neuron stimulation selectively attenuates Y1R neuron activity during long term inflammatory pain but not during acute nociceptive stimuli. Together, our work suggests that lPBN Y1R neurons are integrating hunger and nociceptive information to prioritize food intake over the response to persistent pain to promote survival.

Disclosures: **N. Goldstein:** None. **J.R.E. Carty:** None. **J.N. Betley:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.21

Topic: D.02. Somatosensation – Pain

Support: T32 NS 007421
R01NS118504
R01NS106301
New York Stem Cell Foundation
McKnight Endowment Fund for Neuroscience
Brain Research Foundation

Title: A cell atlas of the amygdala enables synergistic pharmacology against pain unpleasantness

Authors: *D. BERG[1,4,5,6,7], D. LEE[1,2], J. KRZESKI[1], M. CHEN[5,8], X. JIANG[4,5,6,7], Y. KE[4,5,6,7], S. QUAKE[5,8,9], M. J. SCHNITZER[4,5,6,7], G. SCHERRER[1,3,10];
[1]UNC Neurosci. Center, Dept. of Cell Biol. and Physiol., [2]Dept. of Pharmacol., The Univ. of North Carolina at Chapel Hill, Chapel Hill, NC; [3]Dept. of Pharmacol., The Univ. of North Carolina at Chapel Hill, Stanford, CA; [4]Dept. of Biol., [5]Dept. of Applied Physics, [6]Howard Hughes Med. Inst., [7]CNC Program, [8]Dept. of Bioengineering, Stanford Univ., Stanford, CA; [9]Chan Zuckerberg Biohub, San Francisco, CA; [10]New York Stem Cell Fndn. – Robertson Investigator, New York, NY

Abstract: Pain is a complex experience with sensory, emotional and cognitive dimensions. We recently reported the discovery of a discrete ensemble of neurons in the basolateral amygdala (BLA) that encodes, and is causally responsible for, pain negative emotions (*i.e.*, the unpleasant quality of pain) (Corder, Ahanonu et al., Science, 2019). To determine the molecular identity of nociceptive amygdalar neurons, we profiled the gene expression of individual neurons active during nociception using single-cell RNA-sequencing (scRNA-seq). We labeled active neurons using the TRAP2;Ai14 reporter system, in which induction of the immediate-early gene *c-Fos* results in tdTomato expression in neurons during administration of 4-OHT. To generate activity in nociceptive circuits, we used an established model of pain, injecting the algogen formalin into the left hindpaw. Cells of the amygdala were dissociated and sorted into 384-well plates, and the

libraries were processed with SmartSeq2. From 13747 sequenced cells, we selected 9695 high-quality cells for further analysis. We distinguished neuronal (5092) from glial (4603) cells based on clustering and analysis of known markers *Snap25* and resolved GABAergic and glutamatergic subpopulations of neurons by expression of *Gad1* and *Gad2 versus Slc17a7* and/or *Slc17a6,* respectively. We classified 17 major neuron types and 28 subtypes of amygdalar neurons. Mapping the spatially resolved patterns with which the marker genes defining cell types are organized establishes a comprehensive amygdalar neuron atlas.
Next, we integrated our scRNA-seq results with published literature regarding the function and cell types of amygdalar neurons to guide the discovery of amygdalar analgesic drugs. For example, we identified *Rspo2*-expressing BLA neurons that serve well-established functions in negative valence-related behaviors. We initially focused our search for therapeutic targets on enriched G protein-coupled receptors (GPCRs), which are highly druggable biological targets. We have tested agents engaging these GPCR targets in several pain assays to determine their potential clinical utility. Our data suggest that several of the target GPCRs indeed show antinociceptive activity. Considering our prediction that the tested drugs modulate multiple pain-related pathways, we derived a combination strategy of three drugs that synergize to enhance analgesia. Together, our findings establish the molecular structure of the nociceptive amygdala and identify highly druggable targets for the development of analgesics against pain unpleasantness across pain types.

Disclosures: D. Berg: None. **D. Lee:** None. **J. Krzeski:** None. **M. Chen:** None. **X. Jiang:** None. **Y. Ke:** None. **S. Quake:** None. **M.J. Schnitzer:** None. **G. Scherrer:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.22

Topic: D.02. Somatosensation – Pain

Support: NIH F32DE030003
1R01NS118504
New York Stem Cell Foundation – Robertson Investigator.

Title: Transcranial magnetic stimulation of the motor cortex requires endogenous opioid signaling in the rostral ventromedial medulla to generate analgesia

Authors: *N. MERCER LINDSAY[1,3], T. M. BAER[2], M. J. SCHNITZER[4,5], G. SCHERRER[3,6]; [1]Dept. Biol., [2]Stanford Photonics Res. Ctr., Stanford Univ., Stanford, CA; [3]Dept Cell Biol. and Physiol., The Univ. of North Carolina Chapel Hill, Chapel Hill, NC; [4]Depts. Biol. & Applied Physics, Stanford Univ. Dept. of Biol., Stanford, CA; [5]Howard Hughes Med. Inst., Stanford, CA; [6]New York Stem Cell Fndn. – Robertson Investigator, Chapel Hill, NC

Abstract: Motor cortex stimulation (MCS) reduces pain experience in chronic pain patients. However, how motor cortex modulates activity in pain circuits to induce analgesia is not known. Here, we engineered a miniature transcranial magnetic stimulation (TMS) device that models the human subjects' most commonly used non-invasive stimulation technique to investigate MCS-induced analgesia mechanisms in mice. Similar to TMS devices used in the clinic, this device induces a magnetic field of ~1 Tesla by passing an electric current through an electromagnetic coil. We tested this TMS device in a mouse model of trigeminal neuropathic pain induced by the constriction of the infraorbital nerve. Strikingly, we observed that TMS targeting the mouse motor cortex decreased reflexive and affective-motivational pain behaviors in a dose-dependent manner for up to two weeks. Furthermore, subcutaneous injection of naloxone, a nonselective opioid receptor antagonist, prior to TMS of the motor cortex blocked MCS-induced analgesia. This result suggested that MCS relies on endogenous opioid signaling, either during or after stimulation, to reduce pain. Thus, we next used viral and genetic tools to identify candidate brain regions activated during MCS, focusing on several pain experience-related areas such as the amygdala, periaqueductal gray (PAG), and rostral ventromedial medulla (RVM). Our data, alongside fMRI studies in human subjects, suggested that descending pain control pathways might contribute to MCS antinociception. To determine whether opioidergic descending pain control pathways are required for MCS to generate analgesia, we injected naloxone intracranially into the RVM before MCS treatment. Mice injected with naloxone but not vehicle into the RVM showed no pain reduction, suggesting endogenous opioid signaling in the RVM mediates MCS antinociception. Together, this research describes a new device to model TMS in rodents and establishes the molecular and circuit mechanisms by which MCS generates analgesia. Collectively, these approaches provide a promising route to developing optimized neurostimulation protocols for treating chronic pain patients.

Disclosures: N. Mercer Lindsay: None. **T.M. Baer:** None. **M.J. Schnitzer:** None. **G. Scherrer:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.23

Topic: D.02. Somatosensation – Pain

Support: NSERC

Title: The social approach toward pain is controlled by the midcingulate cortex and opioid receptors in mice

Authors: N. LIDHAR, S. KHAN, A. MUTASA, C. MUI, ***L. MARTIN**;
Univ. of Toronto, Mississauga, ON, Canada

Abstract: The observation of another undergoing a painful or stressful experience elicits emotional and physiological events that are detectable by analyzing general neural activation in the cingulate cortex and requires neural activity in insular neurons to elicit approach behavior. At a neurophysiological level, the endogenous opioid receptor system plays an important role in attention to social visual cues, endogenous pain modulation and pain perception; however, the role of opioid receptors in modulating empathetic or social approach behavior has not well characterized. Here, we examined the role of mu (MOR), delta (DOR), and kappa opioid receptors (KOR) in modulating social approach behaviors towards a sibling in pain. Systemic blockade of opioid receptors (MOR, DOR and KOR) all reversed social preference for a female sibling in pain. C-fos expression was elevated in the midcingulate cortex (MCC) of females interacting with a sibling in pain compared to controls. Treatment with a MOR, DOR or KOR antagonist also reduced c-fos in the MCC. Inactivation of the ACC resulted in reversed approach behaviour in female siblings, indicating that the ACC is necessary for social approach to a sibling in pain. Together these findings point towards distinct modulation of pain-related behaviours due to social context.

Disclosures: **N. Lidhar:** None. **S. Khan:** None. **A. Mutasa:** None. **C. Mui:** None. **L. Martin:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.24

Topic: D.02. Somatosensation – Pain

Support: NSERC
Louise and Alan Edwards Foundation

Title: Chemogenetic inhibition of anterior insula attenuates pain-induced facial grimacing

Authors: A. ZUMBUSCH[1], M. VALYEAR[2], L. ABDUL-REDA[2], T. BROGARD[2], J. S. MOGIL[2];
[1]Psychology, McGIll Univ., Montréal, QC, Canada; [2]McGill Univ., Montréal, QC, Canada

Abstract: Introduction: Lesion studies show that ablating the anterior insula attenuated facial expressions of pain as measured by the Mouse Grimace Scale (MGS), while leaving the intensity of pain unchanged as measured via reflexive behaviour. Our aim was to replicate this finding using chemogenetics, in service of a broader goal of mapping the neuroanatomical and neurochemical basis of grimacing behaviour and pain affect.
Methods: We expressed inhibitory designer receptors [designer receptors exclusively activated by designer drugs (DREADDs)] in the anterior insula of adult male and female CD-1 mice. After 4-6 weeks of viral transfection, we evaluated pain behaviour in these mice using the acetic acid (AA) abdominal constriction test Abdominal constrictions (i.e., writhing behaviour) and facial

grimacing (via the MGS) were captured using videos collected for 30 minutes both before (baseline) and immediately after an intraperitoneal (i.p.) injection of 10 ml/kg of 0.9% acetic acid immediately followed by either the DREADD-activating ligand clozapine-N-oxide (CNO+AA) or vehicle solution (VEH+AA). Results: Mice that received CNO+AA showed significantly attenuated grimacing compared to mice that received VEH+AA. There was no such difference in abdominal constrictions (writhing).
Conclusions: Chemogenetically inhibiting the insula to control the affective component of pain is a crucial step in illuminating circuit-type and cell-type specificity in different aspects of the pain experience. Ongoing research using Cre lines will allow us to identify with specificity and directionality, a proposed pain network wherein we can map the circuitry involved in different aspects of the pain experience.

Disclosures: A. Zumbusch: None. **M. Valyear:** None. **L. Abdul-Reda:** None. **T. Brogard:** None. **J.S. Mogil:** None.

Poster

540. CNS Pain and Touch Mechanisms in Preclinical Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 540.25

Topic: D.02. Somatosensation – Pain

Title: Role of orexin receptors within the dentate gyrus in antinociception induced by chemical stimulation of the lateral hypothalamus in animal models of acute and persistent inflammatory pain

Authors: *A. HAGHPARAST, B. RASOULI, M. SHAREGHI BROJENI, M. RASHVAND; Neurosci. Res. Ctr., Neurosci. Res. Center, Shahid Beheshti Univ. of Med. Sci., Tehran, Iran, Islamic Republic of

Abstract: Pain is a complex experience consisting of sensory, affective-motivational, and cognitive dimensions. Hence, identifying the multiple neural pathways subserving these functional aspects is a valuable task. The role of the dentate gyrus (DG) as a relay station of neocortical afferents in the hippocampal formation (HF) in the mediation of antinociceptive responses induced by lateral hypothalamus (LH)-stimulation in different animal models of pain is still a matter of controversy. Adult male Wistar rats weighing 220-250 g were unilaterally implanted with two separate cannulae into the LH and DG. Intra-DG administration of the orexin-1 receptor (OX1R) antagonist, SB334867, or the orexin-2 receptor (OX2R) antagonist TCS OX2 29 was performed just 5 min before intra-LH carbachol microinjection. Animals then underwent the tail-flick test as a model of acute pain and the formalin test using injection into the plantar surface of the hind paw as a model of persistent pain. The results showed that OX1R and OX2R antagonists dose-dependently decreased antinociceptive effects of carbachol in both tail-flick and formalin tests. In addition, the results obtained from the tail-flick test demonstrated the more prominent role of OX1R in the DG in carbachol-induced antinociception compared to that

of OX2Rs in this region. According to the formalin test results, the preventive effect of SB334867 or TCS OX2 29 on carbachol-induced antinociception was approximately equal in both early and late phases of formalin nociception. Pain modulatory role of the orexinergic system through a neural pathway from the LH to DG region suggests an alternative approach to developing more efficient therapeutic agents in the clinical setting.

Disclosures: A. Haghparast: None. **B. Rasouli:** None. **M. Shareghi Brojeni:** None. **M. Rashvand:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.01

Topic: D.02. Somatosensation – Pain

Support: Brain Research UK

Title: The stress regulator FKBP51 drives the exacerbation of inflammation-induced hypersensitivity by prior stress exposure

Authors: *O. B. MORGAN[1], F. HAUSCH[2], A. ANDREOU[3], S. M. GÉRANTON[1];
[1]CDB, Univ. Col. London, London, United Kingdom; [2]Max Planck Inst. of Psychiatry, Munich, Germany; [3]Wolfson Ctr. for Age-Related Dis., King's Col. London, London, United Kingdom

Abstract: *Objectives & rationale*
Stressful life events are known to exacerbate pain states but the underlying mechanisms linking pain with stress remain inadequately understood. A common regulator of stress and pain is the FK 506 binding protein 51 (FKBP51). FKBP51 modulates the glucocorticoid receptor (GR) sensitivity and is therefore important for the regulation of the stress response. Recently our lab has shown that FKBP51 is expressed in rodent central pain circuits and plays a key role in the development of chronic pain states, with mice lacking the protein FKBP51 being less hypersensitive after hind-limb joint inflammation or nerve damage than wild-type controls. Here, we explored whether stress in adulthood could exacerbate inflammation-induced hypersensitivity, hypothesising that FKBP51 contributes to the underlying mechanisms of the comorbidity between stress and pain.
Methods
Adult male C57BL6 mice were exposed to either restraint stress alone (1h per day for 3 consecutive days), or restraint stress and subsequent hind-limb inflammation, induced by intra-plantar injection of the inflammatory agent Complete Freund's Adjuvant (CFA). Mechanical thresholds were assessed using Von Frey filaments. Blood, brains and spinal cords of experimental mice were collected to investigate molecular mechanisms using ELISA, RT-qPCR, IHC, RNAscope and DNA methylation arrays.
Results & conclusion

Restraint stress alone induced mechanical hypersensitivity in the hind-limb in C57BL6 WT male mice, which resolved approximately one week after the end of the restraint and was accompanied by an increase in blood corticosterone levels and spinal upregulation of stress-related genes, including *FKBP5*.
Furthermore, restraint stress induced a latent state of hypersensitivity, with prior stress leading to an increase in duration of hyperalgesia following hind-paw inflammation compared to inflammation alone. Prior stress also led to changes in central pain circuits as it increased inflammation-induced spinal expression of (1) the immediate early gene, cFos, and (2) *FKBP5*.
Crucially, blocking FKBP51 during the restraint stress period, using the specific FKBP51-inhibitor SAFit2, did not alter stress induced hypersensitivity, but prevented the exacerbation of inflammation-induced hypersensitivity.
Ongoing DNA methylation analysis focusing on stress-induced changes to the *FKBP5* gene methylome are likely to reveal a role for epigenetic alterations in this form of priming.
In conclusion, this study provides further understanding of the interactions between stress and pain and strengthens our hypothesis that FKBP51 drives the exacerbation of pain states by stress.

Disclosures: O.B. Morgan: None. **F. Hausch:** None. **A. Andreou:** None. **S.M. Géranton:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.02

Topic: D.02. Somatosensation – Pain

Support: CIC UMSNH 26.10
CIC UMSNH 30.2

Title: Coadministration of metformin and carbamazepine in rat formalin test

Authors: *L. F. ORTEGA-VARELA[1], E. BENITEZ-FAJARDO[2], J. G. TORRES-ALVARADO[2], C. J. GUTIERREZ-GARCIA[4], D. GODINEZ-HERNÁNDEZ[3], M. Y. GAUTHEREAU-TORRES[5];
[1]Univ. Michoacana de San Nicolas de Hidalgo, Salud Publica. Univ. Michoacana Sn Nicolas De H ., Morelia, Mexico; [2]Facultad de Quimico Farmacobiologia, [3]IIQB, Univ. Michoacana de San Nicolas de Hidalgo, Morelia, Mexico; [4]Dept. de Ingeniería química y Bioquímica, Tecnológico Nacional de México/Instituto Tecnológico de Morelia, Morelia, Mexico; [5]Facultad de Ciencias Medicas y Biologicas Dr Ignacio Chavez, Univ. Michoacana De San Nicolas De Hidalgo, Morelia, Mexico

Abstract: Combination therapy is a pharmacotherapeutic strategy for the treatment of various clinical disorders, especially pain, which remains as a public health challenge. This study was achieved to assess the interaction between metformin and carbamazepine orally administered in

the rat model of formalin test. Female Wistar rats (220-300 g) were injected into the dorsal surface of the right hind paw with 50 μ g of 1% formalin. This substance induced a flinching pain-related behavior, the reduction of such behavior is considered as antinociception. The percent of antinociceptive effect was determined by the oral administration of metformin (30-1000 mg/kg), topiramate (10-300 mg/kg), and their combination. To analyze the nature of the interaction, isobolographic analysis was used in a fixed-dose radio combination (0.5:0.5), on the basis of their ED50 values: metformin (908.63 ± 280.87 mg/kg) and carbamazepine (198.94 ± 56.01 mg/kg). The metformin- carbamazepine combination significantly reduced the number of flinches in the second phase of the formalin test. For the isobolographic analyses, the theoretical effective dose 50 for the combination (ED50 T) was 553.79 ±143.21mg/kg. Experimentally, the effective dose 50 (ED50 E) was significantly lower (460.58 ± 50.57mg/kg), indicating the presence of synergism for the combination. Results show that the oral coadministration of metformin and carbamazepine can interact synergistically and could provide a therapeutic alternative for inflammatory pain.

Disclosures: L.F. Ortega-Varela: None. **E. Benitez-Fajardo:** None. **J.G. Torres-Alvarado:** None. **C.J. Gutierrez-Garcia:** None. **D. Godinez-Hernández:** None. **M.Y. Gauthereau-Torres:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.03

Topic: D.02. Somatosensation – Pain

Support: SIP-20220300 (MDC)

Title: Antinociception is increased by melatonin in combination with N-palmitoylethanolamide or paracetamol

Authors: *M. DECIGA-CAMPOS[1], M. Y. RIOS[2], R. VENTURA-MARTÍNEZ[3], G. E. ÁNGELES-LÓPEZ[3];
[1]Escuela Superior de Medicina IPN, Inst. Politécnico Nacional, Mexico City, Mexico; [2]Ctr. de Investigaciones Químicas, Univ. Autónoma del Estado de Morelos, Morelos, Mexico; [3]Facultad de Medicina, Univ. Autónoma de México, Mexico City, Mexico

Abstract: Inflammatory pain is the most common pain affecting the quality of life and imposing a significant economic burden. Thus, novel therapeutic strategies are required. Melatonin has been shown to have an antinociceptive effect. Their administration, together with other drugs, could enhance the antinociceptive effect. This study assessed the antinociceptive effects of melatonin in combination with paracetamol and N-palmitoylethanolamide (PEA). In this study, we used the formalin test to determine the antinociceptive effects in mice. Melatonin, paracetamol, and PEA were administered intraplantar (paw) alone or combined to mice. A

concentration-response curve was generated to determine the concentration needed to reach 30% of the maximal antinociceptive effect (EC_{30}). Melatonin, paracetamol and PEA induced a concentration-dependent antinociceptive effect in both phases of the formalin test, being PEA more potent (EC_{30} = 7.4 ± 0.2 μg/paw) than melatonin (EC_{30} = 20.5 ± 3.1 μg/paw) or paracetamol (EC_{30} = 41.8 ± 2.6 μg/paw). Melatonin coadministered with paracetamol or PEA induced antinociceptive effect concentration-dependent. Isobolographic analysis showed that melatonin combinations with paracetamol or PEA induced a synergistic interaction. The experimental values of EC_{30} were significantly smaller than those calculated theoretically. Hence we concluded from this study that combined melatonin and paracetamol or PEA produced synergistic antinociception that may be helpful in facilitating clinical management of acute nociceptive pain.

Disclosures: M. Deciga-Campos: None. **M.Y. Rios:** None. **R. Ventura-Martínez:** None. **G.E. Ángeles-López:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.04

Topic: D.02. Somatosensation – Pain

Title: Evaluation of the antinociceptive effect of Snow mountain garlic

Authors: M. T. G. CHÁVEZ-LÓPEZ[1], J. R. ZAPATA-MORALES[3], Y. TERÁN-FIGUEROA[1], I. CLARCK-MONTOYA[2], O. A. JARAMILLO-MORALES[4], *J. V. ESPINOSA-JUAREZ[5];
[1]Enfermería y Nutrición, [2]. Doctorado en Ciencias Farmacobiológicas. Facultad de Ciencias Químicas, Univ. Autónoma de San Luis Potosí, San Luis Potosí, Mexico; [3]Farmacia, Univ. de Guanajuato, Guanajuato, Mexico; [4]Dept. de Enfermería y Obstetricia, Univ. de Guanajuato, Ciudad de México, Mexico; [5]Escuela de Ciencias Químicas, Univ. Autonoma de Chiapas, Chiapas, Mexico

Abstract: Snow mountain garlic (SMG) has promising medicinal properties and has shown some degree of remedial effects against hypertension, arteriosclerosis, diabetes, cancer, and immunomodulatory activity. However, there is no scientific evidence to corroborate its analgesic effect. The objective of the present study was to evaluate the antinociceptive effect of lyophilized Snow mountain garlic in a pain model. Methods. The vegetal material was obtained from a local market. An aqueous extract of garlic was obtained by crushing it and the extract obtained was lyophilized, and resuspension in a known volume of water was used for evaluation of the antinociceptive effect. The antinociceptive activity was evaluated in rats in which nociception was induced intraplantar with 5% formalin. Flinching nocifensive behavior was counted in both phases induced-formalin injection, neurogenic and inflammatory. Results. The intragastric administration of aqueous extract lyophilized SMG (100 mg/kg) showed (an antinociceptive

effect) and significantly decreased the flinching experimental values in the inflammatory phase. Conclusión. Our results indicate for the first time the antinociceptive activity of aqueous extract lyophilized SMG in inflammatory pain.

Disclosures: M.T.G. Chávez-López: None. **J.R. Zapata-Morales:** None. **Y. Terán-Figueroa:** None. **I. Clarck-Montoya:** None. **O.A. Jaramillo-Morales:** None. **J.V. Espinosa-juarez:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.05

Topic: D.02. Somatosensation – Pain

Support: JSPS KAKENHI, grant number 20K09510

Title: Vacuum Phenomenon in intervertebral discs was associated with upregulation of pain-related molecules, might lead to discogenic low back pain.

Authors: *M. MIYAGI[1], K. UCHIDA[1], S. INOUE[1], A. KURODA[1], E. SHIRASAWA[1], Y. YOKOZEKI[1], N. HIROSAWA[2], G. INOUE[1], M. TAKASO[1];
[1]Kitasato Univ., Kitasato Univ., Sagamihara City/Kanagawa Prefecture, Japan; [2]Chiba Univ., Chiba, Japan

Abstract: Objectives: Vacuum phenomenon (VP) in degenerated intervertebral discs (IVDs) was reported to be associated with lumbar spinal instability, potentially leads to low back pain (LBP). This study aimed to elucidate the relationships among VP, clinical findings, and pain-related-molecule expression in human degenerated IVDs. Methods: Degenerated-IVD samples from 35 patients (23 men and 12 women) including 19 with lumbar spinal stenosis, 10 with adult spinal deformity, and 6 with IVD herniation during spinal interbody fusion surgery were harvested. Harvested IVD-derived mononuclear cells were obtained and pain-related molecules, including tumor necrosis factor alpha (TNF-alpha), interleukin (IL)-6, calcitonin gene-related peptide (CGRP), microsomal prostaglandin E synthase-1 (mPGES1), and nerve growth factor (NGF), were determined, using real-time polymerase chain reaction (RT-PCR). We also recorded preoperative clinical findings, including Oswestry Disability Index (ODI) and visual analogue scale (VAS) of LBP, and the presence of VP in computed tomography scan images. Further, we compared pain-related molecules expression and clinical findings between the VP (-) and (+) groups, and evaluated the correlations among clinical finding and each pain-related molecules expression.Results: In the VP (+) group, mPGES-1 levels were significantly higher than in the VP (-) group. Additionally, mPGES-1 expression was significantly correlated with CGRP and NGF expression ($r=0.56$, $r=0.39$;$p<0.05$). In addition, NGF expression was significantly correlated with TNF-alpha and IL-6 expression ($r=0.88$, $r=0.42$;$p<0.05$). However, clinical findings including ODI and VAS of LBP were not associated with the presence of VP and any

pain-related molecules expression.Conclusion: Because the patho-mechanism of LBP might be multifactorial, LBP scores were not associated the presence of VP as well as any pain-related molecules expression in the present study. On the other hands, the presence of VP might be associated with the up-regulation of mPGES1 in IVDs. mPGES1 potentially leads to the expression of CGRP and NGF, and NGF also leads to the expression of inflammatory cytokines in human degenerated IVDs, potentially leading to chronic discogenic LBP.

Disclosures: **M. Miyagi:** None. **K. Uchida:** None. **S. Inoue:** None. **A. Kuroda:** None. **E. Shirasawa:** None. **Y. Yokozeki:** None. **N. Hirosawa:** None. **G. Inoue:** None. **M. Takaso:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.06

Topic: D.02. Somatosensation – Pain

Support: NIH Grant R01NS100788
NIH Grant R01NS114018

Title: Inhibition of nonsense-mediated decay causes hyperalgesic priming in mice

Authors: J. DE LA PENA[1], R. CHASE[2], N. KUNDER[2], T.-F. LOU[2], P. SURESH[2], A. STANOWICK[2], **T. SHUKLA**[1], Z. CAMPBELL[1];
[1]Univ. of Wisconsin, Madison, Madison, WI; [2]UT Dallas, Richardson, TX

Abstract: RNA stability permeates neurobiology. Yet, the contribution of a dominant pathway of RNA metabolism termed nonsense-mediated decay (NMD) in pain is unclear. NMD safeguards against translation of mRNAs that harbor premature termination codons. It also controls the stability of roughly 10% of typical protein-coding mRNAs. NMD hinges on the activity of a conserved kinase designated SMG-1. We sought to determine the role of SMG-1 and NMD in pain. We found that SMG-1 is expressed in sensory neurons throughout the DRG. We identified SMG-1 targets in DRG neurons using high-throughput sequencing. We analyzed multiple features present in mRNA to establish the characteristics of NMD substrates in DRG neurons. The most dominant feature was structural content in the 3'UTR. This implies that factors that bind to structured motifs likely recruit NMD machinery to specific mRNAs. We identify numerous NMD targets linked to pain including components of the integrated stress response such as ATF-4. We found that inhibition of SMG-1 with a small molecule results in hyperalgesic priming in mice. Given that ATF-4 is targeted by NMD, we next asked if priming was due to activation of the integrated stress response. Indeed, a small molecule inhibitor of the integrated stress response called ISRIB prevented priming triggered by inhibition of NMD. Collectively, our results indicate that (i) inhibition of NMD is pro-nociceptive, (ii) priming induced by NMD blockade requires the integrated stress response, (iii) and structure is a defining

feature of NMD targets. This work establishes a clear link between NMD and pain-associated behaviors in mice.

Disclosures: J. de la Pena: None. **R. Chase:** None. **N. Kunder:** None. **T. Lou:** None. **P. Suresh:** None. **A. Stanowick:** None. **T. Shukla:** None. **Z. Campbell:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.07

Topic: D.02. Somatosensation – Pain

Title: The neurochemical mediation of conditioned hyperalgesia in mice.

Authors: *A. SKVORTSOVA[1], L. VASCONCELOS LIMA[1], S. CARRIER[2], L. LIANG[1], R. CONTOFALSKY[1], J. S. MOGIL[3];
[1]Psychology, McGill Univ., Montreal, QC, Canada; [2]Psychology, Univ. du Québec à Trois-Rivières, Trois-Rivières, QC, Canada; [3]Psychology, McGill Univ. Integrated Program in Neurosci., Montreal, QC, Canada

Abstract: Introduction/Aim. Conditioned hyperalgesia has been found in male (but not female) mice and humans after a single pairing of an environmental context with pain. The aim of this study was to investigate the neurochemical mediation of this phenomenon with a focus on cholecystokinin (CCK), a neurotransmitter that has previously been demonstrated to affect nocebo hyperalgesia in humans. **Methods.** A conditioning paradigm with one training and one test day was used. On the training day, following the determination of thermal pain sensitivity (radiant heat paw-withdrawal test), a tonic noxious stimulus (intraperitoneal injection of 0.9% acetic acid; 10 ml/kg) was paired with a particular testing context. On the test day, mice were re-tested for thermal pain sensitivity either in the same or in a different context. Mice were randomly assigned to one of the following drug conditions: nonspecific CCK receptor antagonist proglumide (50 mg/kg); CCK-B antagonist LY-225910 (1 mg/kg); CCK-A antagonist loxiglumide (10 mg/kg) or saline. Drugs were administered immediately before the tonic stimulus on the training day and at the start of the test day. **Results.** A significant increase in pain sensitivity was observed in saline-treated male but not female mice tested in the same context on the test day. This conditioned hyperalgesia was blocked by LY-225910 and proglumide, but not affected by loxiglumide. All drugs had no effect on pain in female mice.
Discussion/Conclusions. LY-225910 and proglumide block conditioned hyperalgesia in male mice, indicating that CCK-B and not CCK-A receptors are involved in mediating this phenomenon. CCK-B receptors are distributed primarily in the central nervous system and CCK-B has various psychophysiological functions, such as regulation of anxiety and memory. Ongoing studies are attempting to elucidate whether the effect of CCK on conditioned hyperalgesia are mediated by stress or memory.

Disclosures: **A. Skvortsova:** None. **L. Vasconcelos Lima:** None. **S. Carrier:** None. **L. Liang:** None. **R. Contofalsky:** None. **J.S. Mogil:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.08

Topic: D.02. Somatosensation – Pain

Support: Academy of Finland
Medical Society of Finland
International Association for the Study of Pain

Title: Characterization of the effects of spared nerve injury on pain and sleep architecture in mice

Authors: *T. KILPELÄINEN[1], W. DAI[1], E. A. KALSO[2], V. PALADA[3];
[1]Dept. of Physiol., Univ. of Helsinki, Helsinki, Finland; [2]Pharmacol., [3]Fac. of Med., Helsinki Univ., Helsinki, Finland

Abstract: The relationship between sleep and pain is bidirectional since insufficient sleep can precede and exacerbate pain while pain can disturb the quality of sleep [1]. It has been estimated that up to 68% of patients with chronic neuropathic pain (NP) suffered from severe sleep disturbance [2]. However, the effects of chronic NP on the sleep architecture remain poorly understood. In this study, we wanted to assess the effects of well characterized spared nerve injury (SNI) mouse model of NP on sleep-wake cycle using telemetric electroencephalography (EEG). Experiments were performed for control sham, SNI and EEG+SNI C57BL female and male mice (n=3 animals/gender/group). EEG-recordings were done at the baseline and 7, 14 and 21 days after the ligation of tibial and peroneal nerves. Spike 2 was used for sleep scoring to quantify the duration of wakefulness, REM and non-REM sleep before and after SNI. Mechanical and thermal hyperalgesia were assessed using Von Frey filament test, dynamic test, acetone test and hot and cold plate tests. Behavioral testing was conducted at the baseline and 3, 7, 14, 21 days after SNI. After 24 days the mice were sacrificed and prefrontal cortex, hypothalamus, periaqueductal grey, dorsal root ganglions (L3-L5) and lumbar spinal cord were collected for qPCR analysis. The SNI and EEG+SNI groups showed significantly reduced mechanical pain thresholds 7 days after nerve ligation compared to sham operated mice. EEG+SNI females showed cold hyperalgesia on the acetone test 7 days after SNI as well as SNI males with reduced latency at the cold plate test. EEG recordings showed some changes in sleep architecture 7 days after SNI compared to baseline. Both female and male mice had decreased amount of REM sleep and increased amount of wakefulness during the 36 h recording compared to their baseline. Interestingly, decreased REM sleep in male was pronounced only during the dark time, while female had decreased amount of REM sleep throughout the recording period. In conclusion our results provide a detailed insight into the effects of neuropathic pain on sleep-

wake cycle in mice and might be highly relevant to better understand the mechanisms behind the comorbidity between sleep and pain.
1. Palada V, Gilron I, Canlon B, Svensson CI, Kalso E. The circadian clock at the intercept of sleep and pain. Pain. 2020
2. Lampl C, Schweiger C, Haider B, Lechner A. Pregabalin as mono- or add-on therapy for patients with refractory chronic neuropathic pain: a post-marketing prescription-event monitoring study. J Neurol. 2010

Disclosures: T. Kilpeläinen: None. **W. Dai:** None. **E.A. Kalso:** None. **V. Palada:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.09

Topic: D.02. Somatosensation – Pain

Support: JUST 2021/53

Title: Evaluation of sucrose induced antinociception on long term effects of neonatal noxious stimulation in rats

Authors: *K. NUSEIR[1], A. ALTARIFI[2], K. ALZOUBI[1];
[1]Clin. Pharm., [2]Pharmacol., Jordan Univ. of Sci. and Technol., Irbid, Jordan

Abstract: Pain during infancy have dire consequence both short and long term. Procedural pain is induced as tests and treatments are needed when premature and mature babies are admitted to NICU. Sadly, pain management is either not available or not adequate. Sweet solutions have been shown to decrease some of this pain, as well as prevent negative sequalae of neonatal pain. Mechanism of sweet solution induced pain relief is largely unknown yet. Previous research from our lab had shown a comparable antinociceptive effect of sucrose solution to common analgesics. This work is an extension of our previous research to explore the effects of sucrose solution on Sprague Dawley rats; males and females rats were included. Induced repetitive nociceptive stimulation during infancy model was employed; briefly, a small needle was inserted and removed quickly in rat pups' paws during their first two weeks of life. Sucrose or saline were applied to the tongues of these pups just before noxious stimulation. At adulthood, a battery of behavioral tests was run in sequences to explore deficiencies and inadequacies that result from this nociceptive stimulation. By employing both male and female rats we aimed to examine sex differences if any. Biochemical markers were tested to add to our knowledge of the mechanisms of sweet induced analgesia.

Disclosures: K. Nuseir: None. **A. Altarifi:** None. **K. Alzoubi:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.10

Topic: D.02. Somatosensation – Pain

Support: ANID-FONDECYT 1211082
NIH grants R01AA025718
Millennium Nucleus for the Study of Pain (MiNuSPain). MiNuSPain is supported by the Millennium Scientific Initiative NCN19_038 of the Ministry of Science, Technology, Knowledge and Innovation, Chile

Title: Assessment of nociception and chronic pain in a glycine receptor alpha1 subunit knock-in mouse model

Authors: *V. P. SAN MARTÍN[1,2], V. PÉREZ[1,2], A. M. MARILEO[1,2], C. O. LARA[1,2], Á. SAZO[1,2], J. FUENTEALBA[1], G. MORAGA[1], L. G. AGUAYO[1], G. E. YEVENES[1,2];
[1]Dept. of Physiology, Fac. of Biol. Sciences, Univ. of Concepcion, Concepcion, Chile;
[2]Millennium Nucleus for the Study of Pain (MiNuSPain), Santiago, Chile

Abstract: Glycine receptors (GlyRs) are pentameric chloride-permeable ion channels. GlyRs are key regulators of motor activity, muscle tone, sensory processing, and pain control. Previous studies using chronic pain models have shown alterations of the glycinergic inhibition at the level of the dorsal horn. These studies, for example, have shown a critical role of the alpha3 GlyR subunit on the maintenance of chronic inflammatory pain. However, the role of the alpha1 GlyR subunit in chronic pain has not been yet established. Using a genetically modified mouse line carrying two-point mutations within the large intracellular domain of the alpha1 GlyR which render the receptor resistant to modulation by G protein βγ subunits (i.e. alpha1 GlyRKK385-386AA mice), we explore whether this signaling pathway have a role on sensory processing and on chronic pain. To this end, we performed behavioral studies in male and female wild-type and alpha1 GlyRKK385-386AA mice. We first analyzed the baseline nociception by measuring thermal and mechanical thresholds with Hargreaves test and Von Frey filaments, respectively. Both assays showed no differences between wild-type and alpha1 GlyRKK385-386AA mice ($P=0.3726$ (mechanical) and $P=0.7466$ (thermal), male mice). In addition, both mice groups displayed an undistinguishable response to noxious mechanical stimulation ($P>0.9999$, male mice). We next used the chronic constriction injury (CCI) of the sciatic nerve as a model of neuropathic pain and the Complete Freund Adjuvant (CFA) model to induce peripheral inflammation. The subcutaneous injection of CFA into the left hind paw showed a differential time course of pain sensitization between wild-type and alpha1 GlyRKK385-386AA mice. Interestingly, both male and female alpha1 GlyRKK385-386AA mice displayed a more sustained mechanical pain hypersensitivity ($P< 0.0001$, male mice). On the other hand, the time course of the neuropathic sensitization elicited by the sciatic ligation showed no significant differences between both mice groups ($P= 0.0884$, male mice). Altogether, our findings suggest that the regulation of alpha1 GlyR by G protein βγ plays a role in the maintenance of chronic

inflammatory pain. On the other hand, this signaling pathway appears to be not related with acute nociception or with chronic neuropathic pain.

Disclosures: **V.P. San Martín:** None. **V. Pérez:** None. **A.M. Marileo:** None. **C.O. Lara:** None. **Á. Sazo:** None. **J. Fuentealba:** None. **G. Moraga:** None. **L.G. Aguayo:** None. **G.E. Yevenes:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.11

Topic: D.02. Somatosensation – Pain

Support: UAB start up funds to WRR

Title: Role of inflammatory markers in a nerve growth factor-induced low back pain model.

Authors: *C. LIMA, P. LI, W. REED;
Univ. of Alabama At Birmingham, Birmingham, AL

Abstract: Low back pain (LBP) is a global health concern with approximately 85-95% of the cases lacking an identifiable pathological etiology. Approximately 25% of acute cases of LBP progress into chronic LBP making it the leading cause of years lived with disability. Unresolved inflammation within soft tissues is among the reported contributors to the transition from acute to chronic musculoskeletal pain via excitation of nociceptors and tissue acidosis. Additionally, nerve growth factor (NGF) has been proposed to contribute to the pathophysiology of chronic pain through indirect modulation of inflammatory cytokines. **Purpose:** To determine pro- and anti-inflammatory changes in the CSF induced by NGF intramuscular injections in an animal model of muscular LBP. **Methods:** Female Sprague Dawley rats (N=13; 212.3-266.1 g) were unilaterally injected with either NGF (50 µl; 2.0 µM) or phosphate buffer saline (50µl; control) on Days 0 and 5 in the left multifidus muscle under isoflurane anesthesia (1-2%/2L O_2). On Day 17, animals (n=6-7/group) were anesthetized, and CSF collected via cisterna magna puncture. Samples were stored at −80°C until assay analysis was performed. Frozen, undiluted samples containing a minimum of 75 uL were analyzed in duplicates via the rat cytokine/chemokine 27-Plex Discovery Assay (RD27 - Eve Technologies®) and the mean values used to determine the concentration of 27 inflammatory markers. Differences in inflammatory marker concentration between experimental groups were determined by t-test and a value of $p < 0.05$ was considered statistically significant. **Results:** No significant differences in cytokine/chemokine concentrations between groups were found at Day 17 ($p > 0.05$). **Conclusion:** These preliminary findings suggest a limited role of CSF cytokines and chemokines to the maintenance of muscular pain in this NGF-induced LBP model at later stages. In previous female rat studies using this same NGF LBP model, bilateral trunk mechanical pain was present by Day 7 and fully resolved by Day 21. One plausible explanation for the limited contribution of inflammatory markers in

this LBP model could be that CSF collection on Day 17 was beyond the timeframe of peak inflammation and nearing mechanical pain resolution. To better define their role, temporal effects of NGF on CSF and plasma cytokine/chemokine regulation in this NGF and other muscular models of LBP should be investigated to enhance our understanding of the pathophysiological mechanisms responsible for the transition from acute to chronic LBP.

Disclosures: C. Lima: None. **P. Li:** None. **W. Reed:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.12

Topic: D.02. Somatosensation – Pain

Support: NIH Grant R01NS100788
NIH Grant R01NS114018

Title: A conserved RNA-binding protein contributes to nociceptive pain

Authors: N. KUNDER[1], *J. DE LA PENA[2], **T. SHUKLA**[2], T.-F. LOU[1], R. CHASE[1], A. STANOWICK[1], B. J. BLACK[3], Z. CAMPBELL[2];
[1]Dept. of Biol. Sci., Univ. of Texas at Dallas, Richardson, TX; [2]Dept. of Anesthesiol., Univ. of Wisconsin-Madison, Madison, WI; [3]Dept. of Biomed. Engin., Univ. of Massachusetts Lowell, Lowell, MA

Abstract: Nociceptors rely on post-transcriptional modes of gene control. For example, noxious stimuli can induce rapid translation in nociceptors. In prior work, we examined translational efficiency on a genome-wide basis in DRG neurons using ribosome profiling. We found that a single motif was highly enriched in the 3'UTRs of preferentially translated mRNAs. It contains an AU-rich element bound by HuR. Here, we show that HuR is expressed broadly throughout the PNS. To determine the role of HuR in pain, we made use of a recently described small molecule inhibitor termed CMLD-2. It reduced spontaneous firing of mouse and hiPSC derived sensory neurons. Furthermore, inhibition of HuR in vivo with CMLD-2 blocks hyperalgesic priming. Elimination of HuR from sensory neurons similarly reduces hyperalgesic priming. Collectively, this work reveals HuR as a key factor involved in nociceptive plasticity.

Disclosures: N. Kunder: None. **J. De La Pena:** None. **T. Shukla:** None. **T. Lou:** None. **R. Chase:** None. **A. Stanowick:** None. **B.J. Black:** None. **Z. Campbell:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.13

Topic: D.02. Somatosensation – Pain

Support: BBSRC BB/V010344/1

Title: Elucidating inflammatory signaling mechanisms at ER-PM junctions in sensory neurons

Authors: C. PALFREY, N. GAMPER, ***S. SHAH**;
Univ. of Leeds, Leeds, United Kingdom

Abstract: Nociception is referred to as neural encoding of noxious signals, allowing this information to be detected and propagated from the periphery to higher centres. To allow this process to be managed efficiently, signals emanating from various painful stimuli must be tightly controlled and regulated to allow 'beneficial' pain sensation. An emerging mechanism for controlling specificity of pain signaling, are sensory signaling complexes assembled within vast areas of close apposition between the endoplasmic reticulum and plasma membrane (<30nm) known as ER-PM junctions. Examples of such signaling complexes in dorsal root ganglion neurons (DRG) include: (1) Anoctamin 1 (ANO1) with inositol triphosphate receptors (IP_3R) and (2) Ca^{2+}-release activated channel (CRAC) proteins STIM1 and Orai1 in inflammatory pain. We recently identified junctophillin 4 as an essential scaffold for the CRAC complex but with regards to ANO1-IP_3R coupling, it is currently unknown which linker or scaffolding proteins (if any) are involved with forming these junctional complexes. Extended Synaptotagmin 1 (Esyt1), a Ca^{2+}-dependent junctional protein known to reduce the distance between the ER and PM in a dynamic manner, is a binding partner of ANO1 and has been proposed to play a role in ANO1 trafficking to the PM. Therefore, we hypothesised that Esyt1 may be involved in ER-PM junction formation where ANO1-IP_3R complexes are formed. To this end, we performed immunohistochemistry in DRG and trigeminal ganglia (TG), which revealed high expression levels of Esyt1, mainly in small-diameter neurons. Proximity ligation assay (PLA) was also conducted to assess proximity between ANO1, IP_3R and Esyt1. There was a level of proximity between both ANO1-Esyt1 (1.62 ± 0.136 puncta per cell (ppc), n=65) and IP_3R1-Esyt1 (21.0 ± 2.68 ppc, n=82) under control conditions. Interestingly, an inflammatory mediator bradykinin (BK) significantly increased the number of puncta between ANO1-Esyt1 (11.2 ± 1.29 ppc, $p<0.0001$, n=93 neurons) however, the number of puncta between IP_3R-Esyt1 was significantly reduced (7.41 ± 0.678 ppc, $p<0.001$, n=75 neurons), indicating robust, stimulus-dependent rearrangements within the ANO1-containing signaling complex upon stimulation. Live cell total internal reflection fluorescence (TIRF) microscopy performed in DRG neurons overexpressing Esyt1-GFP revealed that BK, significantly increased the TIRF signal (0.11 ± 0.02 $\Delta F/F_0$, n=6) suggestive of ER movement towards the PM or Esyt1-GFP translocation closer to the PM within existing ER-PM junctions. So far, these data suggest that Esyt1 may play an active role in BK-induced inflammatory signaling.

Disclosures: C. Palfrey: None. **N. Gamper:** None. **S. Shah:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.14

Topic: D.02. Somatosensation – Pain

Support: NYS DOH C35596GG
DoD W81XWH-18-1-0746

Title: Contribution of CaV3.2 calcium channels to nociceptors' hyperexcitability and chronic neuropathic pain following spinal cord injury.

Authors: K. GUNARATNA, H. LIU, E. SIPPLE, J. LAUZADIS, M. KACZOCHA, ***M. PUOPOLO**;
Stony Brook Med., Stony Brook, NY

Abstract: A hyperexcitable state and spontaneous activity of nociceptors following spinal cord injury (SCI) have been suggested to play a primary role in the development of SCI-induced neuropathic pain (SCI-pain). Previous work showed that the increased activity of T-type calcium channels induced by the injury is responsible for driving nociceptors' hyperexcitability and their spontaneous activity (Lauzadis et al., J Neurosci. 2020 Sep 16;40(38):7229-7240). The aim of this work is to test the hypothesis that the increased activity of the CaV3.2 isoform induced by the injury is responsible for driving nociceptors to a hyperexcitable state and for promoting the development/maintenance of SCI-pain. Wild type and CaV3.2-/- mice were used in this study. SCI was performed by a midline spinal cord contusion at T10 by using an Infinite Horizon Impactor (50 kilodynes, 1-s dwelling time). The patch clamp technique was used in dissociated dorsal root ganglia (DRG) neurons isolated from SCI, sham, and naïve mice. The mechanical allodynia was assessed with the von Frey filaments by using the up-down method with the 50% threshold. The conditioned place preference (CPP) paradigm was used to assess the spontaneous pain. In wild type mice, the 50% mechanical threshold dropped from 1.66±0.19 g (pre-injury) to 0.85±0.20 g (post-SCI). TTA-P2 (10 mg/kg, a blocker of T-type calcium channels) rescued the 50% mechanical threshold to 1.36±0.15 g at 1-hour post-injection. In CaV3.2-/- mice, the 50% mechanical threshold dropped from 1.34±0.12 g (pre-injury) to 1.14±0.19 g (post-SCI). TTA-P2 (10 mg/kg) rescued the 50% mechanical threshold to 1.32±0.17 g at 1-hour post-injection. The same cohort of wild type and CaV3.2-/- SCI mice were subjected to the CPP paradigm. Wild type SCI mice showed an increase in the TTA-P2-paired chamber of 52±36 sec and a decrease in the vehicle-paired chamber of -62±33 sec. CaV3.2-/- SCI mice showed an increase in the TTA-P2-paired chamber of 30±76 sec and an increase in the vehicle-paired chamber of 12±108 sec. In current-clamp recordings from nociceptors isolated from wild type SCI mice, the frequency-current (FI) relationship was reduced from 16±2 Hz in control (with 100 pA current injection) to 9±3 Hz in 1 µM TTA-P2 (with 100 pA current injection). In nociceptors isolated from CaV3.2-/- SCI mice, the FI relationship was reduced from 10±4 Hz in control (with 100 pA current injection) to 8±3 Hz in 1 µM TTA-P2 (with 100 pA current injection). Taken together, our data suggest that the increased activity of CaV3.2 calcium channels induced by the injury plays a primary role in driving SCI-nociceptors to a hyperexcitable state and contributes to chronic neuropathic pain following SCI.

Disclosures: **K. Gunaratna:** None. **H. Liu:** None. **E. Sipple:** None. **J. Lauzadis:** None. **M. Kaczocha:** None. **M. Puopolo:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.15

Topic: D.02. Somatosensation – Pain

Title: Evaluation of the antinociceptive effect of Strawberries

Authors: J. V. ESPINOSA-JUÁREZ[1], J. R. ZAPATA-MORALES[2], L. A. MORENO-ROCHA[7], E. FRANCO-ROBLES[3], C. OZUNA-LÓPEZ[4], E. DÍAZ-CERVANTES[5], ***O. A. JARAMILLO-MORALES**[6];
[1]Ciencias Químicas Sede Ocozocoautla, Univ. Autónoma de Chiapas, Chiapas, Mexico; [2]Farmacia, Univ. de Guanajuato, Guanajuato, Mexico; [3]Veterinaria y Zootecnia, [4]Alimentos, Univ. de Guanajuato, IRAPUATO, Mexico; [5]Alimentos, Univ. de Guanajuato, Tierra Blanca, Guanajuato, Mexico; [6]Enfermería y Obstetricia, Univ. de Guanajuato, Irapuato, Mexico; [7]Sistemas Biológicos, Univ. Autónoma Metropolitana, Unidad Xochimilco, Mexico, Mexico

Abstract: Strawberries has promising medicinal properties and has shown some degree of remedial effects against cancer, cardiovascular disorders, diabetes, obesity and numerous infections. However, there is no scientific evidence to corrobate its analgesic effect. The objective of the present study was to evaluate the antinociceptive effect of lyophylized strawberries in a pain model. Methods. Strawberry freeze-dried was obtained from Frutas Lio by Alpafe. The antinociceptive activity was evaluated in rats in which nociception was induced intraplantar with 5% formalin. Flinching nocifensive behavior was counted in both phases induced-formalin injection, neurogenic and inflammatory. Results. The intragastric administration of Strawberry freeze-dried (100 mg/kg) showed significantly decreased in the flinching experimental values in the inflammatory phase (antinociceptive effect). Conclusion. Our results indicate for the firs time the antinociceptive activity of Strawberry freeze-dried in a inflammatory pain.

Disclosures: **J.V. Espinosa-Juárez:** None. **J.R. Zapata-Morales:** None. **L.A. Moreno-Rocha:** None. **E. Franco-Robles:** None. **C. Ozuna-López:** None. **E. Díaz-Cervantes:** None. **O.A. Jaramillo-Morales:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.16

Topic: D.02. Somatosensation – Pain

Support: NIH grant R01NS120395

Title: Kappa opioid receptor agonists produce sexually dimorphic and prolactin-dependent hyperalgesic priming

Authors: *C. M. KOPRUSZINSKI[1], H. J. STRATTON[4], M. WATANABE[2], A. L. MARTINEZ[2], L. MOREIRA DE SOUZA[2], D. W. DODICK[5], A. MOUTAL[6], V. NEUGEBAUER[7], F. PORRECA[3], E. NAVRATILOVA[2];
[1]Univ. of Arizona, [2]Pharmacol., [3]Dept Pharmacol., Univ. of Arizona, Tucson, AZ; [4]Barrow Neurolog. Inst., Tucson, AZ; [5]Mayo Clin., Phoenix, AZ; [6]Pharmacol. and physiology, St. Louis Univ., St Louis, MO; [7]Pharmacol. and Neurosci., Texas Tech. Univ. HSC, Lubbock, TX

Abstract: Introduction: Repeated stress is maladaptive and promotes negative outcomes including negative hedonic states. Hyperalgesic priming in preclinical models might provide mechanistic insights regarding pain chronification. In this paradigm, a first hit stimulus produces neural adaptation and induce sensitization to a subsequent subthreshold second hit stimulus. In mice, three episodes of stress produces hyperalgesic priming, but the underlying mechanisms remain unclear.**Objective:** As stress engages kappa opioid receptors (KOR), we hypothesized that repeated systemic administration of U-69593 or nalfurafine, non-biased and biased KOR agonists, respectively, could mimic hyperalgesic priming of repeated stress. We hypothesized that the priming effect of KOR agonists was independent of signaling bias and that KOR-induced priming would be female selective and dependent upon PRL-induced nociceptor sensitization.**Methods:** Mice received three daily doses of intraperitoneal U-69593 or nalfurafine as a "first hit" stimulus followed by assessment of periorbital allodynia and circulating prolactin. Sixteen days later, a subthreshold dose of inhalational umbellulone, a TRPA1 agonist, provided the second hit and periorbital allodynia was assessed. Cabergoline, a dopamine D2 agonist, was administered to inhibit circulating PRL in additional cohorts. PRL receptor (PRLR) isoforms were quantified in the V1 region of the trigeminal ganglion after repeated doses of U-69593 using Western blot. In addition, current clamp recordings were performed from cultured sensory neurons following overnight exposure to 50nM PRL and excitability was assessed. Experimenters were blind to treatments and mice were randomly divided into control and experimental groups. **Results:** In both sexes, KOR agonists increased circulating PRL and produced a transient allodynia. Hyperalgesic priming, revealed by umbellulone-induced allodynia in animals previously treated with KOR agonists, occurred in both sexes. Repeated U-69593 triggered PRLR-L downregulation in trigeminal neurons of female mice. Umbellulone-induced allodynia was prevented by cabergoline co-treatment during priming in female, but not male, mice. Furthermore, sensory neuron excitability was dramatically enhanced following exposure to overnight PRL in female mice but was only minimally affected in males.**Conclusions:** Hyperalgesic priming therefore occurs in both sexes following either biased or non-biased KOR agonists. However, a PRL/PRLR-dependent sexual dimorphism is observed only in female nociceptors possibly contributing to the female-prevalence of stress-related pain disorders.

Disclosures: **C.M. Kopruszinski:** None. **H.J. Stratton:** None. **M. Watanabe:** None. **A.L. Martinez:** None. **L. Moreira de Souza:** None. **D.W. Dodick:** None. **A. Moutal:** None. **V. Neugebauer:** None. **F. Porreca:** None. **E. Navratilova:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.17

Topic: D.02. Somatosensation – Pain

Support: NIH Grant NS091759
NIH Grant NS111521

Title: Memory-related induction mechanisms can trigger persistent hyperexcitability of nociceptors

Authors: M. Y. ZHU, ***A. BAVENCOFFE**, C. W. DESSAUER, E. T. WALTERS; Integrative Biol. and Pharmacol., McGovern Med. Sch. At UTHealth, Houston, TX

Abstract: Persistent hyperactivity of primary nociceptors contributes to diverse forms of chronic pain. Much is known about mechanisms that maintain persistent nociceptor hyperexcitability (e.g., Garza Carbajal et al., 2020, J Neurosci 40:6522; Bavencoffe et al., 2022, J Neurosci, in press), but little is known about induction mechanisms. However, some parallels have been noted between processes that can induce long-lasting nociceptor hyperexcitability and cell signaling mechanisms involved in memory formation (e.g., Weragoda et al., 2004, J Neurosci 24:10393; Price et al., 2015, Prog Mol Biol Transl Sci, 131:409). To test the hypothesis that cellular processes important for the induction of late long-term synaptic potentiation (L-LTP) and memory in the brain can also induce long-term hyperexcitability (LTH) of dissociated nociceptor somata, we have used an in vitro model to test whether nociceptor LTH can be induced by cAMP signaling involving PKA activity, CREB activity, gene transcription, and protein synthesis. DRG neurons from adult male rats were dissociated and cultured overnight. Beginning 1 h after dissociation, neurons were incubated for 6 h with a Gs-coupled serotonin receptor (5-HT4R) agonist, prucalopride. Other neurons received co-treatment with either a PKA inhibitor, H89; a CREB inhibitor, 666-15; a transcription inhibitor, actinomycin D; or a translation inhibitor, cycloheximide, at concentrations reported to block induction of hippocampal L-LTP. Whole-cell patch recordings under current-clamp were made 12-24 h after washout. Early treatment with prucalopride induced LTH manifested as an increase in incidence of ongoing activity (OA) shown as repetitive firing of action potentials (APs) when neurons were depolarized artificially to -45 mV for 30 s. Blocking 5-HT4Rs with a specific inhibitor, GR113808, overnight beginning immediately after prucalopride washout failed to prevent LTH. Co-treatment during induction by prucalopride with H89, 666-15, actinomycin D, or cycloheximide significantly reduced LTH. Specific electrophysiological manifestations of LTH (including changes in resting membrane potential, AP voltage threshold, and depolarizing spontaneous fluctuations of membrane

potential) induced in vitro will be compared to those reported previously in diverse persistent pain models.
Our results support the hypothesis that conserved, memory-related mechanisms can induce persistent, pain-related nociceptor activity. This simple, in vitro preparation enables detailed examination of potential induction mechanisms, and might reveal new therapeutic targets for preventing the transition to chronic pain.

Disclosures: **M.Y. Zhu:** None. **A. Bavencoffe:** None. **C.W. Dessauer:** None. **E.T. Walters:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.18

Topic: D.02. Somatosensation – Pain

Support: MBRU Internal Research Grant (MBRU-CM-RG2018-12)
Al Jalila Foundation

Title: The role of nociceptive ion channels in pain development in a pre-clinical model of inflammatory prostatitis

Authors: ***A. CYRIL**[1], Y. LOZON[2], M. OTHMAN[1], Y. AL GAFFARI[1], N. KARUVANTEVIDA[1], J. S. KUMAR[1], H. JAVED[3], S. A. SHEHAB[3], R. RADHAKRISHNAN[1]; [1]Col. of Med., Mohammed Bin Rashid Univ. of Med. and Hlth. Sci., Dubai, United Arab Emirates; [2]Dubai Pharm. Col. for Girls, Dubai, United Arab Emirates; [3]Col. Med. & Hlth. Sci., Fac Med. & Hlth. Sci., Al-ain, United Arab Emirates

Abstract: Prostatitis is a common painful urological problem in the United Arab Emirates and worldwide. Of these patients, 90% suffer from nonbacterial chronic prostatitis/chronic pelvic pain syndrome (CP/CPPS), a chronic pain condition classified as NIH category III prostatitis. Although it is not a life-threatening condition, the symptoms significantly affect the quality of life in these patients, and effective treatments are sparse. The main objective of this study was to determine the molecular mechanism of pain in CP/CPPS by investigating the expression of pain transducer ion channels namely acid-sensing ion channel type 3 (ASIC3), purinergic channel type 3 (P2X3), and transient receptor potential vanilloid type 1 channel (TRPV1) in the dorsal root ganglia (DRG - Lumbar 5 and Lumbar 6) of prostate inflamed rats and compare their expression with non-inflamed rat tissue using immunohistochemistry and RT-PCR. Prostatitis was induced in male Wistar rats by injecting 3% carrageenan into the prostate. The results showed an increased expression of the three channels in the L5-L6 DRGs prostate-inflammed rats. Using fluorescent immunohistochemistry, the percentage of fluorescent cells were determined and compared between the two study groups. For the TRPV1 and P2X3 expression, the prostate-inflamed group showed statistically significant increased expression in the L5 and

L6 DRG compared to the control group (TRPV1; p= 0.012 and p=0.04 for L5 and L6 DRG respectively, P2X3; p= 0.03 and p=0.001 in L5 and L6 DRG respectively) while a higher level of ASIC3 expression, that was statistically not significant (p = 0.075 and 0.08 for L5 and L6 respectively). Furthermore, the RT-PCR results showed that the mRNA levels of all the three channels were significantly increased in the L5-L6 DRG of prostate-inflamed rats (p= 0.007 (ASIC3); p=0.003 (TRPV1); p=0.01 (P2X3). The expression fold change values for ASIC3, P2X3, and TRPV1 were 2.89, 1.7, and 3.81 respectively. Our results suggest a significant involvement of molecular receptors - ASIC3, TRPV1 and P2X3 in mediating the generation of pain in rats with carrageenan-induced prostatitis. If these findings translate to humans, these ion channels can be targeted to develop new drugs to treat pain in patients with NIH category IIIA CP/CPPS.

Disclosures: A. Cyril: None. **Y. Lozon:** None. **M. Othman:** None. **Y. Al Gaffari:** None. **N. Karuvantevida:** None. **J.S. Kumar:** None. **H. Javed:** None. **S.A. Shehab:** None. **R. Radhakrishnan:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.19

Topic: D.02. Somatosensation – Pain

Support: 75N95019D00026

Title: In vivo PK, side effect profile, and efficacy of multiple classes of analgesics in rats

Authors: *E. DUGAN[1], D. BUDAC[1], C. MCDONNELL[1], M. URBAN[1], S. A. WOLLER[2], S. IYENGAR[2], T. HANANIA[1], M. A. VARNEY[1];
[1]Psychogenics, Paramus, NJ; [2]NIH/NINDS, Rockville, MD

Abstract: In collaboration with the NIH HEAL Initiative Preclinical Screening Platform for Pain (PSPP), we evaluated clinically used analgesics, including morphine, gabapentin, duloxetine and ketoprofen through the tiered approach established to profile potential novel analgesics. First, pharmacokinetic studies were conducted to guide dosing, select the route of administration, and to determine the time course, supporting subsequent behavioral studies. Next, the modified Irwin (n=4) and rotarod tests (n=10) were conducted to evaluate potential neurologic, physiologic, and fine motor effects that may impact outcome measures in the pain models. Following side effect profile assessment, efficacy was evaluated in the plantar incisional pain (n=10) and L5/L6 spinal nerve ligation (SNL; n=10) models. The rat plantar incisional pain model is an established model of acute post-operative pain induced by incision of the skin and the plantaris muscle (Brennan et al. 1996). The model is characterized by transient hind paw tactile allodynia and spontaneous guarding behaviors. SNL is a model of peripheral neuropathic pain resulting from chronic nerve compression in which tactile and cold allodynia are produced (Kim and Chung, 1992). All

experiments were conducted in a blinded manner with both sexes included. Power analysis was used to determine the group sizes for the various assays. The results of these studies of clinically used analgesic standards demonstrate the validation of the models and endpoints within the PSPP program and highlight the goal of providing a robust platform to accelerate the discovery and preclinical development of non-opioid, non-addictive treatments for pain.

Disclosures: **E. Dugan:** A. Employment/Salary (full or part-time):; Psychogenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **D. Budac:** A. Employment/Salary (full or part-time):; Psychogenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **C. McDonnell:** A. Employment/Salary (full or part-time):; Psychogenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **M. Urban:** A. Employment/Salary (full or part-time):; Psychogenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **S.A. Woller:** None. **S. Iyengar:** None. **T. Hanania:** A. Employment/Salary (full or part-time):; Psychogenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **M.A. Varney:** A. Employment/Salary (full or part-time):; Psychogenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.20

Topic: D.02. Somatosensation – Pain

Support: NIH Contract No. 75N95019D00026.

Title: Evaluation of the abuse liability of oxycodone in male and female rats using two approaches: intravenous self-administration and conditioned place preference

Authors: *Q. CHANG[1], E. DUGAN[2], W. MIN[1], W. LACSINA[1], P. V. SEVERINO[1], H. BUECHLER[1], D. A. NICHOLSON[1], S. A. WOLLER[3], S. IYENGAR[4], T. HANANIA[2];

[1]PsychoGenics Inc., Paramus, NJ; [2]Behavioral Pharmacol., PsychoGenics, Inc., Paramus, NJ; [3]DTR, NIH/NINDS, Rockville, MD; [4]NINDS/NIH, Rockville, MD

Abstract: In collaboration with the NIH HEAL Initiative Preclinical Screening Platform for Pain (PSPP), we evaluated the abuse liability properties of oxycodone in the intravenous self-administration and conditioned place preference (CPP) tests in male and female Sprague Dawley rats. Intravenous drug self-administration took place in sound attenuated operant chambers (Med Associates, VT) where rats pressed an active lever that delivered the test compound intravenously through a jugular vein catheter. Rats were allowed to self-administer saline (negative control), morphine (0.6 mg/kg/infusion) or oxycodone (0.01, 0.03, 0.06 and 0.1 mg/kg) by pressing the active lever on an FR3 schedule. Acquisition training lasted 20 days. In male and female rats, oxycodone (0.03, 0.06 and 0.1 mg/kg/infusion) showed higher infusion rate compared to saline. In the CPP study, an independent group design (N=16) was used for both male and female rats with five treatment groups (saline, cocaine 15 mg/kg, and oxycodone 1, 3 and 5 mg/kg) injected intraperitoneally (IP). Perceptive cues were applied to create a distinctive texture and visual features for the two compartments. A 10-day protocol was used in this study. The study was videotaped on Day 1 (baseline) and on Day 10 (bias test). The time spent in the different chambers was scored by an experimenter blinded to the treatment. Days 2-9 of the test were conditioning days in which differentiation between “drug compartment” and “saline compartment” was achieved. Rats were treated with saline on days 2, 4, 6 and 8, and with either cocaine or oxycodone on days 3, 5, 7, 9. Animals were confined in the “drug compartment” or “saline compartment” immediately after drug administration for 20 minutes. Compared to saline, cocaine, used as the positive control, induced significant CPP. Oxycodone (1, 3 and 5 mg/kg; IP) induced even larger bias between the two compartments ($P<0.001$) in both male and female rats. These data confirm the high abuse potential of oxycodone in male and female rats and both assays can be used to screen the potential abuse liability of novel therapies as part of the NIH HEAL Initiative’s PSPP program towards discovering novel non-addictive analgesics.

Disclosures: **Q. Chang:** A. Employment/Salary (full or part-time):; Full time employee, PsychoGenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **E. Dugan:** A. Employment/Salary (full or part-time):; Full time employee, PsychoGenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **W. Min:** A. Employment/Salary (full or part-time):; Full time employee, PsychoGenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **W. Lacsina:** A. Employment/Salary (full or part-time):; Full time employee, PsychoGenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **P.V. Severino:** A. Employment/Salary (full or part-time):; Full time employee, PsychoGenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **H. Buechler:** A. Employment/Salary (full or

part-time):; Full time employee, PsychoGenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **D.A. Nicholson:** A. Employment/Salary (full or part-time):; Full time employee, PsychoGenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO. **S.A. Woller:** None. **S. Iyengar:** None. **T. Hanania:** A. Employment/Salary (full or part-time):; Full time employee, PsychoGenics. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; CRO.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.21

Topic: D.02. Somatosensation – Pain

Support: NIH/NIGMS R35GM138168

Title: The role of proinflammatory multi-protein complex NLRP3 in a rat model of inherited, persistent hyperalgesia

Authors: *C. K. REY, L. F. FERRARI, N. E. TAYLOR;
Univ. of Utah, Univ. of Utah, Salt Lake City, UT

Abstract: The nucleotide-binding oligomerization domain (NOD)-like receptor 3 (NLRP3) has been proposed as a key regulator of inflammation and pain in fibromyalgia (FM). However, no studies have been conducted in animal models or in FM patients investigating the mechanism of increased NLRP3 activity in widespread pain conditions. Elucidating these mechanisms could identify novel therapeutic targets to treat pain. We utilized a novel model of inherited, persistent, hyperalgesia, the Dahl S rat (SS), to test our hypothesis that inherited increases in reactive oxygen species (ROS) and toll-like receptor 4 (TLR4) activity could be responsible for increased NLRP3 activity and pain. We used both pharmacologic (dexamethasone, NLRP3 inhibitor MCC950, ROS scavenger Tempol) and genetic approaches (SS-NcF2-/- and SS-TLR4-/-) to inhibit the activities of TLR4, NLRP3 and ROS and measured their effects on mechanical paw withdrawal thresholds (Randall-Selitto test) in SS rats. Dexamethasone dose-dependently increased paw-withdrawal thresholds of SS rats (15%-29% increase compared to control, $P<0.0001$), indicating the involvement of systemic inflammation. Similarly, the NLRP3 inhibitor MCC950 also produced a significant increase in the paw withdrawal thresholds (13%-42% increase compared to control, $P<0.0001$). To begin to test why NLRP3 activity might be elevated, we found that scavenging ROS using tempol attenuated the SS rat hyperalgesia (30%-

62% increase compared to control, P<0.0001). In addition, higher mechanical nociceptive thresholds were also observed in the NcF2-M1 knockout rats (17% increase compared to the SS rat, P<0.0001). This strain lacks the gene for p67phox, the activator of the NADPH oxidase Nox2 responsible for the increased ROS production in SS rats. Finally, paw-withdrawal thresholds were also higher in TLR4 knockout rats (12% increase compared to the SS rat, P<0.001) suggesting a role of this receptor in SS rat hyperalgesia. Together, the results suggest that chronic systemic inflammation contributes to the low mechanical nociceptive threshold phenotype observed in SS rats, in a mechanism involving NLRP3 and its upstream mediators TLR4 and ROS. Future studies are needed to directly test the effects of ROS and TLR4 on NLRP3 activity in SS rats. The findings support the use of the SS rat as a strain to investigate how inflammation pathways contribute to conditions involving inherited chronic pain conditions.

Disclosures: C.K. Rey: None. **L.F. Ferrari:** None. **N.E. Taylor:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.22

Topic: D.02. Somatosensation – Pain

Support: SC INBRE P20GM103499-20

Title: The effects of arnica montana extracts as a non-opioid treatment for post operative pain

Authors: *R. TURNER[1], D. HOLMES[1], T. PRICE[2], W. LEI[3];
[1]Presbyterian Col., Clinton, SC; [2]The Univ. of Texas at Dallas, Richardson, TX; [3]Pharm., Manchester Univ., North Manchester, IN

Abstract: *Arnica Montana* (Arnica) is an herbaceous perennial plant that has been traditionally used intreating trauma, bruises, inflammation, or tissue injuries. However, the molecular mechanisms of Arnica's medicinal properties are largely unknown. The objective of this study is to evaluate the effects of the Arnica extracts on post-operative pain in CD-1 mice. The extracts were made by mixing Arnica powder with one of the following solvents: 100% ethanol, ethanol: water (7:3,v/v), methanol, and acetone. The extracts were dried at room temperature and mixed in PCCAVersaBase gel at 1% by weight, with or without 1% of resveratrol. The analgesic effect of Arnica extracts was tested in a post-operative pain model in CD-1 male and female mice. Mice were treated with gel alone, gel with 1% Arnica extract, or gel with 1% Arnica extract + 1% resveratrol (a generally recognized as safe compound) for one hour under anesthesia after the paw incision surgery. After recovered, the von Frey assay was performed at 3, 5, 24, and 48 hours after surgery. We found that all Arnica extracts exhibited analgesic effects to reduce the post-operative pain. However, the co-treatment with resveratrol only enhanced the activity of ethanolic aqueous (7:3) extract and acetone extracts. The tissues around the incision have been harvested for measuring the concentration of pro-inflammatory mediators, such as interleukin

(IL)-1β, IL-6, and tumor necrosis factor alpha (TNF-α) using Western Blot. These findings further confirm the activity of Arnica on pain relief and will provide a better understanding of the mechanism of analgesic effect of Arnica.

Disclosures: **R. Turner:** None. **D. Holmes:** None. **T. Price:** None. **W. Lei:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; SC INBRE.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.23

Topic: D.02. Somatosensation – Pain

Support: College of Pharmacy, University of Minnesota
Winston and Maxine Wallin Neuroscience Discovery Fund
Center for Drug Design, University of Minnesota
Department of Anesthesiology, University of Minnesota

Title: Development of novel non-opioid alpha-2 adrenergic receptor agonists for the treatment of pain

Authors: *C. PETERSON[1], L. CAYE[2], W. XIE[3], A. NOLL[4], K. F. KITTO[5], G. L. WILCOX[5], L. S. STONE[4], S. MORE[3], C. A. FAIRBANKS[1];
[1]Pharmaceutics, [2]Mol. Pharmacol. and Therapeut., [3]Ctr. for Drug Design, [4]Anesthesiol., [5]Neurosci., Univ. of Minnesota, Minneapolis, MN

Abstract: Alpha 2 adrenergic receptor (α_2-AR) agonists offer an effective, non-opioid analgesic strategy. While clonidine remains the best characterized α_2-AR agonist, it is limited in its widespread use by side effects including hypotension and sedation which limit patient compliance and clinical utility. Guanabenz (GB) is an FDA-approved antihypertensive and has been shown to be an effective analgesic in pre-clinical models of pain. Its use, however, is also limited by sedative side effects. We therefore sought to develop a library of GB-based α_2-AR agonists and characterize their analgesic efficacy, side effect profile, kinetics, and α_2-AR subtype affinity.
Methods: We designed and synthesized novel GB analogues with an intent to differentiate their analgesic properties from the sedative side effects. To determine dose ranges, female and male ICR mice (21-30 g) were injected intrathecally with substance P to induce transient scratching and biting behaviors. Mice were pre-treated with either in-class control, the parent compound GB, or the newly developed α_2-AR agonists as single agents or in combinations and the maximum percent effect (%MPE) in reduction of the Substance P (SP)-induced scratching and biting behaviors was calculated as compared to saline control. Following MPE calculation, high

effective [LSS1] doses were tested in the rotarod motor coordination assay and open field assay to investigate motor impairment and sedation. Additionally, these compounds were assessed in models of neuropathic pain (spared nerve injury), low back pain (SPARC-null transgenic mouse), and inflammatory pain (complete Freund's adjuvant (CFA) injection).
Results: We observed that increasing doses of the GB-based, α_2-AR agonists were effective when delivered singly or in combination in reducing transient scratching and biting pain behaviors in the sub-acute SP assay. The efficacy of the GB-based compounds was inhibited by idazoxan but not naloxone co-administration, supporting action at α_2-AR and not opioid receptors. The GB-based compounds were also effective at reducing expression of pain behavior in acute (inflammatory, and chronic (low back pain, neuropathic pain) rodent models of pain. Additionally, the compounds showed attenuated sedation as compared to the parent compound, GB and a positive in-class control, clonidine. Pursuit of guanabenz-related analogs may represent an improved non-opioid analgesic strategy and an alternative to opioid for the management of chronic pain.

Disclosures: C. Peterson: None. **L. Caye:** None. **W. Xie:** None. **A. Noll:** None. **K.F. Kitto:** None. **G.L. Wilcox:** None. **L.S. Stone:** None. **S. More:** None. **C.A. Fairbanks:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.24

Topic: D.02. Somatosensation – Pain

Support: VA RR&D SPiRE 1I21RX003453
VA TTP BRAVE 2020-144

Title: Therapeutic effect of boldine in spare nerve injury-induced pain in mice

Authors: J. PAN[1], W. BAUMAN[1], C. CARDOZO[1,2], ***W. ZHAO**[1,2];
[1]James J Peters VA Med. Ctr., James J Peters VA Med. Ctr., Bronx, NY; [2]Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Objective: Chronic pain is a major health problem in the health care system. It is well established that chronic pain, such as inflammation pain, neuropathic pain, and cancer pain, is an expression of neural plasticity both in the peripheral nerve system (PNS) and in the central nerve system (CNS). Chronic neuropathic pain is caused by a lesion or disease of the somatosensory (peripheral and/or central) nervous system. In this study, we evaluated boldine, an alkaloid derived from the bark and leaves of the Chilean tree *Peumus boldus*, for its capacity to reduce neuropathic pain induced by peripheral nerve injuries, and potential underlying mechanisms.
Methods: 4-month-old C57BL6 mice were administered with boldine or vehicle, and subjected to spared nerve injury (SNI). Pain phenotypes including mechanical allodynia, thermal preference and spontaneous pain were measured using Von Frey filament test, thermal gradient

test (TGT) and dynamic weight bearing (DWB) test, respectively. Brain and spinal cord were harvested 14 days post injury (dpi), RNAs were extracted and subjected to qPCR. To test the effect of boldine on inflammasome signaling, astrocytes and microglia were stimulated with LPS and treated with boldine. Major components of the NLRP3 inflammasome and inflammation markers were quantified by qPCR. Results: Our *in vivo* studies conducted in a mouse model of SNI suggest that boldine administration leads to a significant reduction of nerve injury-induced mechanical pain hypersensitivity. As measured by DWB, boldine also significantly prevents the SNI-induced decrease of ipsilateral weight support at 7 dpi. Temperature preference along a continuous temperature gradient (16-55°C) was also performed. Before injury, animals show a baseline of temperature preference at 34.7°C. At 14 dpi, sham-operated animals still show a temperature preference at 34.7°C, while SNI animals spend much less time around 34.7°C and appear more often at colder zones (16-22°C) instead. SNI animals that received boldine administration demonstrate a temperature preference at 32.7°C, suggesting that boldine has a beneficial effect on improving the temperature preference deficit induced by SNI. The behavioral changes are accompanied with decreased expression of some inflammation markers in the brain and spinal cord including CCL2, IL-1β and TNF-α. *In vitro* studies in astrocytes and microglia cells show that boldine could drastically inhibit the NLRP3 inflammasome activation and IL-1β expression. Conclusion: Oral administration of boldine could alleviate spare nerve injury-induced neuropathic pain, possibly through inhibiting the NLRP3 inflammasome activation.

Disclosures: J. Pan: None. **W. Bauman:** None. **C. Cardozo:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Co-inventor for provisional patent (US Application No. 63/043,572). **W. Zhao:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Co-inventor for provisional patent (US Application No. 63/043,572).

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.25

Topic: D.02. Somatosensation – Pain

Support: AANA Foundation Art Zwerling Grant 2022-G-5

Title: Development of glutamate transporter modulators as a novel, non-opioid treatment for neuropathic pain

Authors: *R. TEMMERMAND, A. C. FONTANA;
Pharmacol. & Physiol., Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: Neuropathic pain (NP), a disease of the somatosensory nervous system, afflicts many people and adequate management with the current pharmacotherapies remains ineffective. NP is

associated with excess levels of glutamate, which overstimulates post-synaptic glutamate receptors and increases pain transmission. Excitatory amino acid transporters (EAATs) remove extracellular glutamate and, thus, are crucial for the regulation of excitatory and nociceptive transmission. EAATs are downregulated in NP states, suggesting a potential target for therapeutic treatment. We have identified a series of potent, selective, positive allosteric modulators (PAMs) that increase the activity of EAAT2, the main transporter responsible for glutamate clearance in the CNS. Preliminary data in male rats showed an anti-nociceptive effect of our lead EAAT2 PAM compound, NA-014, after spared nerve injury (SNI), a clinically relevant model of NP. We extended these studies to mouse models of NP and included additional behavioral assays. We hypothesized that NA-014 would provide anti-nociception in mice after peripheral nerve injury. In male mice, NA-014, was tested for anti-nociception at 7, 14 and 21 days after SNI surgery. Mice (n =6/group) were administered NA-014, gabapentin (positive control) or vehicle via IP. Behavioral testing (pre-surgery, and before and after compound administration) included Von Frey to measure mechanical allodynia and dynamic weight bearing (DWB) to measure spontaneous pain behavior. Mice administered NA-014 (100 mg/kg) demonstrated a tolerance to increasing forces of Von Frey filaments compared to vehicle ($p < 0.05$) on day 7 after SNI, but not on days 14 and 21 days. Conversely, mice administered gabapentin showed a significant difference in pain thresholds compared to vehicle at all time points. These results suggest EAAT2 PAMs, with further optimization, are a potential early pain therapy after nerve injury. In DWB, SNI animals demonstrated a decrease in weight-bearing on the injured paw after SNI, which was not reversed by NA-014 or gabapentin at any time point, suggesting the DWB assay requires further optimization. In future studies we will increase the group size and include the mechanical conflict-avoidance assay to study EAAT2 PAMs on the motivational and affective aspects of pain. Our future studies will also include female animals to investigate sex differences. Finally, we will investigate different NP models (e.g., taxol administration, a model of chemotherapy-induced neuropathy). Overall, increasing the efficacy of EAATs may serve as a novel way to successfully treat those suffering from NP.

Disclosures: R. Temmermand: None. **A.C. Fontana:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.26

Topic: D.02. Somatosensation – Pain

Support: FRQ-NT Grant 2018-PR-207591
CIHR Grant FDN-148413

Title: Pepducins allosterically modulate neurotensin receptor type 1 signalling and function

Authors: *R. L. BROUILLETTE[1], É. BESSERER-OFFROY[2], C. MONA[2], J. COTE[1], J.-M. LONGPRÉ[1], É. MARSAULT[1], M. GRANDBOIS[1], P. SARRET[1];

[1]Pharmacology-Physiology, Univ. de Sherbrooke, Sherbrooke, QC, Canada; [2]David Geffen Sch. of Med., UCLA, Los Angeles, CA

Abstract: “Pepducins” represent an innovative strategy in targeting G protein-coupled receptors (GPCRs), a highly successful protein target class in drug development. Unlike traditional orthosteric ligands, pepducins interact at the intracellular receptor-effector interface and modulate GPCR signaling by acting as allosteric agonists or allosteric modulators. They are composed of an N-terminal palmitic acid and a peptide sequence mimicking one of the intracellular loops of a GPCR of interest. We previously designed a series of pepducins derived from the first intracellular loop of the human neurotensin type 1 receptor (hNTS1), a class A GPCR that mediates many neurotensin (NT) effects, including analgesia and hypotension. Notably, these pepducins have been shown to relieve pain in animal models of acute, tonic, and chronic pain. Here, we further investigate the cellular and functional actions of this pepducin series. First, we demonstrated that our pepducins inhibit orthosteric binding of ^{125}I-labeled NT in CHO-K1 cells stably expressing hNTS1, at high concentrations (≥10 μM). Using BRET-based biosensors, we also found that they preferentially promote GαoA and Gα13 activation over β-arrestin recruitment, and inhibit NT-mediated β-arrestin recruitment. Thus, they appear to act as both biased allosteric agonists and negative allosteric modulators of NTS1. Moreover, despite its inability to promote β-arrestin recruitment, this pepducin induced NTS1 internalization (33%, 1 h post-stimulation), as detected using a β-lactamase-fused construct. Additionally, since NTS1 has been reported to self-assemble into homomeric complexes, and form heteromeric complexes with the apelin GPCR APJ, we used BRET to monitor NTS1-NTS1 and APJ-NTS1 interactions. Our data confirm that, in both cases, this dimerization process is constitutive, rather than ligand-induced. Interestingly, pepducin treatment (100 μM) enhanced NTS1 homomerization. When injected systemically in rats, this pepducin exerted more potent hypotensive effects at 55 nmol/kg than NT(8-13) did at the same dose, with a sustained mean drop in blood pressure of 30 mm Hg. Finally, an alanine scan of this NTS1-derived pepducin, tested on CHO-hNTS1 cells using the Electric Cell-substrate Impedance Sensing (ECIS) method, revealed that the N-terminal RKK motif is critical for pepducin action. In summary, our data show that the NTS1-derived pepducin series allosterically modulates NTS1 receptor function, and can be used to design novel NT-based drugs.

Disclosures: R.L. Brouillette: None. **É. Besserer-Offroy:** None. **C. Mona:** None. **J. Cote:** None. **J. Longpré:** None. **É. Marsault:** None. **M. Grandbois:** None. **P. Sarret:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.27

Topic: D.02. Somatosensation – Pain

Support: PRODEP-SEP 2018 511-6/18-8833

Title: Antinociceptive, antineuropathic and anti-inflammatory effect of the diclofenac-folic acid combination in experimental models in rats

Authors: *A. L. MARTINEZ-MARTINEZ[1], J. J. MARISCAL-RUIZ[1], A. SERRANO-MEDINA[1], J. M. CORNEJO-BRAVO[2], J. J. MANRÍQUEZ-TORRES[1], E. OCHOA-RUIZ[1], G. PINEDA-GARCÍA[1];
[1]Facultad de Medicina y Psicología, [2]Facultad de Ciencias Químicas e Ingeniería, Univ. Autónoma de Baja California, Tijuana, Mexico

Abstract: Pain is the third most common cause of medical care. Although the pharmacological management of pain offers significant relief, in some chronic diseases that occur with inflammatory pain and neuropathic pain, treatment with this type of drugs is not completely successful and the adverse effects associated with them, and their high cost limit its use. It is for these reasons that it is necessary to continue with the search for novel therapeutic alternatives that not only solve the problem of pain, but also minimize the presence of adverse effects and their cost. A viable option is to combine drugs used to treat pain and inflammation. The logic that underlies this strategy is based on two considerations. First, individual drugs do not always provide satisfactory relief: the combination of drugs that act by different mechanisms can increase their effectiveness. Second, individual drugs that provide satisfactory relief can cause adverse effects at the same time. The combination of drugs may allow the reduction in the amount of individual components to achieve the same effect with a lower incidence of adverse effects. Clearly, this is true if the interaction of drugs is in favor of the desired effect, rather than toxicity. This study aims to assess the antinociceptive, antineuropathic and anti-inflammatory effect of the diclofenac-folic acid combination in experimental models in rats. Isobolographic analysis showed that diclofenac and folic acid combination produces a synergistic antinociceptive effect in the formalin test in Wistar rats. Potency of the combination was approximately 4 times greater than expected from the addition of individual drug effects. On the other hand, administration of 30.87 mg/kg of diclofenac plus 11.62 mg/kg of folic acid produces a significant anti-inflammatory effect on carrageenan-induced edema and an anti-allodynic and antihyperalgesic effect in rats. This synergistic effect could be useful clinically because therapeutic efficacy is enhanced without exacerbating the adverse effects of diclofenac, since the combination of both drugs does not damage gastric mucosa. Additionally, in this study it was found that opioid receptors participate in the antinociceptive effect produced by diclofenac and folic acid combination. In conclusion, diclofenac and folic acid combination could result in a good pharmacological strategy to treat inflammatory pain and neuropathic pain.

Disclosures: A.L. Martinez-Martinez: None. **J.J. Mariscal-Ruiz:** None. **A. Serrano-Medina:** None. **J.M. Cornejo-Bravo:** None. **J.J. Manríquez-Torres:** None. **E. Ochoa-Ruiz:** None. **G. Pineda-García:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.28

Topic: D.02. Somatosensation – Pain

Support: California State University Program for Education & Research in Biotechnology
Center for Student Research, Cal State East Bay
Department of Psychology, Cal State East Bay
College of Science, Cal State East Bay

Title: Antinociceptive effects of β-caryophyllene against acute and persistent pain in male rats

Authors: *S. SANCHEZ, A. FERNANDEZ, C. CHIN, R. KANDASAMY;
Dept. of Psychology, California State University, East Bay, Hayward, CA

Abstract: Humans use various strains of Cannabis that contain several hundred different compounds that are not either Δ^9-tetrahydrocannabinol (THC) or cannabidiol (CBD). β-caryophyllene (BCP) is a sesquiterpene found in the essential oils of Cannabis. Despite some early studies, the extent to which these compounds produce pain relief in assays of acute and persistent pain is unclear. Further, the effects of BCP on pain-evoked behaviors and pain-depressed behaviors has never been evaluated. We hypothesized that BCP would inhibit acute pain, mechanical allodynia, and thermal hyperalgesia. We also hypothesized that BCP would restore pain-depressed wheel running activity in male Sprague-Dawley rats with inflammatory pain. We administered varying doses of BCP and evaluated their antinociceptive effects on the hot plate and tail-flick tests in adult male Sprague-Dawley rats. One intraperitoneal injection of 30 mg/kg BCP produced antinociception on the hot plate test and the tail-flick test approximately 15 minutes after injection. Vehicle and doses of 3 mg/kg and 10 mg/kg did not produce antinociception on the hot plate and tail-flick tests. Three different doses of BCP (10, 30, and 100 mg/kg) or vehicle were administered to rats via an intraperitoneal injection after hindpaw inflammation induced by an intraplantar injection of Complete Freund's Adjuvant (CFA). Neither the low dose (10 mg/kg) nor the medium dose (30 mg/kg) of BCP reversed mechanical allodynia of the inflamed hindpaw after intraperitoneal injection. However, a high dose of BCP (100 mg/kg) reversed mechanical allodynia on the von Frey test; however, this dose did not reverse thermal hyperalgesia. A hindpaw injection of 0.1 mL CFA decreased wheel running activity as is consistent with a painful stimulus. However, no dose of BCP restored pain-depressed wheel running. These same doses of BCP did not affect wheel running in uninjured control rats. Therefore, a high dose BCP produces pain relief, although it only does so against mechanical allodynia. No dose of BCP restores normal activity. Therefore, although pain may be eliminated following BCP administration, a return to normal levels of activity may not be possible, which raises questions about the utility of BCP to treat pain. Future studies of the pain-relieving effects of Cannabis constituents must include tests of many pain-related behaviors to understand dose-response relationships and their therapeutic potential.

Disclosures: S. Sanchez: None. **A. Fernandez:** None. **C. Chin:** None. **R. Kandasamy:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.29

Topic: D.02. Somatosensation – Pain

Support: NIH Grant RFNS113881

Title: Upregulated NIS-IncRNA in injured dorsal root ganglion is required for neuropathic pain development and maintenance

Authors: *X. FENG[1], S. DU[1], S. WU[1], B. WANG[1], A. BEKKER[1], S. DAVIDSON[2], **Y. TAO**[1]; [1]Rutgers New Jersey Med. Sch., Anesthesiol., Newark, NJ; [2]Univ. of Cincinnati, Anesthesiol. Neurosci., Cincinnati, OH

Abstract: Neuropathic pain is one of major clinical problems since it can influence more than 7% population in the world. Current treatments of this debilitating disorder have limited efficiency and/or produced severe adverse effects. The evidence demonstrated that maladaptive changes of nerve injury-associated genes in dorsal root ganglion (DRG) are critical for neuropathic pain genesis. Long non-coding RNAs (*lncRNAs*) are considered to be gene transcriptional regulators. Here, we reported a nerve injury-specific lncRNA (*NIS-lncRNA*) for its upregulation in injured DRG exclusively in response to peripheral nerve injury. We demonstrated that *NIS-lncRNA* had two splice variants, full length 435 nt variant 1 and 2,469 nt variant 2, in mouse DRG, a full-length 429 nt *NIS-lncRNA* in rat DRG and a full length 1,263 nt *NIS-lncRNA* in human DRG. Expression of *NIS-lncRNA* was low or undetectable under normal conditions, but its expression was dramatically upregulated in injured DRG neurons after peripheral nerve injury. This upregulation occurred on day 3 post-nerve injury and persisted for at least 28 days after nerve injury. *NIS-lncRNA* was not detected in satellite glial cells and microphages/monocytes in injured DRG. Blocking nerve injury-induced upregulation of *NIS-lncRNA* through DRG microinjection of *NIS-lncRNA* siRNA in CD1 mice or AAV5-Cre in *NIS-lncRNA*$^{fl/fl}$ mice attenuated the development and maintenance of neuropathic pain. Mimicking this upregulation through DRG microinjection of AAV5 expressing full-length *NIS-lcnRNA* or their variants led to neuropathic pain-like behaviours in mice with the absence of peripheral nerve injury. Our findings suggest that DRG *NIS-lncRNA* upregulation is required for neuropathic pain induction and maintenance.

Disclosures: X. Feng: None. **S. Du:** None. **S. Wu:** None. **B. Wang:** None. **A. Bekker:** None. **S. Davidson:** None. **Y. Tao:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.30

Topic: D.02. Somatosensation – Pain

Support: NIH Grant RF1NS113881

Title: Nerve injury-specific long noncoding RNA promotes FUS-triggered CCL2 expression in injured primary sensory neurons

Authors: S. DU, S. WU, X. FENG, B. WANG, A. BEKKER, ***Y. TAO**;
Anesthesiol., Rutgers New Jersey Med. Sch., Newark, NJ

Abstract: Nerve injury-induced dysregulation of pain-associated gene expression in dorsal horn ganglion (DRG) is considered molecular basis of neuropathic pain genesis. Long noncoding RNAs (lncRNAs) are key regulators of gene transcription. We recently identified a novel and native lncRNA specifically responded to peripheral nerve injury in DRG, named nerve injury-specifc lncRNA (*NIS-lncRNA*). Our preliminary data demonstrated that nerve injury-induced upregulation of *NIS-lncRNA* in injured DRG neurons contributed to neuropathic pain development and maintenance. However, the mechanism underlying this phenomenon is still unclear. Here, we showed that blocking *NIS-lncRNA* upregulation attenuated the nerve injury-induced increase in the C-C chemokine ligand 2 (CCL2) in injured DRG. Mimicking nerve injury-induced *NIS-lncRNA* upregulation elevated CCL2 expression and increased CCL2-mediated excitability in DRG neurons. Moreover, *NIS-lncRNA* bound to not only the promoter region of *Ccl2* gene but also to Fused in sarcoma (FUS, an RNA-binding protein). FUS also interacted with *Ccl2* gene promoter and could trigger its transcriptional activity. Thus, upregulated *NIS-lncRNA* recruited more binding of increased FUS to the *Ccl2* gene promoter and augmented *Ccl2* transcription in injured DRG. Given that CCL2 in DRG neurons is an endogenous initiator in neuropathic pain and that *NIS-lncRNA* co-expressed with FUS and CCL2 in DRG neurons, *NIS-lncRNA* participates in neuropathic pain likely by promoting FUS-triggered DRG CCL2 expression and may be a novel target in neuropathic pain management.

Disclosures: S. Du: None. **S. Wu:** None. **X. Feng:** None. **B. Wang:** None. **A. Bekker:** None. **Y. Tao:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.31

Topic: D.02. Somatosensation – Pain

Support: NIH Grant DA032568

Title: Direct activation of TLR7 in nociceptive sensory neurons by single-stranded RNAs

Authors: *O. CHEN[1], S. CHANDRA[2], H. DING[3], C. BAO[1], T. BERTA[4], B. GRAY[1], M. LAY[1], M. CONVERTINO[5], N. DOKHOLYAN[6], A. BORTSOV[7], S. ABRAHAM[1], B. SULLENGER[1], M.-C. KO[3], R.-R. JI[7];
[1]Duke Univ., [2]Duke Univ., Durham, NC; [3]Wake Forest Univ., Winston-Salem, NC; [4]Univ. of Cincinnati Med. Ctr., Cincinnati, OH; [5]Univ. of North Carolina at Chapel Hill, Chapel Hill, NC; [6]Penn State Col. of Med., Hershey, PA; [7]Duke Univ. Med. Ctr., Med. Ctr., Durham, NC

Abstract: MicroRNAs (miRNAs) are small single-stranded non-coding RNAs which regulate gene expression intracellularly. Extracellular miRNAs are also present in various body fluids including serum and cerebrospinal fluid (CSF), and emerging evidence has implied secreted miRNAs may serve as biomarkers for diseases. Extracellular miRNAs may have biological roles via interactions with specific surface receptors. Recently, extracellular let-7b was found to induce apoptosis of cortical neurons via activation of neuronal toll-like receptor (TLR7) contributing to neurodegenerative disease[1]. However, the unique role of extracellular let-7b in modulating cell signaling in nociceptive transmission is virtually unknown. TLR7 expression in immune cells regulates innate and adaptive immunity, but neuronal signaling from TLR7 is not well understood. Using immunohistochemistry staining and RNAscope *in situ* hybridization, we find TLR7 is expressed in primary sensory neurons of dorsal root ganglion (DRG) from different species, including mouse, monkey, and human. Calcium imaging and patch clamp recording each reveal that let-7b can rapidly activate nociceptor neurons across species through TLR7. Computational simulations indicate that let-7b core sequence GUUGUGU interacts with TLR7, and GUUGUGU is sufficient to activate nociceptors and elicit spontaneous pain via TRPA1 ion channel. Extracellular application of let-7b further activates TLR7 and TRPA1 in nociceptor central terminals, enhancing excitatory synaptic transmission in spinal dorsal horn neurons via presynaptic modulation. Spinal blockade of endogenous let-7b reduced pathological pain in mice. Let-7b also produced potent and TRPA1-dependent nociception in monkeys. Collectively, our findings reveal an unconventional signaling by miRNAs through TLR7 in nociceptive sensory neurons across species. This study provides new insights into miRNA function in cell signaling and pain and identifies let-7b as a novel neuromodulator.

Disclosures: O. Chen: None. **S. Chandra:** None. **H. Ding:** None. **C. Bao:** None. **T. Berta:** None. **B. Gray:** None. **M. Lay:** None. **M. Convertino:** None. **N. Dokholyan:** None. **A. Bortsov:** None. **S. Abraham:** None. **B. Sullenger:** None. **M. Ko:** None. **R. Ji:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.32

Topic: D.02. Somatosensation – Pain

Support: 2022R1C1C1008226

Title: Amyloid-beta 1-42 alleviates TRPV1-dependent pain via LRP1-SHP2 pathway

Authors: J. ROH[1], S.-M. HWANG[1], J. PAN[2], S. KIM[1], G. CHUNG[3], C.-K. PARK[1], *Y. KIM[1]; [1]Gachon Pain Ctr. and Dept. of Physiol., Col. of Medicine, Gachon Univ., Incheon, Korea, Republic of; [2]Dept. of Histology and Embryology, Med. Sch. of Nantong University, Nantong, Jiangsu, China; [3]Dept. of Oral Physiol. and Program in Neurobiology, Sch. of Dentistry, Seoul Natl. Unive, Dept. of Oral Physiol. and Program in Neurobiology, Sch. of Dentistry, Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: A neurodegenerative disease accompanying impairment of cognitive and motor function is a disease caused by neuronal cell death of particular subsets of neurons, unlike the normal aging process. Alzheimer's disease is characterized by brain impairment, memory loss, and cognitive impairment that starts slowly and progressively worsens, resulting from the accumulation and aggregation of amyloid-beta (Aβ). However, the Aβ monomer form is rather neuroprotective. A recent study has shown that endogenous Aβ in the spinal cord attenuates pain. Although various types of cells regulate pain threshold in the spinal cord, specific cell types and mechanisms that mediate Aβ's analgesic effect are unknown. Our study has shown that Aβ inhibits TRPV1 activation in vivo as well as in vitro. Indeed, peripheral administration of Aβ alleviated heat hyperalgesia in the mouse SNI model. We confirmed that this TRPV1 inhibitory effect is mediated via Low-density lipoprotein receptor–related protein-1 (LRP1) by using chemical and genetic inhibition of LRP1. Also, we identified that Src homology region 2-containing protein tyrosine phosphatase 2 (SHP2) contributed to TRPV1 inhibition following LRP1 activation. Furthermore, we verified that TRPV1 inhibition by LRP1 activation using α2 macroglobulin (α2M), a potent LRP1 agonist. α2M also inhibited TRPV1 activation in vitro and in vivo levels and activated the SHP2 pathway. Our study found a new mechanism for alleviating chronic pain by modulating the peripheral LRP1-SHP2-TRPV1 pathway.

Disclosures: J. Roh: None. **S. Hwang:** None. **J. Pan:** None. **S. Kim:** None. **G. Chung:** None. **C. Park:** None. **Y. Kim:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.33

Topic: D.02. Somatosensation – Pain

Support: NIH 1RF1NS113883-01
NIH R01 NS045594

Title: Local vasoconstriction in the DRG, via Piezo2, triggers synchronized cluster firing and spontaneous pain following peripheral nerve injury

Authors: W. XIE[1], D. D. LÜCKEMEYER[1], *J. A. STRONG[1], J. ZHANG[1], K. A. QUALLS[1], Q. ZHENG[2], X. DONG[2], J.-M. ZHANG[1];
[1]Univ. of Cincinnati, Cincinnati, OH; [2]Johns Hopkins Univ., Baltimore, MD

Abstract: Previously, we showed spontaneous pain after peripheral nerve injury was associated with intermittent synchronized firing of spatially clustered neurons in the DRG (as observed with in vivo Ca^{2+} imaging using GCaMP6s expressed in sensory neurons) and depended on sympathetic nerve sprouting in the DRG. We now report that cluster firing and spontaneous pain may be induced by blood vessel movements within the DRG in mice with peripheral nerve injury. I.p. injection of the vasoconstrictor phenylephrine (PE) significantly increased

spontaneous pain behaviors and cluster firing in mice with spared nerve injury (SNI; day 21-35). PE did not increase single-neuron spontaneous firing or induce cluster firing in normal mice without peripheral nerve injury. Tracing experiments suggested that injured neurons, not those with intact axons, participated in the cluster firing. Effects of PE on spontaneous pain and cluster firing were reduced by siRNA-mediated knockdown of mechanoreceptor Piezo2 in lumbar DRGs 3-5 days prior. siRNA efficacy was confirmed by Western blotting. The short term siRNA injection did not affect Piezo2 levels in the neuroma, providing further evidence for the DRG as the site of action. In addition, cluster firing could be induced by local injection of PE into the DRG. PE-induced cluster firing and spontaneous pain behaviors were also reduced by knockdown of floxed Piezo2 in the DRG mediated by virally expressed Cre. Effects of i.p. or local PE on cluster firing could be reduced by local DRG application of 2 blockers of mechanically activated channels (GsMTx4 plus (d-)GsMTx4). These blockers also reduced baseline cluster firing. We hypothesized that PE induces vasoconstriction in the DRG, mechanically stimulating Piezo2. After SNI, we observed abundant Piezo2-positive sensory nerve sprouting inside the DRG. In other experiments, we used a glass micropipette to gently poke blood vessels on the DRG surface, evoking local vasoconstriction directly. In the DRGs from SNI but not normal mice, this induced cluster firing, which was also reduced by Piezo2 blockers. Gently poking the DRG cellular area did not induce cluster firing, only single neuron activation. The percentage of mechanical pokes that evoked cluster firing was also reduced by Piezo2 siRNA mediated knockdown. A second vasoconstrictor, angiotensin II, could also evoke spontaneous pain and cluster firing when injected systemically. We propose that vasoconstrictors acting on blood vessels inside DRG mechanically activate Piezo2 channels in nearby sensory fibers/neurons, and evoke cluster firing in the DRG after SNI, thus increasing spontaneous pain behaviors.

Disclosures: W. Xie: None. **D.D. Lückemeyer:** None. **J.A. Strong:** None. **J. Zhang:** None. **K.A. Qualls:** None. **Q. Zheng:** None. **X. Dong:** None. **J. Zhang:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.34

Topic: D.02. Somatosensation – Pain

Support: NIH Grant 1 R35 GM141802

Title: Direct application of a novel neuroactive steroid, (3-alpha, 5-alpha)-3-hydroxy-13,24-cyclo-18,21-dinorchol-22-en-24-ol (CDNC24) on the rat dorsal root ganglia alleviates post-incisional pain

Authors: *M. E. WALZ[1], N. USEINOVIC[1], V. JEVTOVIC-TODOROVIC[1], D. F. COVEY[2], S. M. TODOROVIC[1];

[1]Anesthesiol., Univ. of Colorado Anschutz Med. Campus, Aurora, CO; [2]Dept Developmental Biol., Washington Univ. Sch. Med., Saint Louis, MO

Abstract: The role of $GABA_A$ modulation in both the spinal cord and dorsal root ganglia (DRG) of rodents in nociception was previously studied in chronic preclinical pain models, however similar studies in an acute incisional model are lacking. The main goal of this study is to investigate the changes in $GABA_A$ A_2 subunit expression within the dorsal root ganglia (DRG) after plantar skin incision in rats and determine potential therapeutic benefit of direct DRG application of $GABA_A$ modulators. For this purpose, we used a novel neuroactive steroid and positive modulator of neuronal $GABA_A$ receptors (3-alpha, 5-alpha)-3-hydroxy-13,24-cyclo-18,21-dinorchol-22-en-24-ol (aka CDNC24). Acute incisional pain was induced in Sprague Dawley female adult rats and mechanical threshold was measured using electronic Von Frey for both hind paws in triplicates before and after incision and compound application. Thirty nanograms or 100 nanograms CDNC24 or equal volume (30 microL) of vehicle (25% 2-hydroxypropyl-beta-cyclodextrin), was applied in a blinded manner directly onto the right L5 DRG at the same time as the paw incision surgery. Western blot analysis was performed on acutely dissected lumbar DRGs (L4-L6) collected on post-operative day (POD) 2 or 10. Post-incisional hyperalgesia in naïve animals lasted for approximately 5 days, with POD 2 showing the most pronounced mechanical hyperalgesia (n=15). Mechanical thresholds recovered back to baseline level by POD 10. Western blot analysis showed about 25% (±10%) reduced protein expression of the $GABA_A$ receptor A_2 subunit in the ipsilateral side on POD 2 (n=12). In contrast, no difference in expression of the A_2 subunit was seen in sham incised animals (n=12) or in incised animals at POD 10 (n=15). Direct DRG application of CDNC24 effectively alleviated hyperalgesia at 30 ng /30 microL (n=7) on POD 1, and at 100 ng /30 microL dose (n=8) up to POD 4 when compared to vehicle. In contrast, CDNC24 applied to sham incised animals did not show any difference in mechanical thresholds. Overall, we show that the acute incisional pain model induced a downregulation of the A_2 subunit of $GABA_A$ receptors in ipsilateral DRG that coincided with most intense periods of hyperalgesia. Application of CDNC24 to the DRG dose-dependently diminished hyperalgesia up to POD 4. Since we previously have shown no effective analgesic effect of CDNC24 when administered intrathecally in the same pain model, this argues for specificity of $GABA_A$ modulation by CDNC24 in the DRG. We conclude that the $GABA_A$ receptor modulation in the DRG plays an important role in pathophysiology of post-operative hyperalgesia and represent promising target for novel pain therapies.

Disclosures: M.E. Walz: None. **N. Useinovic:** None. **V. Jevtovic-Todorovic:** None. **D.F. Covey:** None. **S.M. Todorovic:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.35

Topic: D.02. Somatosensation – Pain

Support: NIH Grant NS108087
NIH Grant NS113991
NIH Grant NS128543

Title: Genetic knockdown of the Magi1 scaffold protein attenuates established neuropathic pain behavior during nerve entrapment injury

Authors: *M. MARTIN[1], A. BHATTACHARJEE[2], G. GUERRERO[3];
[1]State Univ. of New York, Buffalo, Buffalo, NY; [2]Pharmacol. and Toxicology, SUNY-Buffalo, Buffalo, NY; [3]State Univ. at Buffalo, Buffalo, NY

Abstract: Entrapment neuropathies are the most common cause of neuropathic pain. They occur when a nerve becomes compressed, or entrapped, between two other structures in the body. Studies have shown that there is an increase in NaV1.8 sodium channel trafficking and insertion into the neuronal membrane in preclinical models of nerve entrapment injury. The increase in NaV1.8 channel activity is thought to underlie neuropathic pain behavior. While the scaffold proteins responsible for sodium channel and membrane stabilization in DRG neurons have not been fully characterized, our published studies identified the WW domain-containing scaffold protein called Magi1 as a potential candidate (PMID: 30860870). After establishing neuropathic pain using the sciatic nerve cuff model of entrapment injury in male and female mice, Magi1 was then genetically knocked down using an *in vivo* sciatic nerve knockdown approach strategy to test the consequences of Magi1 deficiency on established pain. The Hargreaves thermal, von Frey fiber mechanical sensitivity, and dynamic weight-bearing assays were used to confirm and monitor neuropathic pain. Our previously published study showed that thermal hyperalgesia and dynamic weight-bearing mirror each other, encoding ongoing pain while von Frey mechanical sensitivity persists (PMID: 34917858) in this model**.** After Magi1 was knocked down and confirmed by Western analyses, in the Hargreaves thermal assay, we found that there was an increase in the thermal withdrawal latency during Magi1 knockdown in neuropathic mice compared to the scrambled shRNA-treated mice. We also saw an increase of the percent weight borne on the ipsilateral paw in the Magi-1 knockdown mice using dynamic weight-bearing measurements compared to scrambled shRNA-treated mice. For mechanical von Frey sensitivity, there was an increase in the withdrawal threshold of the Magi1 knockdown mice. We also noted sex differences in response to the Magi1 knockdown. Magi1 is a potential target for analgesia for nerve entrapment-induced neuropathic pain. Future studies will examine the electrophysiology properties of DRG neurons after Magi1 knockdown and nerve entrapment.

Disclosures: M. Martin: None. **A. Bhattacharjee:** None. **G. Guerrero:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.36

Topic: D.02. Somatosensation – Pain

Support: NIH Grant NS102432

Title: Toll-like receptor 4 in development of mechanical and thermal sensitization, and dorsal root ganglion neuro-inflammatory responses after intraplantar Complete Freund's Adjuvant

Authors: *J. LEMES[1], J. NAVIA PEALEZ[2], G. GONCALVES DOS SANTOS[1], B. DRUMMOND[1], J. ZHANG[1], K. MALANGE[1], J. W. LU[2], L. GONZALEZ[2], M. YUN[1], M. CORR[2], Y. MILLER[2], T. YAKSH[1];
[1]Dept. of Anesthesiol., [2]Dept. of Med., Univ. of California San Diego, San Diego, CA

Abstract: Accumulating evidence shows that dorsal root ganglion TLR4 contributes to the development of neuropathic and inflammatory pain phenotypes, through signaling that reflects communication between neuronal and immune cells. In the present work, we examined the role of TLR4 in the mechanical and thermal thresholds changes (von Frey and Hargreaves test), following unilateral intraplantar injections of CFA (complete Freund's adjuvant) in WT and $Tlr4^{-/-}$male mice. Using flow cytometry, we evaluated the number/phenotypes of DRG macrophages, satellite cell activation (GFAP expression), and TRPV1 channels expression in the ipsi (I) vs contralateral © dorsal root ganglia (L3 to L6) of WT and KO, eight days after CFA. The behavioral data showed that a single intraplantar injection of CFA (20 µl) produced a prolonged (> 8 d) increase in paw edema (Clinical score I: 2.231 ± 0.166*) and decreased hind paw tactile (I: 0.619 ± 0.037 vs ©: 1.300 ± 0.059 gm*) and thermal (I: 3.850 ± 0.323 vs ©: 6.450 ± 0.609 sec*) thresholds in the ipsilateral vs contralateral paw WT mice. In contrast, following CFA, $Tlr4^{-/-}$ mice showed a significant reduction on days 6 and 7 in the mechanical allodynia and thermal hyperalgesia as well as a reduction in paw thickness. In the flow cytometry analysis, we observed an increase in satellite glial cell activity and TRPV1 expression in the ipsilateral DRGs of WT mice but not in the $Tlr4^{-/-}$ DRGs. M1 phenotype macrophages counts were significantly elevated in the ipsi vs contralateral only in DRGs from WT mice. These findings suggest that TLR4 receptors exhibit influence in the inflammatory process induced by CFA through increasing polarization of macrophages to pro-inflammatory phenotype, expression of TRPV1 channels, and enhancing satellite glial cells activity in the DRGs which, together, leads to nociceptor sensitization reflecting in peripheral mechanical allodynia and thermal hyperalgesia. (*: $p<0.05$, mean with SEM) (NS102432).

Disclosures: J. Lemes: None. **J. Navia Pealez:** None. **G. Goncalves Dos Santos:** None. **B. Drummond:** None. **J. Zhang:** None. **K. Malange:** None. **J. W. Lu:** None. **L. Gonzalez:** None. **M. Yun:** None. **M. Corr:** None. **Y. Miller:** None. **T. Yaksh:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.37

Topic: D.02. Somatosensation – Pain

Support: Canadian Institutes of Health Research (CIHR) grant FDN 159906
Student Scholarship for excellence from Wallonie-Bruxelles International

Title: Altered subpopulations of mechano-sensitive primary afferents in response to peripheral nerve injury.

Authors: *J. DAMBLON[1], F. WANG[2], Y. DE KONINCK[3];
[1]CERVO Brain Res. Ctr. - Univ. Laval, Quebec, QC, Canada; [2]CERVO Brain Res. Ctr., Quebec, QC, Canada; [3]CERVO Brain Res. Ctr., Laval Univ. / IUSMQ, Quebec, QC, Canada

Abstract: Neuropathic pain is a condition with a high incidence, caused by a disease or lesion of the peripheral or central nervous system. It affects up to 10% of the population globally and has a huge socioeconomic cost. However, it is often undertreated due to its complex pathophysiology that is still not fully elucidated. Mechanical allodynia (innocuous stimuli becoming painful) is the major and most debilitating symptom of neuropathic pain. There is ongoing debate between two potential mechanisms underlying mechanical allodynia: a decrease of the activation threshold of the high-threshold nociceptors vs. an altered signaling of low-threshold mechanoreceptors to transit pain. To address this question, we quantified the mechanical responses of nociceptors from both nerve-injured (n=46) and sham-control mice (n=54). We performed in-vivo calcium imaging on lumbar DRGs from anesthetized NaV1.8-GCaMP6s mice, in which most nociceptors were labelled with the genetically-encoded calcium indicator, GCaMP6s. After mapping the receptive field on the glabrous hind paw, we applied a series of indentation stimuli, of varying pressure, to the center of the receptive field using a force feedback-controlled mechanical stimulator. In both sham-control and nerve-injured mice, higher pressure recruited more neurons and induced stronger activation of individual neurons in a graded fashion. Interestingly, a high proportion of these nociceptors (42% in control mice and 39% in nerve-injured mice) show sustained responses to innocuous pressure (<3.3g/mm2). The size of the receptive field and the activation threshold of the overall population of NaV1.8 neurons appeared unaltered after nerve injury. However, further analysis of the activation threshold revealed two subpopulations of mechanoreceptors in control mice: low-threshold and high-threshold nociceptors. Following nerve injury, the prevalence of the high threshold subpopulation decreased, and the remaining high-threshold neurons had an increased activation rate as compared to those from control mice. Thus, our results do not support the theory of a decrease of the activation threshold of nociceptors. Instead, it points towards a restructuring of a high-threshold sub-population after nerve injury as a novel mechanism of mechanical allodynia.

Disclosures: **J. Damblon:** None. **F. Wang:** None. **Y. De Koninck:** None.

Poster

541. Neuropathic and Inflammatory Pain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 541.38

Topic: D.02. Somatosensation – Pain

Title: The NIH HEAL Initiative Preclinical Screening Platform for Pain (PSPP) efforts to accelerate development of non-opioid, non-addictive pain therapeutics

Authors: *S. A. WOLLER, S. SHARMA, A.-S. WATTIEZ, D. KEMPEGOWDA, S. IYENGAR;
NIH/NINDS, Rockville, MD

Abstract: The NIH HEAL Initiative is a trans-agency effort to providescientific solutions to stem the opioid crisis. Within the NIH HEAL Initiative,NINDS developed the Preclinical Screening Platform for Pain (PSPP) tofacilitate the identification and development of new non-opioid, non-addictivepain therapeutics.PSPP provides researchers from academic, industry, and governmentinstitutions, within the US and internationally, an efficient, rigorous,one-stop *in vivo* resource at no cost, to accelerate development of newpain therapeutics including small molecules, biologics, natural products, anddevices for the treatment of pain. The asset owner's intellectual property andconfidentiality are protected.For accepted participants, the asset is rigorously evaluated underPSPP direction by an NINDS contract facility, PsychoGenics Inc., in a blindedand confidential manner. Assets are evaluated using a tiered approach thatincludes in vitro screens assessing functional activity against a broad panelof human recombinant targets associated with abuse, dependence, and off-targetactivity; brain and plasma protein binding; pharmacokinetics studies; sideeffect profile; efficacy in a suite of pain-related models using evoked andnon-evoked endpoints; and abuse liability. All in vivo studies are conducted inmale and female SD rats using group sizes determined by power analysis. Data and reports are given to the asset ownerat the end of each study. The tiered approach to evaluation of assets, andinformation about each model and endpoint currently used within the program willbe publicly available. Evaluation ofassets in PSPP can provide a key step in transitioning NIH HEAL Initiativepreclinical programs into clinical programs. A key feature of the PSPP programis the flexibility to continuously acquire and validate innovative models andendpoints that more closely represent human pain conditions.This presentation will highlight data generated within the programwith clinically used drugs, including pregabalin and diazepam, and illustratethe rigorous nature of evaluation of novel non-opioid, non-addictive therapeuticapproaches. Results illustrate the efficacy of a drug clinically used in painconditions, e.g., pregabalin, in multiple models and endpoints in the contextof pharmacokinetics, side effect profiles as well as abuse liability potentialand highlight differences from the profile of diazepam, a drug not clinicallyapproved for pain. In summary, the NINDS PSPP program strives to accelerate thedevelopment of novel non-opioid, non-addictive therapeutics for pain.

Disclosures: S.A. Woller: None. **S. Sharma:** None. **A. Wattiez:** None. **D. Kempegowda:** None. **S. Iyengar:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.01

Topic: D.04. The Chemical Senses

Support: Whitehall Foundation Grant
NSF-Simons Center for Quantitative Biology Pilot Grant

Title: An unbiased transcriptomic analysis of taste receptor cells

Authors: *Y. YU, H. LEE;
Northwestern Univ., Evanston, IL

Abstract: Sense of taste is essential for evaluation of potential food items. Each taste receptor cell (TRC) in the mammalian taste bud expresses transmembrane receptors specialized to detect a single quality - sweet, umami, bitter, sour, or salty. With a short life span of only ~2 weeks, TRCs need to be constantly replenished and accurately wired to their respective sensory neurons for the animal to maintain taste sensation. Recent work has identified LGR5 as a genetic marker for progenitor cells responsible for giving rise to TRCs and implicated major signaling pathways (e.g. Shh, Wnt, Notch) in the differentiation process. However, the precise genetic mechanisms that regulate taste tissue homeostasis and define TRC specification remains a mystery. To resolve this, we generated single cell transcriptomics data from taste tissue. We dissociated taste papillae from mice (C57BL/6, male, n=10, age=10 weeks) and performed single-cell RNA sequencing using the 10X Genomics platform. Our approach retrieved ~16,800 cells across 19,034 expressed genes. With the Seurat V4.0 package, we observed robust clustering of ~3,100 TRCs and basal Lgr5+ progenitor cells. By applying mathematical modeling to compare clusters, we found transcripts that are differentially expressed in TRCs for each taste quality. Taken together, our data advances the current understanding of transcriptomic expression of taste system and opens the opportunity to determine key factors that drive TRC specification.

Disclosures: Y. Yu: None. **H. Lee:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.02

Topic: D.04. The Chemical Senses

Support: NIDCD Grant DC015799

Title: Egr4 is critical for postnatal cell-fate determination and phenotypic maintenance of geniculate ganglion neurons that underlie sweet and umami taste

Authors: *D. DUTTA BANIK[1,2], L. J. MARTIN[1,2], B. A. PIERCHALA[1,2];
[1]Indiana Univ. Sch. of Med., Indiana University-Purdue Univ. Indianapolis, Indianapolis, IN;
[2]Anatomy, Cell Biol. & Physiol., Indiana Univ. Sch. of Med. Stark Neurosciences Res. Inst., Indianapolis, IN

Abstract: Early Growth Response 4 (EGR4) belongs to the EGR family of zinc-finger transcription factors and has a critical role in the development of several cell types such as spermatogonia and Dorsal Root Ganglia (DRG) neurons. During our investigation of novel genes important for the development of Geniculate Ganglion (GG) neurons, EGR4 was identified as a gene enriched in PHOX2B-positive oral sensory neurons. Its function in the gustatory system is currently unknown. We observed a severe loss of PHOX2B expression in oral sensory neurons of the GG with a concomitant increase in the BRN3A+ pinna somatosensory neurons. Deletion of EGR4 also disrupted the cell fate determination of these neurons resulting in loss of several known subpopulations of GG oral sensory neurons. A significant reduction in the chemosensory innervation of taste buds as well as taste cell number in Fungiform papillae were also observed in the *Egr4*$^{-/-}$ mice. Chorda tympani nerve recordings demonstrated that *Egr4*$^{-/-}$ mice exhibit deficits in responses to sweet (sucrose) and umami taste stimuli. To understand the downstream mechanism of EGR4 function, we performed RNA-seq on the GG from *Egr4*$^{+/+}$ and *Egr4*$^{-/-}$ mice. We found that axon guidance proteins such as PLEXINB3, ROBO2, and DRAXIN were significantly downregulated in *Egr4*$^{-/-}$ mice. On further investigation, these proteins were also significantly reduced in the axon terminals innervating taste buds in Fungiform papillae. These results indicate that EGR4 plays an integral role in cell fate determination of oral sensory neurons in the GG and controls the expression of the axon guidance molecules required for the proper neuronal innervation and/or synapse formation in taste buds.

Disclosures: D. Dutta Banik: None. **L.J. Martin:** None. **B.A. Pierchala:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.03

Topic: D.04. The Chemical Senses

Support: NIH Grant SC2GM130411
Whitehall Grant 2019-12-41

Title: Revisiting the taste map: regional specialization of the tongue revealed by gustatory ganglion imaging

Authors: *L. J. MACPHERSON[1], B. FOWLER[1], S. HUMAYUN[1], J. YE[2], H. LEE[2];
[1]UT San Antonio, The Univ. of Texas at San Antonio, San Antonio, TX; [2]Northwestern Univ., Northwestern Univ., Evanston, IL

Abstract: Edwin Boring's mis-interpretation of a 1901 study popularized the idea of a tongue map with discrete regions of taste sensitivity across the tongue surface. While this representation persisted in textbooks for decades, it was largely discredited by the observation that most taste buds express the full array of taste receptors, irrespective of their position on the tongue. However, gustatory papillae of the anterior and posterior tongue are derived from different

developmental origins and innervated by separate peripheral sensory afferents. Gustatory information from the anterior tongue is relayed by geniculate ganglion neurons and from the posterior tongue by neurons of the petrosal portion of the jugular/nodose/petrosal complex. Here, we revisit the regional specialization of the tongue by using in vivo calcium imaging in mice to compare the encoding of taste information in the geniculate and petrosal ganglia, at single neuron resolution. Our data support an anterior/posterior specialization of taste information coding from the tongue to the ganglia, with petrosal neurons more responsive to umami or bitter and less responsive to sweet or salty stimuli than geniculate neurons. Does this regional difference have any functional consequences? We found a significant effect on salivation based on regional application of taste stimuli; umami promotes salivation when applied to the posterior, but not anterior, tongue. In summary, this suggests a functional taste map of the mammalian tongue where the anterior and posterior taste pathways are differentially responsive to specific taste qualities, and differentially regulate downstream physiological functions of taste, such as promoting salivation.

Disclosures: **L.J. Macpherson:** None. **B. Fowler:** None. **S. Humayun:** None. **J. Ye:** None. **H. Lee:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.04

Topic: D.04. The Chemical Senses

Support: Royal Society Newton International Fellowship NIF/R1/19368
NSERC PDF-546125-2020

Title: Temporal responses of bumblebee gustatory neurons encode sugars

Authors: *R. H. PARKINSON, G. A. WRIGHT;
Dept. of Zoology, Univ. of Oxford, Oxford, United Kingdom

Abstract: Taste permits the recognition of valuable nutrients and the avoidance of potential toxins. The sense of taste is generally understood to be the intensity-dependent detection of stimuli in a few broad categories (e.g., sweet, bitter, etc.), with little ability to distinguish compounds within a single modality. In *Drosophila*, a single taste sensillum houses between 2-4 gustatory receptor neurons (GRNs), with a single GRN from each sensillum responsive to sugars. Bumblebees rely on sugary nectar as their primary energy source, and they have adapted a specialized taste system for the detection of sugars at the expense of high resolution in other taste modalities. Within a single bumblebee galeal taste sensillum there are four GRNs, and we show that three of these GRNs are responsive to sugars. When stimulated with some sugars, the GRNs fire in a bursting pattern, while other sugars do not elicit bursting at any concentration. A clustering analysis of the temporal responses to sugars over a concentration gradient predicts that

bees can distinguish sugars in a few broad categories. We tested this behaviourally and found that bees perform better than the clustering algorithm predicts: bees can distinguish a range of sugars, including those found in nectar (sucrose, fructose, glucose) and honeydew (maltose, melezitose). This suggests that bumblebees have evolved a specialized taste system with high acuity for sugars.

Disclosures: R.H. Parkinson: None. **G.A. Wright:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.05

Topic: D.04. The Chemical Senses

Support: NIH Grant DC006666
NIH Grant DC007703

Title: Mixture of tastes is more than the addition of its components: Evidence from taste responses in the gustatory cortex

Authors: *J.-Y. LIN, D. KATZ;
Psychology, Brandeis Univ., Waltham, MA

Abstract: Tastes of a food critically contributes how the food is perceived and evaluated. Accordingly, the decision to eat or not to eat a specific food, unsurprisingly, depends on the quality of processing taste information within the brain. An area that undertakes this task is the gustatory cortex (GC). Over the past 20+ years, we have found that GC taste responses are dynamic and evolve through various epochs: following the initial detection of taste presented on the tongue, GC activity becomes identity-related and then transitions into a state dominated by palatability (or decision)-related activity. Given that food often is comprised of several of the primary tastes (e.g., salty and sweet), the current research aimed to determine how a mixture of tastes is processed in GC via an active licking paradigm. During each experimental session, rats with multi-channel implants in GC were given the opportunity to lick for a sucrose (0.1 M; S), NaCl (0.1M, N), or mixture (SN) across trials with water rinses interleaved between these taste trials. Preliminary results indicate that the GC neurons that responded to mixtures and their activity differ from those responding to S or N, indicating that the mixture processing in GC cannot be simply understood as an addition of S and N, but rather, it reflects a unique property of mixtures. These results were discussed in the context of configural perception, likely via the mechanism of within-compound associations. Future directions will involve further analyses and experiments to investigate whether GC taste response dynamic changes following the development of configural perception.

Disclosures: J. Lin: None. **D. Katz:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.06

Topic: D.04. The Chemical Senses

Support: NIDCD grant RO1-DC006914

Title: Neural population response to taste and food in the parabrachial nucleus of the pons in the awake unrestrained rat using in vivo one-photon calcium imaging

Authors: F. P. O'CONNELL[1], J. D. VICTOR[2], ***P. M. DI LORENZO**[1];
[1]Psychology, State Univ. of New York, Binghamton Integrative Neurosci., Binghamton, NY;
[2]Feil Family Brain and Mind Res. Inst., Weill Cornell Med. Col., New York, NY

Abstract: Although brainstem responses to taste stimuli have been well-documented, the function of the taste system in natural feeding remains unexplored. Based on previous electrophysiological data, we tested the hypothesis that the taste system is strongly engaged while exploring food, its taste and smell, but becomes relatively disengaged when food is being consumed. Here, we used *in vivo* one-photon Ca^{2+} imaging of cells in the parabrachial nucleus of the pons (PbN) of awake, unrestrained rats as they (i) licked representatives of four basic taste qualities (0.1 M sucrose, 0.1 NaCl, 0.0167 M citric acid, 0.0001 M quinine) as well as artificial saliva (AS) presented alone or paired with either peanut or chocolate odor, and (ii) ate either chocolate or peanuts presented in wells (recessed open boxes at the corners of the experimental chamber). In separate surgeries 3 wks apart, rats were prepared for Ca^{2+} imaging by infusion of GCamp7s and implantation of a 1 mm dia. GRIN lens above the PbN. When recovered, rats were water-deprived and a miniscope (Inscopix, Inc.) was mounted above the GRIN lens. For the 20 min lick session, rats were placed in a chamber with a lick spout for delivery of taste stimuli, AS, AS+peanut odor or AS+chocolate odor. Each taste trial consisted of 10 consecutive taste stimulus licks preceded and followed by 6 AS licks delivered on a VR5 schedule. For odor trials, AS was presented for 10 consecutive licks, the first of which initiated a 3 s puff of an odorant delivered through a port orthogonal to the lick spout. Odorants were generated by flowing compressed air over either chopped salted dry roasted peanuts or chopped milk chocolate (35% cacao, 67% sugar). A 20 min eating session followed the lick session immediately. Doors to the wells were removed and 3 gms of peanuts or chocolate were placed in separate wells, to which the rats had free access. A video camera (CinePlex, Plexon, Inc.) was used to record behavior in the experimental chamber. Results from 2 rats thus far have shown that prior to consumption all taste and odor stimuli evoked spatially widespread responses (>30 cells/rat) with no gustotopic segregation apparent. When rats ate either peanuts or chocolate, however, the number of responsive cells was spatially restricted to a small cluster of cells (~7-10) located in the caudo-medial field. Cells in this cluster also responded to all tastants and odorants tested, while each individual taste or odor stimulus additionally evoked unique spatial patterns of response scattered

across the field. These data suggest that, in the PbN, taste and odor are represented by a dense, distributed code, while food is represented by a sparse and spatially restricted code.

Disclosures: F.P. O'Connell: None. **J.D. Victor:** None. **P.M. Di Lorenzo:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.07

Topic: D.04. The Chemical Senses

Support: NIH NIDCD DC016833

Title: Changes in Patterns of Gustatory Cortex Response After Conditioned Taste Aversion

Authors: M. A. RAYMOND[1], *M. L. FLETCHER[2], J. D. BOUGHTER[1];
[1]Anat. & Neurobio., Univ. of Tennessee Hlth. Sci. Ctr., Memphis, TN; [2]Anat. & Neurobio., The Univ. of Tennessee Hlth. Sci. Ctr., Memphis, TN

Abstract: The role of gustatory cortex (GC) in the processing sensory input and generation of behavior associated with taste is still not fully understood. It has been previously suggested that after condition taste aversion (CTA) learning, the neural representation of a palatable conditioned taste stimulus is altered, becoming more similar to innately aversive tastes. We investigated the neural representation of CTA in the GC of adult male and female mice. Using calcium imaging via head-mounted miniscopes, we recorded the activity of a consistent population of 1061 cells from 8 awake, behaving mice as they progressed through a CTA paradigm, permitting an unprecedented investigation of neural correlates of behavior and learning in real time. Control animals (6 mice, 1037 cells) experienced the same taste exposure paradigm, but did not undergo aversion conditioning. Over the course of several days, animals were first presented with a panel of several tastes, and after conditioning an aversion to one previously accepted taste solution, the panel was presented again. A subset of the conditioned animals (4 mice) went through additional extinction training, where the conditioned stimulus was presented repeatedly until its consumption recovered to pre-conditioning levels. We found that responses to different tastes were initially uncorrelated, but responses to accepted tastes began to intercorrelate over successive exposures. After a CTA to sodium chloride (NaCl) was formed, however, responses to that taste reverted to their previously uncorrelated state. This pattern of change appeared related to, but not fully explained by, quantity of ingestion. The conditioned taste stimulus became less similar to accepted tastes, and more similar to taste solutions that are innately rejected. Notably, cells that had previously been categorized as NaCl-best (N-units) greatly reduced their response to NaCl, and cells previously categorized as bitter- or quinine-best (Q-units) substantially increased their NaCl response. Reversing the aversion via extinction also reversed the effects of learning in GC; correlations between responses to NaCl and those of other accepted tastes again increased their intercorrelation, and tuning changes revert, such that N-

units classified prior to conditioning resumed responding to NaCl, and Q-units return to responding primarily to quinine. In sum, these findings describe activity in gustatory cortex that is initially disorganized, but following behavioral experience becomes increasingly commensurate with taste palatability and ingestive decision making.

Disclosures: M.A. Raymond: None. **M.L. Fletcher:** None. **J.D. Boughter:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.08

Topic: D.04. The Chemical Senses

Title: The role of cortical D1R-expressing neurons in taste-based sensorimotor transformations

Authors: *J. CHEN[1], A. FONTANINI[2];
[1]Stony Brook University, Neurobio. & Behavior, Stony Brook, NY; [2]Dept. of Neurobio. & Behavior, SUNY Stony Brook, Stony Brook, NY

Abstract: To survive, animals must use gustatory stimuli to guide oromotor behaviors like licking and chewing. The anterior-lateral motor cortex (ALM) is involved in the genesis of sensorimotor decisions related to licking. Although licking and taste are invariably linked, it remains unknown if ALM plays a role in the linking the sensorimotor transformation to taste stimuli to lick decisions, and how the underlying circuits are modulated during this process. One candidate mechanism for taste-based sensorimotor transformations is dopamine transmission through D1 receptor (D1R). Blockade of D1R signaling within ALM affects both sensory cue-evoked responses and motor planning of licking, suggesting a potential role for D1R-expressing (D1R+) neurons in sensorimotor guided licking. We tested the hypothesis that D1R+ neurons in ALM exhibit patterns of activity that represent taste cues and preparation of licking, and that these specific and distinct representations are required to drive lick decisions during a taste-guided 2-alternative choice (2-AC) task. In this task, mice must use two taste cues to guide different licking actions (lick left vs lick right) after a delay period. Two-photon calcium imaging in the superficial layers of ALM (<350 um) was performed using transgenic D1R reporter mice to identify and simultaneously record populations of D1R+ neurons and non-D1R expressing neurons (D1R-) during performance in the task. Individual D1R+ and D1R- neurons exhibit epoch-specific responses during the sample (S), delay (D) and response (R) periods of the task, representing taste cue, preparation of licking and lick execution, respectively. Examination of taste coding for single neurons show weak tuning to specific taste cues during the sampling. However, analysis of lick direction coding revealed a stronger bias for contralateral lick trials in D1R+ responses when compared to D1R- responses during both the delay and choice periods. These preliminary findings suggest D1R+ neurons in the superficial layers of ALM represent a unique subpopulation with a preference for contralateral lick choice. To determine if activity of D1R+ neurons is required in taste-guided licking, we will rely on optogenetics to bilaterally

silence D1R+ neurons during different behavioral periods of the 2-AC task. Based on findings from our imaging experiments, we expect silencing during the sample period to not perturb task performance while silencing D1R+ neurons during the delay to produce an ipsilateral bias in lick performance. Altogether, these findings will provide novel understanding of the link between gustatory decision-making and cortical neurons expressing D1Rs.

Disclosures: J. Chen: None. **A. Fontanini:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.09

Topic: D.04. The Chemical Senses

Support: NIH Grant DC007703
HHMI Grant GT15942
NIH Grant MH019929

Title: Investigating the cortical signals driving sensorimotor transformations leading to consummatory responses in rats

Authors: *N. BAAS-THOMAS[1], D. B. KATZ[2];
[1]Brandeis Univ. Grad. Neurosci. Program, Waltham, MA; [2]Dept Psychol, Brandeis Univ., Waltham, MA

Abstract: The consumption system is an ideal model with which to study the neural processes and pathways guiding discriminative, ethologically-relevant behavior in response to sensory stimuli. When a potential taste stimulus reaches the tongue, the CNS has one simple function - to determine whether that stimulus should be consumed or expelled from the mouth. Rats (and many other mammals) produce discriminative orofacial movements reflecting the reaching of a consummatory decision: lateral tongue movements (LTMs) for ingestion and gapes for rejection. Previous work has demonstrated that sensory (gustatory) cortex (GC) is integrally involved in reaching such decisions: taste responses of GC ensembles progress through three firing-rate "epochs", clocking the sensori-motor transformation that ends in the consumption decision. Furthermore, the relationship between the transition to decision-related firing and consummatory behavior initiation has proven causal. However, GC is not in and of itself a motor structure - a central pattern generator localized to the parvocellular reticular formation (RF) directly drives gapes and LTMs. We propose, therefore, that GC provides a modulatory signal to RF which guides the selection and initiation of one of the two consummatory responses. Using optogenetics, I will briefly (0.5s) inhibit GC->RF projections in freely moving rats after taste delivery, while electrophysiologically monitoring behavior - specifically the activity of two orofacial muscles - and neural activity in GC. I predict that this inhibition will perturb stereotyped consummatory responses, but only if it precedes the decision-related epoch of GC

activity. These findings will ultimately enrich our understanding of how sensory information is transformed into appropriate behaviors.

Disclosures: N. Baas-Thomas: None. **D.B. Katz:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.10

Topic: D.04. The Chemical Senses

Support: T32DC000044
R01DC019326

Title: Intraoral thermal processing in the gustatory cortex of awake mice.

Authors: *C. BOUAICHI, R. VINCIS;
Biol. Sci., Florida State Univ., Tallahassee, FL

Abstract: In the past decades, many electrophysiological studies in behaving rodents have described how neurons in the gustatory cortex (primary taste cortex, GC) process taste information. In addition, a growing body of experimental work in humans and primates - as well as pioneering works in anesthetized rats - indicates that GC neurons respond also to non-gustatory components of intra-oral stimuli, including temperature, a salient sensory feature of of food and beverages. While these data implicate the GC as a potential key brain region for the integration of taste and thermal orosensory inputs, they stop short of supplying a fine-grained analysis of its neural responses, and many questions remain.Here, using fiber-photometry and extracellular recording (tetrodes and silicon-based probes) we aim to provide a complete neurophysiological assessment of how thermal orosensory inputs shape GC activity in alert mice. Specifically, we tested 1) whether and how neurons in the GC of actively licking mice are modulated by changes in the temperature of chemically inert drinking solutions and 2) if thermal responses are organized across the dorso-ventral axis (granular, dysgranular and agranular) of the GC. Licking and neural activity was recorded in mice trained to experience (on a fixed ratio schedule) a drop of cool (14°C), ambient (25°C), or warm (36°C) water.Overall our results show that GC processes thermal intra-oral information at both a population and single-unit level without an apparent topographical organization. In conclusion, our data shows that temperature is a salient intra-oral cue represented in the taste cortex and suggests the GC is cortical region important for the integration of thermal and chemosensory stimuli present in food and beverages.

Disclosures: C. Bouaichi: None. **R. Vincis:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.11

Topic: D.04. The Chemical Senses

Support: NIH Grant R01DC013770-03

Title: Characterization of Gastrin-Releasing Peptide-Expressing Neurons in the Gustatory Cortex of Mice

Authors: *L. CZARNECKI[1], J. S. CHEN[3], O. K. SWANSON[2], S. SWAMINATHAN[4], A. MAFFEI[2], A. FONTANINI[5];
[2]Neurobio. and Behavior, [1]SUNY Stony Brook, Stony Brook, NY; [3]Neurobio. & Behavior, Stony Brook University, Neurobio. & Behavior, Edison, NJ; [4]SUNY at Stony Brook, Stony Brook, NY; [5]Dept. of Neurobio. & Behavior, SUNY Stony Brook, Setauket, NY

Abstract: Gastrin-releasing peptide (GRP) is endogenously expressed peripherally and centrally. Peripherally, GRP is involved in enteric activities related to metabolism and digestion. Centrally, GRP has many actions including involvement in circadian rhythm, learning and memory, the spinal cord itch reflex, as well as ingestion. Intravenous administration in humans, and peripheral or intracranial administration in animal models, appears to act as a meal termination signal. While the pharmacological effects of GRP in ingestion have been relatively well described, the neural basis of its central action remains unknown, as is the role of GRP within the taste pathway. Here, we describe GRPergic circuits in the gustatory cortex (GC). GRP-expressing (GRP+) neurons make up approximately 5% of neurons in GC. The distribution shows greatest cell density in layers II/III and IV of dysgranular and granular GC where GRP is expressed in up to ~15% of neurons. Whole cell patch clamp recordings revealed pyramidal cell-like intrinsic properties. To begin investigating the functional role of GC GRP+ neurons, we relied on 2-photon calcium imaging. GCaMP7f was expressed in GC of animals genetically expressing tdTomato in GRP+ cells. Taste-evoked responses of GRP+ neurons were assessed in mice sampling quinine, citric acid, sodium chloride, sucrose, sucralose, water, and Ensure. Preliminary analysis of responses suggests that GRP+ cells respond to tastants, licking, and taste-predictive cues in similar proportion to GRP- cells. Calcium transients were largest in a window ~3-5 sec after taste delivery. In this window, GRP+ neurons show a different taste tuning profile than GRP- neurons, with GRP+ neurons having a larger proportion of sucrose-best neurons. Using viral tracing and electrophysiological methods, we found that GRP-expressing neurons in GC send a dense, functional projection to the basolateral nucleus of the amygdala (BLA). Previous work has shown that GRP administration into the amygdala reduces meal size and increases time between eating bouts. Since GC may be providing a source of GRP into the BLA, this suggests that GC GRP-expressing neurons may be involved in integrating gustatory and homeostatic signals. Future experiments will explore how the activity of GRP+ neurons in GC is modulated by the homeostatic state of the animal.

Disclosures: L. Czarnecki: None. **J.S. Chen:** None. **O.K. Swanson:** None. **S. Swaminathan:** None. **A. Maffei:** None. **A. Fontanini:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.12

Topic: D.04. The Chemical Senses

Support: DC007703

Title: Consideration of individual rat preferences improves correlation in gustatory cortex activity

Authors: E. CROUSE, K. C. MAIGLER, *D. KATZ;
Brandeis Univ., Waltham, MA

Abstract: Rodents, like humans, appear to show individual differences in preferences for tastes. As this preference or palatability value is a crucial variable in the consumption decision, it is important for taste researchers to determine palatability reliably. Historically taste research uses the lick-counts for each tastant in order to establish relative palatability rank order, but there are many other metrics (first bout-length, orofacial behaviors) that can be used to define palatability in rats. A concern with all of these measures is variability—behavior is typically averaged across groups of subjects to generate the palatability rank order. However, we demonstrate that there is significant variability in the decision phase of the primary taste cortex (GC) evoked response. Here, we show this variability, which has traditionally been assumed to be noise, is reflective of individual differences in relative palatability preferences. Using the brief-access task, we collect individual rat palatability data and then record cortical responses from these same rats in GC while delivering tastes intra-orally. Comparing neural data from traditional rank-order calculations to individualized rat palatability sets confirms activity in GC during the palatability encoding phase is better correlated with individualized data. This result underscores the importance of considering inter-rat variability in both brain and behavior, and is novel reinforcing evidence that the late epoch of taste processing in GC genuinely tracks taste palatability.

Disclosures: E. Crouse: None. **K.C. Maigler:** None. **D. Katz:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.13

Topic: D.04. The Chemical Senses

Support: Productos Medix 3247
Fundación Miguel Alemán
Catedra Marcos Mochisky

Title: Diet induced obesity enhanced neuronal responses to sucrose in the lateral hypothalamus of rats

Authors: *O. X. GUERRERO GUTIERREZ[1], M. A. VILLAVICENCIO CAMARILLO[2], E. FONSECA[3], C. I. PEREZ[4], R. GUTIERREZ[5];
[1]CINVESTAV, Gustavo A. Madero, Mexico; [2]Ctr. De Investigación Y Estudios Avanzados, Mexico, Mexico; [3]Ctr. de Investigación y Estudios Avanzados, Princeton, NJ; [4]Inst. of Neurobio., Univ. Nacional Autonoma de Mexico, Juriquilla, Mexico; [5]Pharmacol., CINVESTAV - IPN, Mexico City, Mexico

Abstract: The overconsumption of palatable food, like sucrose and fat, is one of the main factors that contribute to the developing obesity epidemic. Apart from the cardiovascular problems it causes, obesity has been shown to alter taste perception through changes in the tongue's proteome and transduction pathways. In the brain, the attraction towards palatable food is mediated through the Lateral Hypothalamic Area (LHA). However, how obesity alters the gustatory circuit's neural responses to palatable food remains poorly understood. To answer this question, we studied how a DIO model would affect LHA's electrophysiological responses to sucrose. To this end, freshly weaned Wistar rats were fed with a High-Fat Diet (HFD, 45% fat) to induce obesity. Then, they were trained in a Brief Access Taste Test (BATT) to measure their evoked sucrose palatability responses, while LHA single-unit activity was recorded. We found that DIO rats exhibited an altered licking microstructure, resulting in a blunted behavioral response to sucrose's palatability. Recordings in the LHA unveiled a diminished lick-spike coherence in DIO compared to Lean control rats, demonstrating that the coupling between LHA spikes with rhythmic licking is impaired in a DIO model. In contrast, an ensemble of palatability-related neurons in the LHA exhibited increased excitability (and cell number), reflecting an exalted neuronal sensitivity to sucrose in DIO rats. A second ensemble, in which sucrose inhibited their activity, displayed a selective reduction in their number of neurons. Moreover, palatability related neurons in DIO rats decoded sucrose concentrations faster and more accurately than Lean rats. To investigate the neuronal subpopulations involved in this process, we used transgenic mice (Vgat-ires-Cre) that express channelrhodopsin-2 (ChR2) on LHA neurons. With an optogenetic tagging, we were able to discern between GABAergic and non-GABAergic LHA subpopulations. We observed that GABAergic neurons are more involved in palatability-related modulations as opposed to non-GABAergic. This suggests that GABAergic neurons are more affected by obesity. In sum, we showed that the activity of LHA underwent multiple neuronal adaptations in response to exposure to an HFD during childhood and adolescence. These brain adaptations comprise a diminished lick-spike synchronization and perhaps a compensatory mechanism better decoding of sucrose concentrations that paradoxically correlated with the blunted sucrose palatability response observed in obese rats.

Disclosures: O.X. Guerrero Gutierrez: None. **M.A. Villavicencio Camarillo:** None. **E. Fonseca:** None. **C.I. Perez:** None. **R. Gutierrez:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.14

Topic: D.04. The Chemical Senses

Support: Z01AA000135
1ZIANR000035-01
Y1AA-3009

Title: Influence of Excessive Prenatal Caffeine Exposure on Total Sugar Intake, Brain Structure, and Reward Sensitivity in Adolescents

Authors: *K. AGARWAL[1], P. MANZA[2], H. A. TEJEDA[3], A. B. COURVILLE[4], N. D. VOLKOW[5], P. V. JOSEPH[1];
[1]Section of Sensory Sci. and Metabolism, [2]Lab. of Neuroimaging, Natl. Inst. on Alcohol Abuse and Alcoholism/National Inst. of Hlth., Bethesda, MD; [3]Natl. Inst. of Mental Health/National Inst. of Hlth., Bethesda, MD; [4]Natl. Inst. of Diabetes and Digestive and Kidney Diseases/National Inst. of Hlth., Bethesda, MD; [5]NIDA/NIH, NIH, Natl. Inst. On Drug Abuse, Bethesda, MD

Abstract: ***Background:*** Adverse effects of prenatal caffeine exposure (PCE) on children include maladaptive behavior, cognition, developmental delays, abnormal neuro-motor activity, excess body weight, heightened risk for obesity. Since PCE may cause obesity, it may also possibly increase a child's preference for sugar rich foods, which can develop early in childhood. Here we sought to investigate the relationship of PCE with total sugar intake (TSI) and brain activity during reward anticipation. Also, if TSI serves as potential mediator responsible for the indirect effect on relationship between PCE and altered reward sensitivity during reward anticipation in adolescents using the large Adolescent Brain Cognitive Development (ABCD) dataset. ***Methods:*** The analysis was conducted on n=5534 adolescents (9-11 years), excluding children with mothers known to consume alcohol or any other illicit drugs, with no TSI data, as well as those with no data on caffeine use during pregnancy (n=6344 excluded). Separate regression models were built to establish the relationships between (i) PCE, TSI and insular thickness; (ii) PCE and different regions of interest (ROIs) functional brain activity on reward anticipation during the monetary incentive delay (MID) task. We used the categorical variable for PCE (No/daily/weekly/less than weekly exposure), TSI (g) from the Block Kids Food Screener, and cortical insular thickness (bilateral total). ROIs for exploring activation to reward anticipation MID task-functional MRI were middle frontal cortex (MFC), anterior cingulate, insula, orbitofrontal cortex, and nucleus accumbens. Mediation analysis was conducted in [SPSS 28.0] using the Baron and Kenny method. For pairwise comparisons Bonferroni corrections were applied. ***Results and Conclusion:*** As hypothesized, we observed a significant relationship of PCE with TSI ($F_{3,5319} = 3.3$; $p = 0.02$) and rostral MFC activation during reward anticipation ($F_{3,3951} = 2.7$; $p = 0.04$). Children with daily PCE compared to counterparts with exposure occurring at frequencies of less than a week displayed greater TSI (beta = 3.6; $p = 0.01$), but lower MFC activation during reward anticipation ($b = -0.04$; $p = 0.01$). Significant indirect effect

attributed to TSI was associated with the relationship between PCE and rostral MFC activation during reward anticipation ($F_{1,3950} = 5.3$; $p = 0.02$). Interestingly, a relationship between lower insular thickness and elevated PCE exposure and TSI of female compared to male adolescents ($F_{1,5473} = 13.5$; $p < 0.001$; $F_{1,5319} = 92.4$; $p < 0.001$) was seen providing evidence for potential altered taste perception in children pre-exposed to high maternal caffeine doses.

Disclosures: K. Agarwal: None. **P. Manza:** None. **H.A. Tejeda:** None. **A.B. Courville:** None. **N.D. Volkow:** None. **P.V. Joseph:** None.

Poster

542. Taste

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 542.15

Topic: D.04. The Chemical Senses

Support: NIH grant UL1GM118979, TL4GM118980, and RL5GM11897
Psi Chi Faculty Advisor Research Grant
CSULB Student Summer Research Award
CSULB RSCA small faculty grant

Title: Cross-generalization profile to sucrose, quinine and sucrose-quinine mixtures in female and male rats conditioned to avoid 5% and 10% ethanol

Authors: M. A. BARCELOS, T. J. LOPEZ, T. HO, S. R. HESSEL, *Y. TREESUKOSOL; Dept. of Psychology, California State Univ. Long Beach, Long Beach, CA

Abstract: Individual variability in taste responsivity contributes to differences in dietary choices. Like humans, adult female rats display a higher propensity than males to consume ethanol under some conditions. Though bitter-tasting compounds like ethanol are innately avoided by humans and rodents, variability in the taste qualities of ethanol may attribute to variability in intake. Previous findings in the literature suggest conditioned avoidance to alcohol in rats generalizes to compounds that humans describe as bitter and sweet. The current study was designed to test the hypothesis that taste qualities of ethanol differ in female and male rats. Here, female (n=40) and male (n=37) rats were presented either 5% or 10% ethanol followed by administration of 0.15 M LiCl (1.33 ml/100 g body weight; unconditioned stimulus; US) to induce visceral malaise, or saline as a control. Both female and male rats administered LiCl displayed avoidance of ethanol across the 4 conditioning trials. Generalization of the conditioned avoidance was assessed in a brief-access taste test (10-s trials; 30 min session). The test array included water, 0.03 M sucrose, 0.3 M sucrose (representing “sweet” compounds), 0.03 mM quinine, 0.3 mM quinine, (representing “bitter” compounds) and mixtures 0.3 M sucrose - 0.03 mM quinine, and 0.03 M sucrose - 0.3 mM quinine, presented in randomized blocks without replacement. Animals could initiate as many trials as possible during the 30-min test session. The average number of licks to each test stimulus was used to calculate suppression scores, indicating the degree to which rats

generalized the conditioned avoidance of EtOH to each test stimulus. Conditioned avoidance of 5% and 10% ethanol generalized to both concentrations of sucrose and sucrose-quinine mixtures but only to the higher concentration of quinine indicating the sucrose-like component is the more salient orosensory feature of ethanol in rats. Both female and male LiCl-injected rats showed higher suppression scores to sucrose than quinine suggesting sex differences in ethanol ingestion is not primarily driven by differences in qualitative profiles. It is possible that differences in ethanol responses can be attributed by other aspects of taste function, such as reward signaling.

Disclosures: M.A. Barcelos: None. **T.J. Lopez:** None. **T. Ho:** None. **S.R. Hessel:** None. **Y. Treesukosol:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.01

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant R01NS128904
NIH Grant R01AG077681

Title: Mice Demonstrate Asymmetric Context-Dependent Shifts to Phonemes

Authors: *S. MEHAN[1], M. S. WEHR[2];
[1]Univ. of Oregon, Univ. of Oregon Inst. of Neurosci., Eugene, OR; [2]Univ. of Oregon Inst. of Neurosci., Inst. of Neurosci., Eugene, OR

Abstract: Humans are capable of processing phonemes in real time with high accuracy despite widely variable auditory information from different speakers and sources, and we have yet to determine how this is achieved. Recent work from the Wehr Lab has established that mice are able to learn and discriminate between phonetic categories. Humans have demonstrated a context-dependent shift in speech perception, and we investigated whether this effect is present in mice neurometric data. We chronically implanted 12 mice with 32- or 64-channel tetrode arrays in Auditory Cortex (AC) and recorded over 500 neurons while freely moving and listening to synthetically generated phonemes without discrimination training. We found a wide variety of neurons demonstrating speech context effects to preceding speech stimuli, and that mice demonstrate neurometric context effects of preceding speech sounds, but that this shift is asymmetric and not dependent on the identity of the preceding context. This project lays the groundwork for future experiments to elucidate the mechanisms of phoneme non-invariance.

Disclosures: S. Mehan: None. **M.S. Wehr:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.02

Topic: D.05. Auditory & Vestibular Systems

Support: Texas Speech Language Hearing Foundation

Title: How much data are needed to build encoding models for natural speech EEG experiments?

Authors: *M. DESAI[1], L. S. HAMILTON[2];
[1]The Univ. of Texas at Austin, Univ. of Texas, Austin, Austin, TX; [2]The Univ. of Texas at Austin, Austin, TX

Abstract: In many electroencephalography (EEG) studies which investigate auditory and speech processing in the brain, the experiments are often lengthy and tedious. Researchers tend to collect more trials and ultimately have a longer task to ensure that the acquired data are robust and effects are measurable. Recent studies have used naturalistic stimuli to investigate the brain's response to individual or a combination of multiple speech features using multivariate temporal receptive field (mTRF) analyses. In this analysis, the neural data collected from an experiment must be divided into a training set and a test set to fit and validate the mTRF weights. While a good strategy is clearly to collect as much data as possible, it is unclear how much data are needed to achieve stable results. Furthermore, it is unclear whether the specific stimulus used for mTRF fitting and the choice of feature representation affects how much data would be required for a robust and generalizable result. We used two contrasting stimuli from a previously published EEG dataset. We tested models predicting brain activity from different speech features to better understand how much data needs to be collected for naturalistic speech experiments. Our results suggest that we need far fewer trials than the original amount of data collected during the EEG session. Using sentences from the TIMIT corpus, we found that the EEG receptive field structure stabilizes after collecting a training dataset of approximately 100 sentences (about 200 seconds). On the other hand, using audiovisual movie trailers requires more training data, but is still reasonably small (around 600 seconds). From our findings, we seek to provide suggestions on the minimum amount of data that would be necessary for building receptive field models from naturalistic listening data. This is a highly practical concern for working with children, patient populations, or others who may be unable to tolerate long study sessions. Ultimately, the findings from this study will hopefully provide suggestions to future researchers who may want to build a task to answer questions about naturalistic speech processing in healthy and clinical populations while minimizing participant fatigue, yet retain clean signal quality.

Disclosures: M. Desai: None. **L.S. Hamilton:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.03

Topic: D.05. Auditory & Vestibular Systems

Support: CIHR- MOP133450
Canada Research Chair program
the Natural Sciences and Engineering Council of Canada CGS-D
Vector Institute Post Graduate Affiliates Program

Title: The Effects of Speech Masking on Neural Tracking of Acoustic and Semantic Features of Naturalistic Speech

Authors: *S. YASMIN[1], V. IRSIK[2], I. S. JOHNSRUDE[2], B. HERRMANN[3];
[1]Western Univ., , London, ON, Canada; [2]Western Univ., London, ON, Canada; [3]Rotman Res. Inst., Rotman Res. Inst., Toronto, ON, Canada

Abstract: Natural listening environments generally contain background masking sounds, which can degrade speech signals and hinder communication. Speech understanding depends on both acoustic and lexical semantic information in a speech stream: the acoustic information cues the linguistic identity, and lexical semantics enables reconstruction of missed content when speech is degraded. Most research into how acoustic and semantic features influence the neural representation of speech, and its intelligibility, has been conducted using very simple, non-naturalistic listening situations; for example, brief, isolated sentences presented in a repetitive event-related design. Our group has recently shown that listening to engaging naturalistic speech, compared to disconnected sentence-length utterances, qualitatively alters listening behaviour and can facilitate intelligibility of masked speech (Irsik, Herrmann, Johnsrude, 2022). In the current study, we investigate how the neural tracking of the amplitude envelope and the semantic information in engaging, continuous speech changes with level of multi-talker background masking (babble), and how changes in neural tracking relate to changes in speech intelligibility. We tested noise levels from clear to -3 dB SNR. Neural tracking is calculated using a linear mapping between features of the continuous speech stimulus and the corresponding electroencephalographic signals. Neural tracking of the amplitude envelope of speech was larger for speech masked by moderate babble noise than for both clear speech and highly masked speech although background masking generally increased tracking of the amplitude envelope. Tracking of semantics was less affected by noise and remained robust until speech masking was severe (-3dB SNR; intelligibility ~60%; Irsik et al 2022). Our findings show that neural tracking of semantics is more robust to noise than is neural tracking of acoustics, presumably because cognitive processes help to recover and repair the masked signal.

Disclosures: S. Yasmin: None. **V. Irsik:** None. **I.S. Johnsrude:** None. **B. Herrmann:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.04

Topic: D.05. Auditory & Vestibular Systems

Support: Ministry of Education Youth and Sports, Czech Republic, Grant LTAIN 19201

Title: Neuronal responses to tonal stimuli and ultrasonic vocalizations in the inferior colliculus of CBA mice

Authors: K. PYSANENKO, ***D. SUTA**, J. SYKA;
Inst. of Exptl. Med. CAS, Prague, Czech Republic

Abstract: It is very common for mice to emit communication sounds during the social interaction. These species-specific vocalizations are found predominantly in the ultrasonic range with frequencies typically above 50 kHz. In our work, we studied how ultrasonic vocalizations are encoded in the subcortical portion of the central auditory system in CBA mice. Specifically, we focused on the inferior colliculus (IC) of the mouse, the high-frequency region of which is known to contain relatively small number of neurons with characteristic frequencies matching the frequency content of ultrasonic vocalizations. We recorded ultrasonic vocalizations in freely moving mice (1.5-18 months of age) when one female and one male were temporarily housed together in the same cage. The repertoire of communication sounds of the inbred CBA strain was dominated by relatively simple vocalizations consisting of a series of short bouts with upward and downward frequency modulation. The responses of IC neurons to tones, combinations of two tones, frequency-modulated tones and ultrasonic vocalizations were recorded using a multichannel microelectrode in ketamine-xylazine anesthetized mice. Two-tone stimulation was applied to revealed inhibitory regions in the frequency response area. We found not only lower and/or upper inhibitory sidebands surrounding excitatory region at the characteristic frequency but specifically in high-frequency neurons inhibition formed more complex patterns. Neurons in the high-frequency region of the IC frequently displayed responses with several excitatory peaks when stimulated with upward or downward frequency modulated tones, whereas those in the low-frequency region responded predominantly with a continuous excitatory reaction. In principle, only neurons localized in the high-frequency region of the IC responded to species-specific vocalizations and their responses did not follow all segments of the vocalization bouts. This finding suggests that the inhibition might play an important role in the processing of species-specific vocalizations in the subcortical part of the auditory pathway in CBA mice.

Disclosures: K. Pysanenko: None. **D. Suta:** None. **J. Syka:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.05

Topic: D.05. Auditory & Vestibular Systems

Support: Swiss National Science Foundation

Title: The Neural Basis of Spectro-temporal Glimpsing Voice-in-Noise

Authors: *H. SWANBOROUGH, S. FRÜHHOLZ;
Univ. of Zurich, Zürich, Switzerland

Abstract: Successfully perceiving and processing speech-in-noise is fundamental to our everyday listening and communication. A major problem for the auditory system is that the dynamic, changing noise of everyday life unsystematically masks speech. Just as the visual system can identify an object that is partially obscured in the visual field, spectrotemporal 'glimpsing' is the process by which a target voice can be processed despite only some spectrotemporal information being available to the listening brain. While glimpsing is well established within psychoacoustics research, the neural structures or networks that may facilitate the process have not yet been investigated. This project aims to address the gap in the literature, investigating the neural substrates supporting glimpsing of target voices during effortful voice-in-noise listening. In a behavioural-fMRI project, we presented participants (N=26) with affective, non-verbal vocalisations obscured by 'bubbled' noise. Each bubble suppressed a spectrotemporal region of noise to zero energy, ensuring stimuli presented concurrently with noise would be unobscured in those regions. Remaining stimuli spectra were fully obscured at –23dB. Non-verbal stimuli were chosen to minimise the potential non-spectrotemporal information that could be used for signal reconstruction by the auditory system.Classification images derived from the behavioural results showed statistically significant spectrotemporal regions for each stimuli that were beneficial for perception. This finding indicates consistent spectral cue availability across participants despite the challenging nature of the task. Imaging contrasts of correct-vs-incorrect stimuli categorisation saw recruitment of a left-lateralised network involving the putamen and superior temporal gyrus. These results echo existing work showing a left-lateralised, putamen supported network during high effort listening tasks that require greater internal representation of unanalysed speech; tasks requiring reflexive auditory learning; and error processing. We propose that these results show the putamen plays a key supporting role to the auditory system for processing perceptually incomplete voice stimuli, via this left lateralised network. Crucially, we demonstrate that this process exists even when the auditory system only has access to fundamental, non-linguistic information from the target voice. To our knowledge, this is the first study to explicitly attempt to map SIN glimpsing onto the human brain and is an important step to fully understanding how the auditory system successfully processes SIN in dynamic, challenging environments.

Disclosures: H. Swanborough: A. Employment/Salary (full or part-time):; University of Zürich. **S. Frühholz:** A. Employment/Salary (full or part-time):; University of Zürich, University of Oslo.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.06

Topic: D.05. Auditory & Vestibular Systems

Support: Facebook/Meta

Title: Brain representations of distracting sounds during continuous story listening

Authors: L. XU, A. FIELD, S. HASHEMGELOOGERDI, M. DESAI, L. S. HAMILTON, A. G. HUTH;
The Univ. of Texas at Austin, Austin, TX

Abstract: In social conversation, people can be distracted by external sounds while attending to speech. We investigated how the distracting sounds are represented in the brain, and how the information from attended speech is represented differently from distracting sounds using encoding and decoding models with fMRI. We collected fMRI data from 3 participants (2M/1NB, age 22±2.6) while they listened to stories taken from The Moth Radio Hour and other podcasts (the "main stream") with occasional embedded distracting sounds. Distractor sounds included a set of short natural sounds encompassing both speech and non-speech stimuli (e.g. music, environmental sounds), as well as longer distractors taken from other stories and movie trailers. To simulate real world scenarios, we applied a head-related transfer function to simulate the distracting sounds appearing at -45°, 0°, and 45° azimuth relative to the front of the person. Participants were asked to attend to the main stream stimulus and were occasionally probed to respond with what words they heard during a distractor stimulus. We then fit linear regression models to predict fMRI responses using acoustic and linguistic features of the main stream and distractors. Variance partitioning showed that main stream sounds were best encoded in primary auditory, lateral superior temporal gyrus, and inferior frontal speech areas, while distractor sounds were mostly encoded in posterior auditory cortex, and the spatial locations of distractors were encoded contralaterally across left and right hemisphere. Logistic regression models were used to decode distractor onset time, duration, and spatial location, all of which could be decoded with high accuracy (AUC > 0.9 for onset time and duration, AUC > 0.85 for spatial location). Voxels in auditory cortex were the most important for decoding. In conclusion, mainstream and distractors sounds are represented in anatomically distinct brain areas and distractor onset, duration, and spatial location features are all decodable from fMRI responses.

Disclosures: L. Xu: None. **A. Field:** None. **S. Hashemgeloogerdi:** None. **M. Desai:** None. **L.S. Hamilton:** None. **A.G. Huth:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.07

Topic: D.05. Auditory & Vestibular Systems

Support: R34NS118462

Title: Dissociation of tonotopy and ethologically-relevant categories in the auditory midbrain of the echolocating bat

Authors: J. LAWLOR BLONDEL[1], M. J. WOHLGEMUTH[4], P. GUTRUF[5], C. F. MOSS[2], ***K. V. KUCHIBHOTLA**[3];
[1]Psychological and Brain Sci., [2]Dept. of Psychological and Brain Sci., [3]Psychological and Brain Sciences, Neurosci. and Biomed. Engin., Johns Hopkins Univ., Baltimore, MD; [4]Neurosci., Univ. of Arizona, Tucson, AZ; [5]Univ. of Arizona, Tuscon, AZ

Abstract: Navigating our everyday world requires parsing stimulus information from constantly evolving sensory flows. The physical properties of stimuli may vary continuously, while behavioral responses are comparatively discrete. For example humans can readily understand spoken words despite speech spectral properties varying across speakers. How do category-specific representations emerge in the brain? Here, we take advantage of an animal model long-studied for its expert auditory sensing: the echolocating bat. The bat auditory system makes use of echoes from ultrasonic vocalizations to determine the identity and location of objects - and for social interactions with conspecifics. As such, the bat constructs its representation of the external environment using sound, making it a powerful model to investigate the brain's representation of natural acoustic categories. We developed two-photon calcium imaging in the awake big brown bat, Eptesicus fuscus, to assay the activity of a population of neurons with cellular resolution. We expressed GCaMP6f in excitatory neurons of the Inferior Colliculus, a central auditory hub, while using a thinned-skull approach to monitor large populations of cells in head-fixed subjects. We assessed functional auditory properties of thousands of neurons in awake, passively listening bats (n=3 bats) by presenting a large stimulus set, including pure tones of varying duration and playbacks of natural calls. We discovered a superficial fine-scaled tonopy in the superficial layers of the IC shell region with cells' preferred frequencies increasing in both the caudolateral and rostromedial extent. Using two sets of natural call categories-exemplar ultrasonic social vocalizations and temporally matched echolocation calls-we show that the sampled population is category selective. We found that category-specific cells do not follow the tonotopic gradient but rather form small clusters spread across the IC. Large-scale population decoding reveals sharper boundaries across, rather than within, sound category even when stimuli show considerable spectrotemporal variation. Our two-photon calcium imaging in the echolocating bat reveals the relationship between traditionally defined functional auditory features and natural categories of sounds in large neural ensembles with unprecedented spatial fidelity.

Disclosures: J. Lawlor Blondel: None. **M.J. Wohlgemuth:** None. **P. Gutruf:** None. **C.F. Moss:** None. **K.V. Kuchibhotla:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.08

Topic: D.05. Auditory & Vestibular Systems

Support: NIH F32 DC018721-01A1
NIH R01 DC013826-06A1

Title: Acoustic, orofacial, and neural correlates of vocalization in the mouse auditory cortex

Authors: *T. HARMON[1], S. MADLON-KAY[2], A. KUMAR[3], J. M. PEARSON[2], R. D. MOONEY[4];
[1]Neurobio., [2]Biostatistics and Bioinformatics, [3]Physics, [4]Duke Univ., Durham, NC

Abstract: Auditory cortical neurons integrate auditory and non-auditory information, including motor-related signals, arousal-related neuromodulatory signals, and, at least in mice, olfactory signals from other conspecifics. These non-auditory signals are likely to be especially important for how the auditory cortex processes auditory feedback during vocalization, a complex behavior which involves both vocal and non-vocal movements, heightened arousal, and that in male mice is triggered by female odorants. To characterize how auditory and non-auditory signals modulate auditory cortical activity, we developed a paradigm for studying vocal and non-vocal courtship interactions between female and head-fixed male mice. We found that episodes of social interaction correlated with increased arousal, decreased locomotion, and increased likelihood that the male would produce ultrasonic vocalizations (USVs). We found that male USV production correlated with increased orofacial movements, but less reliably with changes in other behavioral parameters, and was strongly dependent on olfactory cues from the female. We used 2p calcium imaging in the head-fixed male to monitor the activity of auditory cortical neurons during USV production and in response to playback of the same USVs when the male was quietly listening. We found a subset of neurons that responded strongly to USV playback stimuli but only weakly during USV production, consistent with the idea that auditory cortical responses are suppressed during vocalization. We are currently analyzing these combined behavioral and neural datasets to identify which non-auditory signals contribute to auditory cortical suppression during USV production.

Disclosures: T. Harmon: None. **S. Madlon-Kay:** None. **A. Kumar:** None. **J.M. Pearson:** None. **R.D. Mooney:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.09

Topic: D.05. Auditory & Vestibular Systems

Support: NIH F32DC018508
NIH R01NS082179

Title: Characterizing a Circuit Linking Auditory Pallium and the Social Behavior Network

Authors: *J. A. SPOOL[1], A. LALLY[2], P. CHEN[1], L. REMAGE-HEALEY[1];
[1]Univ. of Massachusetts, Amherst, Amherst, MA; [2]Wheaton Col., Norton, MA

Abstract: To engage in healthy social interactions, the brain must coordinate processing of social sensory cues (i.e., visual, auditory) with appropriate social responses. While complex features of social signals are processed in the telencephalic pallium, nuclei controlling social behaviors, called the social behavior network (SBN; conserved across vertebrates), reside mainly in the diencephalon. In songbirds, for example, the ability to learn dozens of individuals by their vocalizations depends on auditory pallium, while the SBN are necessary for appropriate social responses to songs and calls. Tremendous progress has been made in studying pallial sensory circuits and the SBN largely in parallel, but apart from mammalian olfactory systems we have little knowledge about their intersection. We asked whether auditory pallial circuits contribute to the SBN responses to social sensory cues. We transiently inactivated auditory pallium of female Zebra finches with inhibitory neurotransmitter receptor agonists during song playback, and examined song-induced immediate early gene (egr-1) activation in SBN nuclei. Auditory pallial inactivation specifically impaired egr-1 responses to song in the lateral ventromedial nucleus of the hypothalamus (VMHl), providing the first evidence in vertebrates of a connection between auditory pallium and the SBN. An ANCOVA additionally revealed a relationship between egr-1 expression in VMHl and feeding behavior. Feeding behavior correlated with VMHl egr-1 in 3 out of 4 experimental treatments: control birds exposed to silence, and birds with inactivated auditory pallium exposed to either song or silence; this is consistent with the dual roles of VMH in homeostatic regulation and social behavior. However, this correlation specifically did not include control animals exposed to auditory playback, indicating that auditory input from pallium to VMHl may mediate a trade-off between social attention and feeding. Electrophysiological recordings from VMHl in both female and male Zebra finches reveal a large proportion of single units that are highly responsive to ecologically-relevant bird songs and calls. Female single units had stronger responses to auditory stimuli compared to males, and male single units were selective for contact calls over playback of male songs. These data highlight a role for an auditory pallium to VMHl circuit in the integration of social auditory stimuli with internal state to influence social decision-making.

Disclosures: **J.A. Spool:** None. **A. Lally:** None. **P. Chen:** None. **L. Remage-Healey:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.10

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant DC014299
NIH Grant DC018525

Title: Evidence for a vocal error signal in marmoset auditory cortex

Authors: *S. J. ELIADES[1], J. TSUNADA[2];
[1]Head and Neck Surgery & Communication Sci., Duke Univ. Sch. of Med., Durham, NC;
[2]Chinese Inst. for Brain Res., Beijing, China

Abstract: During both human speech and non-human primate vocalization, there is a well described suppression of activity in the auditory cortex. Despite this suppression, the auditory cortex remains sensitive to perturbations in sensory feedback, and this sensitivity has been shown to be important in feedback-dependent vocal control. Although the mechanisms of suppression and vocal feedback encoding are unclear, this process has been suggested to represent an error signal encoding the difference between sensory-motor prediction and feedback inputs. However, direct evidence for the existence of such an error signal is lacking. In this study, we investigated the responses of auditory cortical neurons in marmoset monkeys during vocal production, testing frequency-shifted feedback of varying magnitude and direction. Consistent with an error signal hypothesis, we found that population-level neural activity increased with the magnitude of feedback shifts, but were symmetric between positive and negative frequency changes. This feedback sensitivity was strongest in vocally-suppressed units and for units whose frequency tuning overlapped that of vocal acoustics. Individual units tested with multiple feedback shifts often showed preferences for either positive or negative feedback shifts, with only a minority showing sensitivity to feedback shifts in both directions. Frequency tuning distributions were different for units showing preference for one feedback direction over the other. These results suggest that vocal feedback sensitivity in the auditory cortex is consistent with a vocal error signal, seen at both the individual unit and population level.

Disclosures: S.J. Eliades: None. **J. Tsunada:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.11

Topic: D.05. Auditory & Vestibular Systems

Support: NIDCD R01 DC009810
NSF/IOS 1656825

Title: Auditory cortical coding of species-specific acoustics and temporal organization in songbird vocal communication

Authors: *J. A. EDWARDS[1], S. M. N. WOOLLEY[1,2];
[1]Psychology, [2]Zuckerman Inst., Columbia Univ., New York, NY

Abstract: Hearing and speech are foundational to human social communication. Early auditory experience with native language guides vocal learning through the landscape of development and

shapes auditory cortical coding for life. Like humans and unlike other animals, songbirds learn to sing by copying the vocal sounds of adult tutors. As in humans, early vocal learning permanently shapes neural coding and perception of acoustic features. Prior work shows that juvenile songbirds that are reared and tutored by parents of a different species can successfully copy heterospecific song syllables, which are spectrotemporally complex. In contrast to the demonstrated flexibility in syllable learning, cross-species tutoring experiments suggest that the temporal organization of syllables into sequences is largely determined by species genetics. Juveniles copy the syllables of their adoptive tutor's songs, but produce those syllables with the timing and temporal order typical of their own species, even when they have never heard conspecific song. We hypothesize that the secondary auditory cortex (caudal nidopallium; NC), where neurons are tuned by vocal learning, contains two populations of neurons: one that encodes syllable acoustics, and one that encodes temporal patterning. To test this hypothesis, we studied song behavior and auditory cortical coding of four species of songbirds that have known relatedness and specific differences in song acoustics and temporal organization. Using single-unit electrophysiology, we compared the responses of NC neurons to stimuli including the natural songs of multiple species, songs with altered temporal structure, and two classes of synthetic sounds that systematically varied in spectrotemporal acoustics and temporal patterning. Across species, NC neurons responded with highest spike rates toward songs of their own species. In species that produced songs with complex acoustics, tuning to the spectrotemporal modulations found in conspecific song explained firing rate differences to species' songs. In species that produce simple songs, tuning to acoustic frequency explained neuronal responses to songs. Analysis of single NC neurons indicated that neurons were sensitive to either acoustics or timing, but rarely both. A subset of neurons was sensitive to the position of a syllable within song, suggesting sensitivity to syllable order. Results indicate that separate subpopulations of secondary auditory cortical neurons process spectrotemporal acoustics and sequence organization of vocal communication sounds, which could be controlled by distinct cellular mechanisms to shape the vocal learning landscape.

Disclosures: **J.A. Edwards:** None. **S.M.N. Woolley:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.12

Topic: D.05. Auditory & Vestibular Systems

Support: (Schweizerischer Nationalfonds zur Förderung der Wissenschaftlichen Forschung, SNSF 100014_182135/1

Title: Real-time modulation of the amygdalo-hippocampal network by live affective speech

Authors: *F. STEINER[1,2], C. TREVOR[1], S. FRUEHHOLZ[1,2,3];
[1]Cognitive and Affective Neurosci. Unit, Univ. of Zurich, Zurich, Switzerland; [2]Neurosci. Ctr.

Zurich, Univ. of Zurich and ETH Zurich, Zurich, Switzerland; [3]Dept. of Psychology, Univ. of Oslo, Oslo, Norway

Abstract: Affective speech communication is an essential part of human social interaction. Speakers encode information about their emotional state in specific acoustic voice features that listeners then process using their cortico-limbic vocal affect processing system. Since speakers and listeners continuously react and adapt to each other, real-life affective speech communication is dynamic. However, previous research has identified the affective voice processing network mainly with pre-recorded and thus non-social and non-adaptive voices, thereby underrepresenting the socio-dyadic and dynamic nature of such communication. To address this gap, we have developed an innovative real-time fMRI setup that factors these social and adaptive dynamics into the experiment by creating a closed-loop situation between speakers' live affective speech and listeners' live limbic connectivity. As a proxy for listeners' affect recognition, we extracted a connectivity metric between the amygdala (AMY) and the hippocampus (HC) in the left hemisphere, which served as feedback to the speakers. Speakers (n = 10, 5 female) dynamically produced and continuously modulated their voices in direct response to the live limbic connectivity feedback of listeners (n = 26, 10 female, age: M = 28.7 y, SD = 6.5) to induce an increase in listeners' AMY-HC connectivity. We ran two control conditions to differentiate activity due to dynamic and live-adaptive speech and our feedback target. First, speakers received static feedback, and second, feedback from right hemispheric AMY-HC connectivity. Our results show that affective speech produced in response to listeners' left AMY-HC connectivity directly increased the functional connectivity and activated the cortico-limbic vocal affect processing system of listeners more than non-adaptive speech (static feedback). Additionally, in response to the live-adaptive voices, we found more inter-connectivity between the different areas of the vocal affect processing system. An acoustic analysis of the speakers' voices indicated a higher variability among central affective voice features in live-adaptive versus non-adaptive voices signifying their dynamic vocal adaption to listeners' AMY-HC connectivity. These results suggest that interactive and individually adaptive affective communication intensifies the activation of the intralimbic vocal affect processing system. Therefore, our experimental setup is a substantial effort to better capture the full processing capacities of the neural system involved in dynamic real-life affective communication and emotional responding.

Disclosures: F. Steiner: None. **C. Trevor:** None. **S. Fruehholz:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.13

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant F32MH123016

Title: Neural circuitry and plasticity underlying top-down control of vocalization-guided maternal behavior

Authors: *A. M. LEMESSURIER[1], R. C. FROEMKE[2];
[1]Neurosci. Inst., NYU Sch. of Med., New York, NY; [2]Otolaryngology, NYU Med., New York, NY

Abstract: The ability of mothers to detect and respond to sensory cues from infants is essential for survival in mammals. A key maternal behavior in mice is retrieving isolated pups into the nest in response to infant ultrasonic vocalizations (USVs). Retrieval is performed robustly by experienced mothers (dams), but can be learned by virgins co-housed with a dam and litter. Maternal experience induces plasticity in left auditory cortex that broadens tuning curves for USVs and increases reliability of responses, and activity left auditory cortex is required for retrieval (Carcea et al., Schiavo et al. 2020, Marlin et al. 2015). How does this enhanced coding support pup retrieval? Descending projections from auditory cortex to subcortical targets may be crucial for retrieval. We used 2-photon *in vivo* calcium imaging and *in vivo* channelrhodopsin-assisted patching to examine encoding of USVs throughout maternal experience in auditory projection neurons, labeled via retrograde viral tracing. We measured responses to USVs in neurons projecting to either the inferior colliculus or posterior striatum. In corticocollicular neurons, imaging experiments revealed a striking increase in baseline activity during epochs of repeated USV presentations compared to tone presentation epochs. In contrast, baseline activity in corticostriatal neurons was equivalent during tone and USV presentation epochs. Time-locked responses to USVs were also substantially larger in corticocollicular compared to corticostriatal neurons (evoked dF/F: 5.6 +/- 0.55, N=158 corticocollicular neurons from 4 mice; 2.9 +/- 0.2%, N=271 corticostriatal neurons from 3 mice; $p<0.001$). *In vivo* patch measurements from optogenetically-identified neurons reflect this trend (evoked firing rate: 2.04 Hz +/- 0.72, N=8 corticocollicular neurons from 3 mice; 0.24 Hz +/- 0.44 N=5 corticostriatal neurons from 5 mice, $p=0.12$). We tested the involvement of select populations of neurons in pup retrieval using chemogenetics. Suppressing activity in left auditory cortex layer 5 neurons impaired retrieval performance in retrieving females (vehicle: 90 +/- 10% of pups retrieved; CNO: 41 +/- 7%, N=3 mice, 10 trials per condition). However, bilateral suppression of activity in corticostriatal neurons did not impair performance (vehicle: 87 +/- 5% of pups retrieved; CNO: 96 +/- 2%, N=3 mice, 10 trials per condition). Together, these results suggest that excitability is increased in corticocollicular neurons in retrieving females during prolonged exposure to USVs.

Disclosures: A.M. LeMessurier: None. **R.C. Froemke:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.14

Topic: D.05. Auditory & Vestibular Systems

Title: Differences between generic and individualized TRF-models for tracking the cortical processing of speech

Authors: *O. BIALAS, E. LALOR, A. NIDIFFER;
Biomed. Engin., Univ. of Rochester, Rochester, NY

Abstract: Over the past decade, applying temporal response functions (TRFs) to EEG recordings provided novel insights into the brain's processing of naturalistic speech. TRFs can be used as forward models, using stimulus features to predict brain responses or as backward models, using brain responses to reconstruct stimulus features. Forward models have been used to show that EEG reflects the phonetic processing and semantic understanding of speech, while backward models have been used to measure a listener's speech comprehension or determine selective attention in a multi-speaker scenario. Thus, the TRF-technique could be used to study speech perception in populations where psychophysical tests are difficult to administer such as children or neurologically impaired patients. However, fitting a TRF-model requires a minimum amount of data which might be hard to acquire in those populations. This can be remedied by averaging the TRF-coefficients across multiple subjects to compute a generalized model which can then be used to analyze limited amounts of data in a test subject. However, since they fail to capture individual differences in the stimulus-to-response mapping, generalized TRFs are outperformed by individualized models given sufficient subject-specific training data. Generalized models might also fail on subjects who's brain-to-stimulus mapping deviates strongly from the group. Thus, while for most subjects, the performance of both models increases monotonically with training time, there are cases where an individualized model performs well while generalized models fail. Here, we investigate how the amount of training data and choice of features impact performance for generalized and individualized TRF-models across multiple datasets. We do this for both backward models based on the speech envelope, as well as forward models based on acoustic, phonetic, and semantic features. We test model performance across multiple frequency bands to investigate whether cases where the performances of individualized and generic models deviate are explained by their reliance on different neural features. Our results can be used to inform the design and modeling strategy of studies that wish to use TRFs to investigate the perception of speech in clinical populations.

Disclosures: **O. Bialas:** None. **E. Lalor:** None. **A. Nidiffer:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.15

Topic: D.05. Auditory & Vestibular Systems

Support: National Science Foundation NSF grant 1355065
National Institute of Health NIH DC015138 01 (HLR, Co-I)
University of Connecticut 12 Brain-Computer Interface Core.

Title: Sound duration perception predicted by Bayesian decision-theoretic model incorporating the empirical co-variations of onset slope and durations found in natural vocalizations.

Authors: M. JANE[1], S. PISUPATI[2], *J. CHROBAK[1], **H. READ**[1];
[1]Psychological Sci., Univ. of Connecticut, Storrs, CT; [2]Princeton Neurosci. Inst., Princeton Univ., Lawrenceville, NJ

Abstract: It is well known that animals rely on multiple sources of information in order to successfully identify sounds in natural environments, to make decisions that are optimal for their survival. For example, rats use duration and pitch cues to respond appropriately to prosocial and distress vocalizations (Saito et al., 2019). Vocalization duration cues are known to co-vary with other temporal cues (Khatami et al., 2018), yet little is known about whether animals rely upon such co-variations to successfully discriminate sounds. In the current study, we find natural alarm vocalizations in rats have onset and offset slopes that are correlated with their duration. Accordingly, vocalizations with faster onset slopes are more likely to have shorter dura- tions. Given that vocalization slopes begin and end within milliseconds, they could provide rapid perceptual cues for predicting and discriminating vocalization duration. To examine this possibility, we train rodents to discriminate duration differences in sequences of synthetic vocalizations and examine how artificially changing the slope impacts duration judgments. We find animals are biased to misjudge a range of synthetic vocalizations as being shorter in duration when the onset and offset slopes are artificially fast. Moreover, this bias is reduced when rats are exposed to multiple synthetic vocalization bursts. The observed perceptual bias is accurately captured by a Bayesian decision-theoretic model that utilizes the empirical joint distribution of duration and onset slopes in natural vocalizations as a prior during duration judgments of synthetic vocalizations. This model also explains why the bias is reduced when more evidence is accumulated across multiple bursts, reducing the prior's influence. These results support the theory that animals perceive fine-grained statistical co-variations in auditory timing cues and integrate this information optimally with incoming sensory evidence to guide their decisions.

Disclosures: **M. Jane:** None. **S. Pisupati:** None. **J. Chrobak:** None. **H. Read:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Elemind Technologies, Inc..

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.16

Topic: D.05. Auditory & Vestibular Systems

Support: Belgian Kids Fund
FNRS Grant F.4508.22

Title: Development of speech perception in noise: neural correlates of stream segregation in children and adolescents

Authors: *J. F. JOHNSON, E. BENOCCI, M. COUVIGNOU, L. LEONARDY, A. CALCUS; Univ. Libre De Bruxelles, Brussels, Belgium

Abstract: In noisy backgrounds, listeners perform auditory scene analysis: the parsing of different auditory streams ("stream segregation"), and selective focus on one particular stream as it unfolds over time ("selective attention"). This project is part of a larger research program aimed at evaluating the neural mechanisms of both stream segregation and selective attention. Here, we present a replication study aimed at uncovering the neural signature of auditory stream segregation in healthy adults. Nine normally-hearing adults were presented with sequences of sounds consisting of stochastic variations of figures and backgrounds - i.e., figure-ground stimuli (Teki et al., 2011). The difficulty level was parametrically varied by changing the duration and coherence degree of the figure within the sequences. Recently, these figure-ground sequences have been shown to elicit distinct "signature" EEG responses, including object-related negativity (ORN) reflecting concurrent auditory object processing. In addition, participants were presented with a consonant identification task. Here, the consonant was presented during relative silence, in the presence of one interfering talker, and in the presence of speech-shaped-noise. Overall, our results replicate existing data showing that the ORN is modulated by the stream segregation task difficulty: Better performance is accompanied by larger ORN amplitudes. We found a significant correlation between the behavioral figure-ground performance and speech intelligibility in the presence of one interfering talker. There was no significant correlation between the amplitude of the ORN and intelligibility in noise. This is in line with recent data from our laboratory showing that perception of speech in noise develops from childhood until early adulthood. Yet selective attention remains immature until late adolescence. Data is currently being collected in a younger sample (8-18 years old), to evaluate the exact contribution of selective attention and stream segregation to speech intelligibility in noise over development.

Disclosures: J.F. Johnson: None. **E. Benocci:** None. **M. Couvignou:** None. **L. Leonardy:** None. **A. Calcus:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.17

Topic: D.05. Auditory & Vestibular Systems

Support: ERC AdG 788240
ANR-18-CE37-0022-03

Title: Fmri-guided electrophysiology of the macaque temporal voice areas

Authors: *M. GIAMUNDO, R. TRAPEAU, S. NOUGARET, X. DE GIOVANNI, L. RENAUD, T. G. BROCHIER, P. BELIN;
Aix-Marseille Université & CNRS, Marseille, France

Abstract: The ability to extract and process voice information is crucial for the social life of humans and other primates. Neuroimaging studies have shown the existence of temporal voice areas (TVAs) selective for conspecific vocalizations in both humans and non-human primates (Bodin, Trapeau et al., 2021), supporting the hypothesis of a functional homology in cerebral voice processing between humans and their closest relatives. But how is voice information treated at the neuronal level in these areas is still not clear.
To tackle this issue, we implanted two rhesus macaques with several high-density multi-electrode arrays in fMRI-localized voice areas of the superior temporal gyrus. Spiking activity was recorded during an auditory stimulation task (pure tone detection task) in which a set of n=96 stimuli from four categories (human voices, macaque vocalizations, marmoset vocalizations, non-vocal sounds) was presented. A total of 1582 auditory-responsive single (n=472) and multi-units (n=1110) was recorded from 4 arrays in the two monkeys.
Analyses indicate that a moderate proportion (29%) of cells was selective for conspecific (macaque) vocalizations, considerably smaller than the proportion of face-selective cells in the middle face patches (Tsao et al., 2006), confirming previous findings (Perrodin et al., 2011). However, at the population level, decoding analysis shows that spiking activity in the different TVAs allows above-chance classification of conspecific vocalizations from non-vocal sounds, with higher accuracy for more anterior TVAs. Spiking activity in the TVAs also allows classification of macaque call types as early as 65ms after the onset of the auditory stimulation, again with higher accuracy for the most anterior TVAs.
Furthermore, a Representational Similarity Analysis of neuronal responses to the 96 stimuli shows that in the anterior TVAs, the Representational Dissimilarity Matrices capturing pairwise spiking activity differences between stimuli show significant association with an ideal categorical model separating conspecific vocalizations from other sounds as early as 75ms after stimulus onset. This difference did not occur for non-vocal sounds.
These results advance our understanding of the neural substrates of voice information processing in macaques, and open a unique comparative window by allowing direct comparison with similar data obtained with humans.

Disclosures: M. Giamundo: None. **R. Trapeau:** None. **S. Nougaret:** None. **X. De Giovanni:** None. **L. Renaud:** None. **T.G. Brochier:** None. **P. Belin:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.18

Topic: D.05. Auditory & Vestibular Systems

Support: DST/INT/CZ/P-04/2020
SERB-CRG/2021/005653
IITKGP/SRIC/Challenge Grant/DMN

Title: Altered vocalization sequence encoding in a mouse model of ASDs

Authors: P. MANDAL[1], S. AGARWALLA[2], *S. BANDYOPADHYAY[1];
[1]IIT Kharagpur, Kharagpur, India; [2]ELECTRONICS AND ELECTRICAL COMMUNICATION ENGINEERING, INDIAN INSTITUTE OF TECHNOLOGY, KHARAGPUR, India

Abstract: Mice emit ultrasonic vocalisations sequences (USVs) to communicate socially relevant information and can thereby serve as a phenotypic marker of various neurodevelopmental disorders like autism spectrum disorders (ASDs). Although ASDs individuals are known to have significant communication problems, the possible reasons behind the impaired communication are poorly understood. In utero exposure of rodents to valproate (VPA) has been proposed to induce a phenotype with behavioral characteristics reminiscent of those observed in ASDs and provides a robust animal model for understanding of social communication and cognitive impairment in ASDs. We hypothesize that VPA exposure may lead to altered auditory processing of sequences of sounds, important in communication. Such deficits may contribute to the deficits in communication observed in individuals with ASDs. In this study, we document auditory cortex responses in mice prenatally exposed to VPA. We recorded in-vivo extracellular responses to mouse USV sequences with and without structure, quantified using mutual information. The informative USV sequences are referred to as natural sequences (SN) and designed sequences created to have random order of syllables are referred to as random sequences (SR) respectively. Interestingly, differential encoding is observed for SN but not for SR between the control and VPA mice. Thus, the auditory processing of USV sequences as a whole are altered in VPA male mice as it exhibits lower selectivity for SN sequences compared to control ones. Our study provides evidence in support of deficit in auditory processing of sound sequences in ASDs model mice as observed in human subjects thereby further establishing the importance of mouse as a powerful neurobiological model for studying vocal communication and related disorders.

Disclosures: P. Mandal: None. **S. Agarwalla:** None. **S. Bandyopadhyay:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.19

Topic: D.05. Auditory & Vestibular Systems

Support: Internal Funds

Title: Dynamic Shifts in Lateralized Activity Patterns Following Exposure to Novel Acoustic Environments

Authors: *B. FUREST CATALDO, P. DADIKA, D. S. VICARIO;
Rutgers Univ., Piscataway, NJ

Abstract: Lateralized activity suggesting functional specialization of corresponding brain regions is observed in sensory and motor systems for many tasks in many species, which may reflect an organizing principle of the nervous system. However, little is known about the degree to which asymmetrical processing depends on developmental experience and how it may be modified, temporarily or permanently, in adulthood. In the present study, we addressed the latter issue by exposing adult Zebra finches (ZFs), a songbird, to unfamiliar acoustic environments and tracking lateralized responses. Humans and songbirds both show lateralized processing for species-specific signals and are among the species of animals that learn their vocalizations from a model during a critical period. In ZFs, the *caudomedial nidopallium* (NCM) is a higher auditory region that exhibits lateralized responses, a bias for conspecific vocalizations, and the ability to form auditory memories; it can be considered functionally analogous to Wernicke's area. In adult birds, we have gone on to show that daily exposure to a novel acoustic environment (HETENV, the sounds of another songbird) for a period of 4-12 days causes the normal pattern to reverse from typical right- to atypical left-biased activity. Using both acute and chronic electrophysiology, we now show that activity returns to typical right-biased patterns after longer exposure (14+ days) to the novel environment; *the reversal and return was correlated with a facilitated discrimination of HETENV exemplars*. These data suggest that exposure to a novel acoustic environment challenges the auditory system differently in the two hemispheres, producing a transient reorganization followed by functional changes in neural processing. In the present study, we expanded this model by chronically recording adult ZFs exposed to two novel acoustic environments sequentially. We confirmed the previously observed transient shifts (i.e. reversal and return) during HETENV1 exposure and we go on to show that a successive HETENV2 exposure led to an additional set of transient shifts; the reversal and return were separately observed for both HETENVs. Upon termination of the exposures, acute microelectrode electrophysiology confirmed that the ZFs exhibited typical right-lateralized responses in NCM. Multiunit-activity analyses suggested that birds exposed to HETENVs displayed modifications in neuronal processing of HETENV exemplars relative to naïve counterparts. Together, these results show that typical lateralization patterns, which appear to be fixed, actually represent a modifiable steady state that can be dynamically updated by new experiences.

Disclosures: B. Furest Cataldo: None. **P. Dadika:** None. **D.S. Vicario:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.20

Topic: D.05. Auditory & Vestibular Systems

Support: ONR N00014-18-1-2062
ONR N00014-19-1-1223
ONR N00014-18-1-2069
ONR N00014-20-1-2709

Title: Source localization of dolphin (Tursiops truncatus) click evoked auditory potentials

Authors: *A. PEI[1], M. D. SCHALLES[2], J. MULSOW[3], D. S. HOUSER[3], J. J. FINNERAN[4], P. L. TYACK[5], B. G. SHINN-CUNNINGHAM[2];
[1]Carnegie Mellon Univ., [2]Neurosci. Inst., Carnegie Mellon Univ., Pittsburgh, PA; [3]Natl. Marine Mammal Fndn., San Diego, CA; [4]Naval Information Warfare Ctr. Pacific, United States Navy Marine Mammal Program, San Diego, CA; [5]Biol. Dept., Woods Hole Oceanographic Inst., Woods Hole, MA

Abstract: Animals use biological sonar (echolocation) to sense their environment through echoes from self-generated clicks. Dolphins excel at echolocation; however, the brain regions in the dolphin that parse echoes to analyze underwater acoustical scenes remain largely unknown. Over the last few decades, human electroencephalography (EEG) has matured to support localization of brain regions involved in neural computations by leveraging dense multi-electrode recording arrays. This study explored whether this paradigm could be applied in dolphins. We recorded 16-channel EEG in two adult male dolphins during a passive-listening task in air. Synthetic clicks were delivered via a jawphone on the animal's left jaw, at presentation rates of 2, 4, and 8 times per second with a 50-ms jittered interstimulus interval. Anterior electrodes 4 cm posterior to the blowhole exhibited a clear P1-N1-P2 complex with latencies ranging from 15 ms to 40 ms for all stimulation presentation rates. Faster presentation rates led to a decrease in the relative magnitudes of the event-related potentials (ERPs). We analyzed these ERPS using the weighted minimum norm eLORETA algorithm to localize neural regions responsible for sound processing. From a manually segmented T1 MRI structural scan of a dolphin, we identified scalp, skull, and brain tissue and generated a 3-layer boundary element model of the head out of triangular meshes, which was used by the eLORETA algorithm to estimate the locations of neural activity. Preliminary results indicate source activity was lateralized towards left parietal regions, which aligns with previous invasive localization of auditory cortices (Supin et al. 1978). However, present estimates in subcortical gray matter regions are inferior to those previous reports, which localized activity to more superficial regions. This study provides a framework for EEG source localization in dolphins using anatomical and electrophysiological data, but requires further validation and testing.

Disclosures: A. Pei: None. **M.D. Schalles:** None. **J. Mulsow:** None. **D.S. Houser:** None. **J.J. Finneran:** None. **P.L. Tyack:** None. **B.G. Shinn-Cunningham:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.21

Topic: D.05. Auditory & Vestibular Systems

Support: NIH R01NS099288-06

Title: Imaging Auditory Cortical Neuron Activity in the Singing Songbird

Authors: *F. DUARTE ORTIZ, R. MOONEY;
Neurobio., Duke Univ., Durham, NC

Abstract: To facilitate effective vocal communication, the auditory system must distinguish self-generated vocalizations from other sounds. In animals that only produce innate vocalizations, one way that this distinction is thought to be made is through vocal motor-related corollary discharge (CD) signals that suppress auditory cortical responses to predictable features of vocalization-related auditory feedback. Whether similar predictive suppression mediated by vocal CD signals characterizes the auditory cortex in animals that produce learned vocalizations remains poorly understood. In fact, prior electrophysiological recordings made in the auditory cortex of songbirds, which learn to sing, concluded that the majority of auditory cortical neurons are excited rather than suppressed during singing. Here, we used pan-neuronal calcium imaging in freely behaving adult zebra finches to determine whether and how corollary discharge signals modulate auditory cortical activity when birds sing. We found that of those neurons in the primary auditory cortex (i.e., Field L and CML) that were modulated during singing, the majority were suppressed compared to activity evoked during non-singing epochs by playback of the bird's song. Additionally, a small proportion of neurons in these regions were modulated up to 500ms prior to song onset. These results are consistent with a vocal corollary discharge signal that operates to suppress vocal feedback during singing. Furthermore, some neurons that were not modulated during normal singing were strongly excited when auditory feedback was perturbed with singing-triggered noise, which may indicate a predictive suppressive mechanism. To determine whether the modulatory effects of singing originate in the auditory cortex, or are instead inherited from upstream areas, we used fiber photometry to image calcium signals of auditory thalamic axon terminals in the primary auditory cortex (Field L2a). In contrast to auditory cortical neuron activity, we found that auditory thalamic terminals in Field L2a displayed similar patterns of activity during singing and song playback. In summary, calcium imaging in the zebra finch auditory forebrain reveals evidence of a vocal CD signal that functions predictively to suppress singing-related auditory feedback and hints that the circuit mechanism underlying these effects operates locally within the auditory cortex.

Disclosures: F. Duarte Ortiz: None. **R. Mooney:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.22

Topic: D.05. Auditory & Vestibular Systems

Title: The evolutionary continuity and divergence of auditory dorsal and ventral pathways in primates

Authors: *Y. ZHANG[1], S. SHEN[1], A. BIBIC[1], X. WANG[2];
[1]Johns Hopkins Univ., Baltimore, MD; [2]Dept Biomed Engin, Johns Hopkins Univ. Sch. Med., Baltimore, MD

Abstract: Auditory dorsal and ventral pathways in the human brain play important roles in supporting language processing. However, the evolutionary course of the dual auditory pathways remains largely unclear. By parcellating the auditory cortex of marmosets, macaques, and humans using the same individual-based analysis method and tracking the fiber pathways originating from the auditory cortex based on multi-shell diffusion-weighted magnetic resonance imaging (dMRI), homologous auditory dorsal and ventral fiber pathways were identified. Ventral pathways were found to be well conserved in the three primate species analyzed but extended to more anterior regions in humans. In contrast, dorsal pathways showed evolutionary divergence in two aspects: first, dorsal pathways in humans have stronger connections to higher-level auditory regions which extended beyond the corresponding regions in non-human primates; second, left lateralization of dorsal pathways was only found in humans. Moreover, dorsal pathways in marmosets are more similar to those in humans than in macaques. These results demonstrate the evolutionary continuity and divergence of dual auditory pathways in the primate brains, suggesting that the putative neural networks supporting human language processing emerged before the lineage of the New-Word primates diverged from the Old-Word primates and continued to parallelly evolve thereafter.

Disclosures: Y. Zhang: None. **S. Shen:** None. **A. Bibic:** None. **X. Wang:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.23

Topic: D.05. Auditory & Vestibular Systems

Support: NIH-NIDCD 5R01DC014279

Title: Distinct neural encoding of glimpsed and masked phonetic features in multitalker speech perception

Authors: *V. S. RAGHAVAN[1], J. A. O'SULLIVAN[1], S. BICKEL[2,3], A. D. MEHTA[2], N. MESGARANI[1];
[1]Electrical Engin., Columbia Univ., New York, NY; [2]Neurosurg., [3]Neurol., Hofstra Northwell Sch. of Med., Manhasset, NY

Abstract: Humans can easily tune in to one talker in a multitalker environment while still picking up bits of background speech; however, it remains unclear how we perceive speech that

is masked and to what degree non-target speech is processed. The glimpsing model of speech perception hypothesizes that speech is perceived in glimpses which provide sufficient information to restore masked portions, but this hypothesis has not been tested neurally. To test this model, we directly recorded the primary and non-primary auditory cortex (AC) in neurosurgical patients as they attended to one talker in multitalker speech, and used temporal response function (TRF) models to predict high gamma neural activity from glimpsed and masked stimulus features. We found that glimpsed speech is encoded at the phonetic level, invariant to attention, in primary AC. Neural sites in non-primary AC also encoded glimpsed speech but with enhanced encoding of the target talker. In contrast, the encoding of masked phonetics was only found for the target, with a greater response latency than glimpsed phonetics, and with a distinct posterior-to-anterior spatial organization. These findings suggest separate mechanisms for the encoding of glimpsed and masked speech and contribute to our understanding of the role top-down attention plays in restoring a complete representation of the target talker to enable robust perception.

Disclosures: V.S. Raghavan: None. **J.A. O'Sullivan:** None. **S. Bickel:** None. **A.D. Mehta:** None. **N. Mesgarani:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.24

Topic: D.05. Auditory & Vestibular Systems

Support: 5R01-DC-017690-02

Title: Time as a supervisor: temporal regularity and auditory object learning

Authors: *R. W. DITULLIO[1], E. PIASINI[2], C. K. PARTHIBAN[1], P. CHAUDHARI[1], V. BALASUBRAMANIAN[1], Y. E. COHEN[1];
[1]Univ. of Pennsylvania, Philadelphia, PA; [2]Cognitive Neurosci., Scuola Internazionale Superiore di Studi Avanzati (SISSA), Trieste, Italy

Abstract: Because objects in the natural world are rarely encountered in a context with explicit supervision, it is likely that sensory systems utilize some regularity of the natural objects in their environment in lieu of an explicit supervisor. We posit that one such regularity that could be used is temporal regularity. In this framework of learning objects, a sensory system identifies the features of a stimulus that change as consistently as possible over time, i.e. are maximally temporally regular. We explored this idea in the auditory system. The auditory system is an ideal test bed because auditory stimuli inherently evolve over time. Further, we know relatively little about the manner in which the auditory system learns to detect and differentiate between auditory objects. Specifically, we tested two related predictions about natural auditory objects: 1) natural auditory objects will have significantly temporally regular features and 2) an

unsupervised algorithm that learns these features will generate a feature space that can support classification. We tested these two predictions with rhesus macaque vocalizations, recordings of applause, and white noise as a control. These stimuli were analyzed using Slow Feature Analysis (SFA), which is an unsupervised temporal learning algorithm. SFA extracts features that either capture just the continuity of frequencies over time (linear SFA) or capture both the continuity of frequencies and the continuity of the correlations between frequencies (quadratic SFA). As an algorithmic control, we compared the SFA results with principal component analysis (PCA). We found: 1) natural auditory objects show significant temporal regularity both in the continuity of frequencies as well as continuity of correlations between frequencies; 2) vocalizations had more regularity than applause or white noise; 3) classification performance was highest for quadratic SFA due to the fact its features capture both continuity and the continuity of correlations; 4) classification performance correlates with temporal regularity; And 5) that these trends held even when the stimuli were embedded in naturalistic clutter. These results suggest that learning continuity of changes in the correlations of auditory stimuli may be sufficient for parsing auditory scenes, providing a powerful computational mechanism that the brain could utilize for auditory perception.

Disclosures: **R.W. DiTullio:** None. **E. Piasini:** None. **C.K. Parthiban:** None. **P. Chaudhari:** None. **V. Balasubramanian:** None. **Y.E. Cohen:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.25

Topic: D.05. Auditory & Vestibular Systems

Support: ERC AdG 788240

Title: Investigations of cortical voice-selectivity in marmosets using anesthetized fMRI at 3T

Authors: M. OBLIGER, R. TRAPEAU, L. BOUDIN, J. SEIN, B. NAZARIAN, J.-L. ANTON, E. RAPHA, L. RENAUD, ***P. BELIN**;
Aix Marseille Univ., Marseille, France

Abstract: Functional MRI studies in humans (Pernet et al, 2015) and macaques (Petkov et al, 2008; Bodin et al, 2021) suggest the existence of a “primate voice patch system”: a set of interconnected areas selective to conspecific vocalizations and subtending representations of increasing abstractness and invariance, potentially analogous to the primate face patch system. To date, a single marmoset auditory fMRI study compared conspecific vocalizations to nonvocal sounds, with results suggesting the existence of bilateral voice patches in the marmoset anterior temporal lobe (Sadagopan et al, 2015). Here we aimed to replicate and extend these results by scanning n=6 marmosets under anesthesia during auditory stimulation with 4 categories: marmoset vocalizations, natural non-vocal sounds, scrambled vocalizations, and silence. We

used a 3T scanner (Siemens PRISMA) and a commercial 16-channel marmoset head coil (Takashima/Rogue Research). Anesthesia was performed using sevoflurane delivered by a mask. A few minutes prior to functional scanning sevoflurane level was lowered to 1.5%-1%. In later scanning sessions N2O was added to the gaseous mix in order to reduce further the sevoflurane level to 0.5-0.8%. Functional scanning was performed with a spatial resolution of 1 (TR=773ms) or 1.25mm (TR=598ms) using an optimized 'clustered-sparse' design: mini-blocks of 3-5 auditory stimuli from a same category presented in silence (no EPI), followed by rapid acquisition of 5-7 EPI volumes. 5-10 functional runs of about 5 minutes were acquired per session. The comparison of EPI volumes acquired after sound stimulation vs. the silent baseline yielded varying results depending on the runs and sessions, with some subjects showing mostly subcortical activation and others showing nice bilateral activation of auditory cortex. Jacknife analyses indicate an effect of sevoflurane levels, with higher t-statistics for lower levels. The comparison of marmoset vocalizations vs. the nonvocal sounds did not yield voice-selective activation, possibly owing to the large difference in spectral distribution between the two categories. However, the comparison with scrambled vocalizations did result in bilateral voice-selective activation in one subject, in a location of the temporal pole very similar to that reported in Sadagopan et al. (2015). Ongoing work is including more subjects and the addition of MION injections as a contrast agent. These results are expected to shed greater light on the neural architecture underlying voice information processing in marmosets, and provide a strong test of the hypothesis of a primate voice patch system.

Disclosures: M. Obliger: None. **R. Trapeau:** None. **L. Boudin:** None. **J. Sein:** None. **B. Nazarian:** None. **J. Anton:** None. **E. Rapha:** None. **L. Renaud:** None. **P. Belin:** None.

Poster

543. Auditory Processing: Natural Sounds and Vocalizations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 543.26

Topic: D.05. Auditory & Vestibular Systems

Support: NIH R01DC015138
NIH R01DC020097

Title: Categorical perception and coding of chimeric fire and water sounds driven by sound and neural summary statistics

Authors: *M. ESCABI[1], M. SADEGHI[2], X. ZHAI[3], D. PEDRICK[4], I. H. STEVENSON[5]; [2]Electrical and Computer Engin., [3]Electrical & Computer Engin., [4]Biomed. Engin., [5]Dept. of Psychological Sci., [1]Univ. of Connecticut, Storrs, CT

Abstract: Categorical perception, the ability to group sounds or images into discrete perceptual categories, enables humans and other animals to rapidly and robustly recognize and respond to stimuli. Categorical representations have been observed for speech and other vocalized sounds

along relatively simple time and frequency dimensions. However, the high-order acoustic features and neural computations underlying categorical representations for more general sound categories are not well understood.We used a texture synthesis (McDermott & Simon celli 2011) to synthesize 'chimeric' auditory textures. Synthetic chimeric textures were generated by morphing summary statistics of *running water* and *crackling fire*sounds. The chimeric sounds were first used in human perception tasks were participants were either required to *identify* a chimeric texture as fire or water or alternately required to *discriminate* two chimeric textures. For both tasks, the chimeric textures statistics were varied by adjusting either the included summary statistics during the synthesis and the morph ratio (MR) from 0 (water extreme) to 1 (fire extreme). Parallel studies were also carried out to study the neural representation of chimeric textures in the inferior colliculus (IC) of unanesthetized rabbits. Neural activity was obtained using multi-channel silicon probes inserted along the principal frequency axis during passive listening of the chimeric textures.We demonstrate that shifting summary statistics by changing the morph ratio produces a robust shift in human listener's perception. Participants readily identified water and fire sounds in a categorical-like fashion with increasing morph ratio where the sound correlation structure was the critical and necessary statistical cue responsible for the categorical effect. The categorical shift is accompanied by lower discrimination accuracy and larger just noticeable difference limen (JND) for within category as compared to across category discrimination. We then demonstrate how the response statistics of neural ensembles in the IC can be used to decode the sound category. Bayesian neural decoders were used to assess how population response summary statistics can categorize or discriminate the chimeric textures. Shifting the morph ratio produces a similar categorical-like shift in neural decoding performance and similar larger JNDs and lower accuracy trends as for human participants.The findings suggest that high-order statistical sound cues can drive categorical-like perception for textures and that the neural response statistics in IC can contribute towards such phenomenon.

Disclosures: **M. Escabi:** None. **M. Sadeghi:** None. **X. Zhai:** None. **D. Pedrick:** None. **I.H. Stevenson:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.01

Topic: D.05. Auditory & Vestibular Systems

Support: Whitehall Foundation Research Grant 2018-08-88
Brain, and Behavior NARSAD Young Investigator Grant 27668
Alzheimer Association Research Grant AARG-21-850571

Title: Auditory cortical mechanisms mediating sound-guided interval time keeping

Authors: *H. SURI[1], G. ROTHSCHILD[2];
[1]Psychology, [2]Psychology, Kresge Hearing Res. Inst. and Dept. of Otolaryngology, Univ. of Michigan, Ann Arbor, MI

Abstract: Everyday decisions such as when to cross a busy street or when it is appropriate to speak in a conversation rely on the ability to implicitly but reliably determine the amount of time passed from incoming sensory cues. However, the neural mechanisms that underlie cue-triggered time estimation on the timescale of seconds in support of such adaptive behavior are not well understood. To address this gap, we developed an appetitive sound-guided interval time keeping behavior in head-fixed mice, which is based on reward-predictive licking across varying sound-reward time intervals. We find that mice trained on this task reliably estimate the time from a sound cue, as demonstrated by learning-dependent timed increases in reward-predictive licking. We next asked whether and how the auditory cortex is involved in this sound-triggered time estimation behavior. Inactivation of the auditory cortex dramatically impaired animals' ability to use sounds to predict the timing of upcoming reward. Mice trained on a sound-reward association with no interleaving time gap showed spared performance following auditory cortical inactivation, suggesting that basic sound detection remained intact. Finally, recordings of neural activity during learning and performance of this behavior reveal auditory cortical neurophysiological signatures underlying sound-guided interval time keeping. Together, our findings identify auditory cortical mechanisms supporting sound-triggered timing-dependent behaviors.

Disclosures: H. Suri: None. **G. Rothschild:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.02

Topic: D.05. Auditory & Vestibular Systems

Support: Whitehall Foundation Research Grant #2018-08-88
NARSAD Young Investigator Grant #27668
Claude D. Pepper Center Grant #AG024824
National Institute of Health grant 2R01MH063649
National Institute of Health training grant T32-DC000011

Title: Integration of sound and locomotion information by auditory cortical neuronal ensembles

Authors: *C. VIVALDO[1], J. LEE[2], M. SHORKEY[1], A. KEERTHY[1], G. ROTHSCHILD[3];
[1]Univ. of Michigan, Ann Arbor, Ann Arbor, MI; [2]UCSD, La Jolla, CA; [3]Dept. of Psychology, Univ. of Michigan, Ann Arbor, MI

Abstract: The ability to process and act upon incoming sounds during locomotion is critical for survival. Intriguingly, sound responses of auditory cortical neurons are on average weaker during

locomotion as compared to immobility and these results have been suggested to reflect a computational resource allocation shift from auditory to visual processing. However, the evolutionary benefit of this hypothesis remains unclear. In particular, whether weaker sound-evoked responses during locomotion indeed reflect a reduced involvement of the auditory cortex, or whether they result from an alternative neural computation in this state remains unresolved. To address this question, we first used neural inactivation in behaving mice and found that the auditory cortex plays a critical role in sound-guided behavior during locomotion. To investigate the nature of this processing, we used two-photon calcium imaging of local excitatory auditory cortical neural populations in awake mice. We found that underlying a net inhibitory effect of locomotion on sound-evoked response magnitude, spatially intermingled neuronal subpopulations were differentially influenced by locomotion. Further, the net inhibitory effect of locomotion on sound-evoked responses was strongly shaped by elevated ongoing activity. Importantly, rather than reflecting enhanced “noise”, this ongoing activity reliably encoded the animal’s locomotion speed. Prediction analyses revealed that sound, locomotive state and their integration are strongly encoded by auditory cortical ensemble activity. Finally, we found consistent patterns of locomotion-sound integration in electrophysiologically recorded activity in freely moving rats. Together, our data suggest that auditory cortical ensembles are not simply suppressed by locomotion but rather encode it alongside sound information to support sound perception during locomotion.

Disclosures: **C. Vivaldo:** None. **J. Lee:** None. **M. Shorkey:** None. **A. Keerthy:** None. **G. Rothschild:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.03

Topic: D.05. Auditory & Vestibular Systems

Support: DOD/MEDCOM/CDMRP W81XWH-18-1-0135

Title: Long-duration sound induced plasticity is altered differently in mice with tinnitus compared to those without tinnitus.

Authors: *A. L. BURGHARD, E. FABRIZIO-STOVER, C. M. LEE, D. L. OLIVER; Neurosci., Univ. of Connecticut Hlth. Ctr., Farmington, CT

Abstract: A presentation of a long-duration sound (LDS) can lead to a change in both spontaneous activity as well as sound-driven activity in the inferior colliculus (IC) in mice. While the majority of sound-driven responses are suppressed, a subset is potentiated after the LDS. This potentiation is more likely in channels with higher spontaneous activity. Since tinnitus is associated with increased spontaneous activity in the auditory system, we hypothesize that tinnitus animals will have more facilitation/less suppression than animals without tinnitus.

Exposing awake CBA/CaJ mice to a unilateral sound resulted in mice with and without behavioral signs of tinnitus. Using multichannel electrodes, we recorded the activity in the inferior colliculus contra- and ipsi-lateral to the sound exposed ear. The spontaneous activity in the contralateral IC was higher in the tinnitus group than in the sound-exposed non-tinnitus and the control (not sound-exposed) group. When comparing LDS-driven plasticity between mice with and without behavioral signs of tinnitus, we find in the contralateral IC that the sound exposed non-tinnitus animals show more suppression than tinnitus animals exposed to the same sound. The tinnitus animals show a response that is more similar to control (not sound-exposed) animals. When comparing the change in spontaneous activity following an LDS presentation, we find that in the contralateral IC tinnitus animals show less afterdischarge activity (significant increase in spontaneous activity) than both the control and the sound exposed non-tinnitus animals. Taken together this indicates an LDS-induced difference in sound-exposed tinnitus vs non-tinnitus animals that might serve as an objective test to differentiate between hearing loss with or without tinnitus.

Disclosures: A.L. Burghard: None. **E. Fabrizio-Stover:** None. **C.M. Lee:** None. **D.L. Oliver:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.04

Topic: D.05. Auditory & Vestibular Systems

Support: DOD/MEDCOM/CDMRP W81XWH-18-1-0135

Title: Long-duration sound-evoked changes as a novel test for tinnitus in mice

Authors: *E. FABRIZIO-STOVER[1], A. L. BURGHARD[2], C. M. LEE[3], D. L. OLIVER[4]; [1]UConn Hlth., rocky hill, CT; [2]Neurosci., Univ. of Connecticut Hlth. Ctr., Farmington, CT; [3]Neurosci., Uconn Hlth., Farmington, CT; [4]Neurosci., Univ. of Connecticut, Farmington, CT

Abstract: An objective, non-invasive electrophysiological test for tinnitus would allow for the efficient diagnosis of tinnitus in laboratory animals and is needed to accelerate tinnitus research. In CBA/CaJ mice of both sexes, a long-duration sound (LDS) can alter both spontaneous firing rate and responses to sound in the central nucleus of the inferior colliculus (ICC). Two changes in sound-driven responses are seen after the LDS: suppression and facilitation. Because tinnitus is associated with higher spontaneous neuronal firing rates, we hypothesize that tinnitus individuals will show less LDS-dependent depression and more facilitation than non-tinnitus individuals. Awake CBA/CaJ mice (n=44) received a unilateral sound exposure (16 kHz center, 2 kHz wide, 113 dB SPL, 60 minutes) that resulted in mice with and without behavioral evidence of tinnitus in an active avoidance task. Auditory brainstem responses (ABRs) were collected from each ear (exposed and unexposed) in sound exposed tinnitus (n=22), sound-exposed non-

tinnitus (n=22), and unexposed control mice (n=10). ABRs to tone pips, chirps, and narrowband noise were collected before and after LDS. For each type of stimulus, responses to three or more frequencies were collected including the frequency of the presumed tinnitus in mice with tinnitus. Multiple analysis methods were used to analyze the ABR waveforms. First, we quantified the effect of LDS-changes and calculated a change score for each wave based on peak-trough amplitudes. Tone pip ABRs evoked by sounds in the exposed ear showed that non-tinnitus mice have lower scores than tinnitus mice. That is, non-tinnitus mice had more suppressed POST-LDS ABRs compared to tinnitus mice (One-way ANOVA, Bonferroni post-hoc test, p=0.020). This effect was most obvious in tone evoked ABRs and not chirp or narrowband noise ABRs. A correlation analysis comparing PRE-LDS and POST-LDS tone pip evoked waveforms showed that non-tinnitus mice were more significantly affected by the LDS than tinnitus mice (One-way ANOVA, Bonferroni pos-hoc test, p=0.046). A differential time frequency analysis showed that in the spectrum of the ABR waveforms over time there was a tinnitus specific 'hotspot' of increased activity at frequency of the tinnitus. These 'hotspots' were not seen for non-tinnitus animals or for non-tinnitus frequencies. These data suggest that ABRs collected before and after an LDS may be a viable method for developing a non-invasive, electrophysiological test for tinnitus.

Disclosures: E. Fabrizio-Stover: None. **A.L. Burghard:** None. **C.M. Lee:** None. **D.L. Oliver:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.05

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant DC009836

Title: Functional dissection of disinhibition versus sensitization in the emergence of cortical hyperexcitability after hearing loss

Authors: *B. AWWAD, Y. WATANABE, O. STEVENSON, L. CASEY, K. CLAYTON, D. B. POLLEY;
Mass Eye and Ear infirmary ,Department of Otolaryngology - Head and Neck Surgery, Harvard Med. Sch., Boston, MA

Abstract: After sensorineural hearing loss, auditory cortex (ACtx) neurons become hyperresponsive to sound - excess central gain -, a core feature of tinnitus and hyperacusis. *Ex vivo* experiments suggest that auditory hyperresponsivity could arise through two mechanisms: either disinhibition via reduced feedforward inhibition or sensitization via enhanced glutamatergic inputs. Here, we developed a novel optogenetic approach to put these ideas to the test in the intact ACtx. We used a triple virus strategy to express ChR2 in parvalbumin+ (PV)

GABAergic neurons and a somatically restricted, red-shifted opsin in contralateral neurons that project to the ACtx via the corpus callosum, allowing independent optical control over select populations of inhibitory (PV) and excitatory (callosal) neurons. High-density translaminar recordings were made from the high-frequency region of A1 in awake, head-fixed mice up to three days following acoustic trauma or an innocuous sham exposure. Sound intensity growth functions from regular spiking putative pyramidal neurons were markedly increased after acoustic trauma (n = 484 units in 6 mice), particularly in layer 5, compared to sham exposure (n=402 units in 5 mice). Dual optogenetic activation revealed that excess auditory gain was accompanied by a striking disinhibition, as measured from reduced PV-mediated suppression of spiking, without any evidence of sensitization to direct activation of excitatory callosal neurons. Hyperresponsivity from deep layer projection neurons via disinhibition could induce strong coupling with limbic brain regions and negative auditory affects after acoustic trauma, a possibility that we are exploring in ongoing experiments via dual recordings from the ACtx and basolateral amygdala.

Disclosures: **B. Awwad:** None. **Y. Watanabe:** None. **O. Stevenson:** None. **L. Casey:** None. **K. Clayton:** None. **D.B. Polley:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.06

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant R00-DC015543
NIH Grant R01-DC012557
NIH Grant P01-NS074972
NIH Grant U19-NS107616
NARSAD Young Investigators Award

Title: Contributions and synaptic basis of diverse cortical neuron responses to task performance

Authors: *B. F. ALBANNA[1], J. TOTH[2], B. DEPASQUALE[3], S. FADAEI[4], T. GUPTA[2], K. KUCHIBHOTLA[5], K. RAJAN[6], R. C. FROEMKE[4,7], M. N. INSANALLY[2];
[1]Neurosci., [2]Otolaryngology, Univ. of Pittsburgh, Pittsburgh, PA; [3]Princeton Neurosci. Inst., Princeton Univ., Princeton, NJ; [4]Skirball Institute, Neurosci. Institute, Depts. of Otolaryngology, Neurosci. and Physiol., New York University, Grossman Sch. of Med., New York, NY; [5]Departments of Psychological and Brain Sciences, Neuroscience, and Biomed. Engin., Johns Hopkins Univ., Baltimore, MD; [6]Neurosci., Icahn Sch. of Med. At Mount Sinai Grad. Training Program In Neurosci., New York, NY; [7]Ctr. for Neural Sci., New York Univ., New York, NY

Abstract: Neuronal responses during behavior are diverse, ranging from highly reliable ‘classical’ responses to irregular or seemingly random ‘non-classically responsive’ firing.

Various spiking patterns (or lack thereof) have been documented throughout brain regions including visual, auditory, parietal, and frontal cortices in response to different sensory inputs, in relation to decision making, motor actions, or other task-related signals. To explore the synaptic origins and contributions of diverse response profiles to network function, perception, and behavior, we combined in vivo cell-attached, extracellular, and whole-cell recordings during behavior with analyses of a novel task-performing spiking recurrent neural network. We recorded from the auditory cortex of rats and mice during a go/no-go auditory recognition task (rats: d' = 2.8±0.1, N = 15; mice: d' = 2.5±0.1, N = 7). In both species, we observed a wide range of single-unit response types from classically responsive cells that were highly modulated relative to pre-trial baseline to non-classically responsive cells with relatively unmodulated firing rates. To relate synaptic structure to spiking patterns over the response-type continuum, we developed a spiking recurrent neural network model incorporating both excitatory and inhibitory spike-timing-dependent plasticity trained to perform a similar go/no-go stimulus classification task as behaving animals. This model captures the distribution of heterogeneous responses observed in the auditory cortex of behaving rodents. Detailed inactivation experiments revealed that classically responsive and non-classically responsive model units contributed to task performance via output and recurrent connections, respectively. Excitatory and inhibitory plasticity independently shaped spiking responses to increase the number of non-classically responsive units while keeping the full network (all units) engaged in performance. Local patterns of synaptic inputs predicted spiking response properties of network units as well as the responses of auditory cortical neurons from in vivo whole-cell recordings during behavior allowing us to predict the functional role of a neuron from the pattern of synaptic inputs. Thus, a diversity of neural response profiles emerges from synaptic plasticity rules with distinctly important functions for network performance.

Disclosures: **B.F. Albanna:** None. **J. Toth:** None. **B. DePasquale:** None. **S. Fadaei:** None. **T. Gupta:** None. **K. Kuchibhotla:** None. **K. Rajan:** None. **R.C. Froemke:** None. **M.N. Insanally:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.07

Topic: D.05. Auditory & Vestibular Systems

Support: NIH NIDCD R01DC015527
NIH NIDCD R01DC014479
NIH NINDS R01NS113241
NIH NIDCD 5T32DC016903-04

Title: Auditory cortex controls categorization acuity

Authors: *J. S. COLLINA[1], M. XIA[2], G. ERDIL[3], J. SHETH[3], K. P. KORDING[2], Y. E. COHEN[3], M. N. GEFFEN[3];
[1]Dept. of Neurosci., [2]Dept. of Bioengineering, [3]Dept. of Otorhinolaryngology, Univ. of Pennsylvania, Philadelphia, PA

Abstract: In everyday life, because both sensory signals and neuronal responses are noisy, important cognitive tasks, such as auditory categorization, are based on uncertain information. At the neuronal level, categorization requires a transformation of sensory representation into a representation of category membership. While categorical representations were found in the cortex, the cell types and auditory neuronal mechanisms supporting the emergence of these representations remains unknown. The broader goal of our project is to understand the neuronal circuits that support decision-making based on information in the region of uncertainty.
Here, to test the role of the auditory cortex (AC) in creating and biasing categorical stimulus representations, we trained mice in a two-alternative-forced choice task in which mice categorize the frequency of a "target" sound into one of two overlapping categories ("low" or "high"). We reversibly suppressed cortical activity through bilateral muscimol injection in AC of trained mice in order to test the hypothesis that AC is involved in categorizing pure tones. Mice maintained their ability to perform the task. However, suppressing AC activity induced an attenuation in categorization accuracy to trained stimuli. In addition, inactivation of the AC resulted in a significant broadening of the psychometric slopes, which is an indicator of how certain mice are about the stimuli near the perceived category boundary when making their decisions. These findings suggest that the auditory cortex controls categorization acuity, but is not necessary for stimulus discrimination in general. Our results lay the groundwork for further exploration of the role of the auditory cortex in categorization behavior under uncertainty.

Disclosures: J.S. Collina: None. **M. Xia:** None. **G. Erdil:** None. **J. Sheth:** None. **K.P. Kording:** None. **Y.E. Cohen:** None. **M.N. Geffen:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.08

Topic: D.05. Auditory & Vestibular Systems

Support: R00MH116100
NIGMS P01GM118269
NIMH R01MH11559
Vanderbilt Startup Funds
the MIT Picower Institute Innovation Fund
The JPB Foundation
ONR N00014-22-1-2453

Title: Predictive processing without consciousness in the auditory cortex

Authors: *Y. XIONG[1], J. A. DONOGHUE[2], M. K. MAHNKE[3], M. LUNDQVIST[4], E. BROWN[4], A. BASTOS[1], E. MILLER[4];
[1]Vanderbilt Univ., Nashville, TN; [2]The Picower Inst. for Learning and Memory, MIT, Boston, MA; [3]MIT, MIT, Cambridge, MA; [4]Massachussetts Inst. of Technol., Massachussetts Inst. of Technol., Cambridge, MA

Abstract: Predictive coding is a fundamental function of the cortex. The Predictive Routing model provides a theoretical framework for predictive coding. Predictions are fed back via deep-layer cortex via alpha/beta (8-30Hz). They inhibit the gamma (40-100Hz) and spiking that would have fed forward predicted sensory inputs. Prediction errors are fed forward gamma/spiking in circuits unaffected by alpha/beta.
To test the Predictive Routing model and its role in consciousness, we collected data from intracranial recordings of macaque monkeys during passive presentation of auditory oddballs before and after loss of consciousness (LOC) due to propofol anesthesia. In local oddballs, a single stimulus is repeated until an oddball appears (AAAAB). In global oddballs, a stimulus pattern is repeated until an oddball pattern appears (AAAAB, AAAAB, AAAAA). Neural responses are generally enhanced to oddballs. Local oddball effects depend on both bottom-up and top-down mechanisms. Global (pattern) oddball effects depend even more strongly on top-down predictions and the frontal cortex. While awake, local oddballs (prediction errors) resulted in increased gamma power and spiking in sensory cortex. By contrast, global oddballs increased alpha/beta power in higher cortex. This is consistent with the Predictive Routing model. After LOC, there was a loss of global prediction error signals from multi-unit activity in higher-order cortex, but not sensory cortex. Gamma power to local oddballs in sensory cortex increased whereas alpha/beta prediction error signals to global oddballs in higher cortex was lost. Furthermore, Current Source Density analysis revealed increased input layer current sink during prediction error in awake sensory cortex. Under unconsciousness, superficial layers displayed larger current sinks to local prediction error.
We hypothesize that the high-amplitude slow waves (~1 Hz) that characterize propofol-induced LOC interfere with predictive routing. Our results show that slow wave phase modulates stimulus processing as well as prediction error processing, such that depolarized phases are related to higher spiking responses. In summary, loss of consciousness is marked by the lack of active top-down inhibition of feedforward prediction error signals, reflected by increased gamma power, spiking, and superficial layer current sinks, as well as lack of alpha/beta prediction signals and lack of prediction error signals in higher order cortex spiking. This supports the predictive routing model's hypothesis that top-down feedback through inhibition of feedforward stream is key to predictive processing and consciousness.

Disclosures: Y. Xiong: None. **J.A. Donoghue:** None. **M.K. Mahnke:** None. **M. Lundqvist:** None. **E. Brown:** None. **A. Bastos:** None. **E. Miller:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.09

Topic: D.05. Auditory & Vestibular Systems

Support: European Community, Human Brain Project
Bettencourt Foundation
ANR (Paradox)
European Research Council (Deepen)

Title: Awake perception is associated with dedicated neuronal assemblies in cerebral cortex

Authors: A. FILIPCHUK[1], J. SCHWENKGRUB[2], B. BATHELLIER[2], ***A. DESTEXHE**[1]; [1]Inst. of Neurosci. (NeuroPSI), CNRS, Paris-Saclay Univ., Saclay, France; [2]Inst. of Audition, Pasteur Inst., Paris, France

Abstract: Neural activity in sensory cortex combines stimulus responses and ongoing activity, but it remains unclear whether they reflect the same underlying dynamics or separate processes. Here we show that during wakefulness, the neuronal assemblies evoked by sounds in the auditory cortex and thalamus are specific to the stimulus and distinct from the assemblies observed in ongoing activity. In contrast, we observed in three different anesthesia, that evoked assemblies are indistinguishable from ongoing assemblies in the cortex. However, they remain distinct in the thalamus. A strong remapping of sensory responses accompanies this dynamical state change produced by anesthesia. Together, these results show that the awake cortex engages dedicated neuronal assemblies in response to sensory inputs, which we suggest is a network correlate of sensory perception.

Disclosures: A. Filipchuk: None. **J. Schwenkgrub:** None. **B. Bathellier:** None. **A. Destexhe:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.10

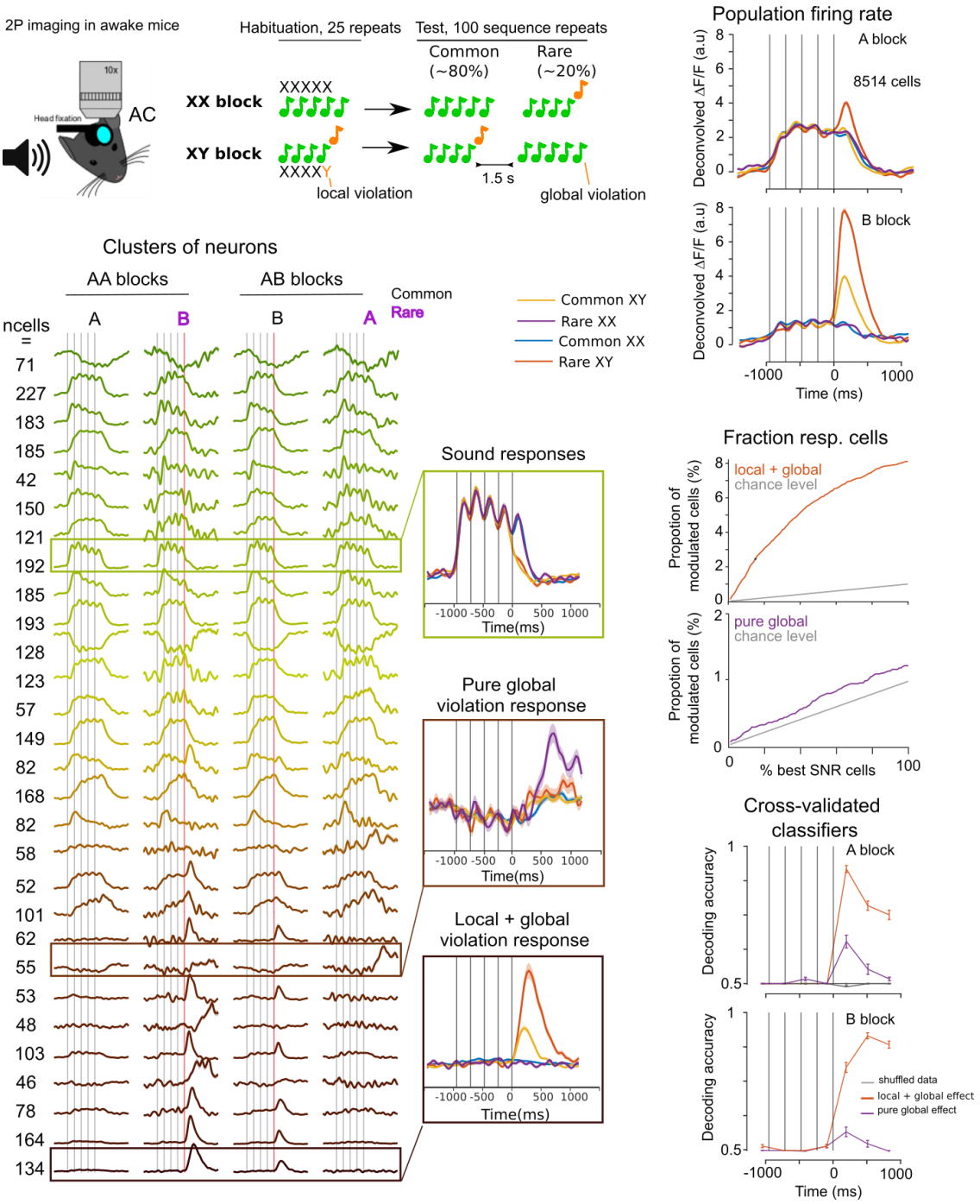
Topic: D.05. Auditory & Vestibular Systems

Support: ERC

Title: Predictive coding of global sequence violation in the mouse auditory cortex

Authors: ***S. JAMALI**[1], S. DEHAENE[2,3], T. VAN KERKOERLE[3], B. BATHELLIER[1]; [1]Hearing institut, Pasteur institut, Paris, France; [2]Collège de France, Paris, France; [3]NeuroSpin, Cognitive Neuroimaging Unit, CEA, INSERM, Univ. Paris-Sud, Univ. Paris-Saclay, NeuroSpin center, Gif sur Yvette, France

Abstract: The ability to extract temporal regularities at different time scales in sensory inputs and detect unexpected deviations from these regularities is a key cognitive ability. The classical auditory oddball paradigm shows that the brain responds to sequence violations at a local time scale, but such responses also occur under anesthesia and therefore seem pre-attentive. In contrast, recent studies in humans and monkeys suggest that when the violation concerns regularities occurring over longer time scales, responses to the violation appear only in conscious, attentive subjects. To investigate whether local and global sequence violation responses exist in the mouse, we recorded from layer 1 to 5 of the auditory cortex using two-photon calcium imaging while mice passively listened to repetitions of 1s-long sequences of five tones. The repeated short sequence contained either a single tone (AAAAA) or a local violation at its end (AAAAB). Purely global violations could be generated by presenting occasionally the AAAAA sequence in a block where AAAAB is repeated. We found that a population of neurons in the auditory cortex specifically responds to such purely global violations at the end of the AAAAA sequence. Although small, this population contained enough information to predict violations on single trials. A larger fraction of neurons boosted their responses to combinations of local and global violations (AAAAB presented in an AAAAA block). These global responses were resistant to a wide increase of inter-sequence interval (1.5s - 25s) ruling out that short-term adaptation causes these responses. However, global responses vanished when the difference between A and B sounds is less salient to the mouse. In anesthetized animals we find a reduced response to the local violations compared to awake animals but the purely global violations disappeared. These results establish that the mouse brain is able to detect global violations in sound sequences in a subgroup of auditory cortex neurons, paving the way for the study of circuit mechanisms underlying long-term temporal regularity detection.



Disclosures: **S. Jamali:** None. **S. Dehaene:** None. **T. Van Kerkoerle:** None. **B. Bathellier:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.11

Topic: D.05. Auditory & Vestibular Systems

Support: NIH R21DC018327
Klingenstein-Simons Fellowship Award in Neuroscience
Hearing Health Foundation Emerging Research Grant

Title: Extratelencephalic neurons encode learned stimulus categories and behavioral choice

Authors: *N. A. SCHNEIDER[1], T. SUAREZ OMEDAS[2], R. F. KRALL[1], R. S. WILLIAMSON[1];
[1]Otolaryngology, Univ. of Pittsburgh, Pittsburgh, PA; [2]Carnegie Mellon Univ., Pittsburgh, PA

Abstract: Auditory-guided behavior is ubiquitous in everyday life, whenever auditory information is used to guide our decisions and actions. Nestled amongst several populations, extratelencephalic (ET) neurons reside in the deep layers of auditory cortex (ACtx), integrate inputs from brain-wide sources, and provide a primary means of routing auditory information to diverse, sub-cortical targets associated with decision-making, action, and reward. To investigate the role of ET neurons in auditory-guided behavior, we developed a head-fixed choice task, where mice were trained to categorize the rate (as high or low) of sinusoidal amplitude-modulated (sAM) noise bursts to receive a water reward. Using bilateral optogenetic inhibition (with GtACR2), we established that ACtx was necessary for performance of this task. We used two-photon calcium imaging and selective expression of GCaMP8s to longitudinally monitor the activity of ET and L2/3 intratelencephalic (IT) populations.

Statistical clustering analyses of both ET and IT populations revealed heterogenous response motifs that correlated with various stimulus and task variables. One such motif, primarily present in ET neurons, corresponded to “categorical” firing patterns (i.e., neurons that responded best to low or high sAM rates), This categorical selectivity was not present early in training, and longitudinal recording revealed that ET neurons shift their response profiles dynamically across learning to reflect these discrete perceptual categories. We also included a sAM rate at the category boundary, rewarded probabilistically, allowing us the ability to investigate stimulus-independent choice. Using statistical approaches to visualize high-dimensional activity in a low-dimensional space revealed that ET activity reflected behavioral choice, regardless of reward outcome. We further quantified this using decoding analyses and confirmed that choice at the category boundary could be robustly predicted from ET activity. Both choice and categorical selectivity was notably lessened in the L2/3 IT population, providing further evidence of ET involvement.

Critically, the shift in ET selectivity was only present when mice were actively engaged in behavior and was not present during passive presentation of identical stimuli within the same imaging session. This suggests that learned categorical selectivity is shaped via top-down inputs that act as a flexible, task-dependent filter, a hypothesis that we are actively pursuing. These results suggest that the ACtx ET projection system selectively propagates behaviorally relevant signals brain-wide and is critical for auditory-guided behavior.

Disclosures: **N.A. Schneider:** None. **T. Suarez Omedas:** None. **R.F. Krall:** None. **R.S. Williamson:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.12

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant 5R01DC012557-09
DARPA N66001-17-2-4010

Title: Experience in auditory conditioning impacts across-animal variability in neural tuning

Authors: *K. A. MARTIN[1,3,4,5], C. J. BREDENBERG[1], J. LEI[1], E. P. SIMONCELLI[1,7], C. SAVIN[1,2], R. C. FROEMKE[6,3,4,1];
[1]Ctr. for Neural Sci., [2]Ctr. for Data Sci., New York Univ., New York, NY; [3]Neurosci. Inst., [4]Dept. of Neurosci. and Physiol., [5]Dept. of Otolaryngology, [6]Otolaryngology, New York Univ. Sch. of Med., New York, NY; [7]Ctr. for Computat. Neurosci., Flatiron Inst., New York, NY

Abstract: Perceptual learning is associated with altered cortical representations of task-relevant stimuli in trained animals relative to naive ones (Polley, et al., 2006; Edeline, et al., 1993; Bao, et al., 2004). While there is substantial variability across animals in the degree of behavioral learning and the associated changes in neural representations, we lack an account of how experiences during learning may drive these differences. To address these limitations, we developed an experimental and computational framework for describing how sensory representations change during auditory perceptual learning. Mice were progressively trained to classify frequencies as a single, center frequency (ranging from 11-16 kHz) or non-center by licking left and right, respectively, for a water reward. Discrimination between center and non-center frequencies improved over 10-45 days through multiple phases of learning (N=72 animals). In parallel, in a subset of animals, we recorded from a population of layer 2/3 excitatory neurons in auditory cortex throughout learning using two-photon imaging (N=14 mice). Despite similar behavioral performance at the end of training, animals exhibited one of two distinct activity profiles in auditory cortex. Specifically, tuning profiles of excitatory neurons exhibited either a relative enhancement or a suppression of responses at frequencies reported as the center frequency (N=14 animals). The response profiles emerged throughout learning, starting in the first phase of learning. Additionally, they were present while the animal was engaged in the behavior, but not when the animal was passively listening to the same set of stimuli. To make sense of the across animal variability in tuning, we developed a computational model to explore whether animal-specific choice preferences seen during learning could explain this individual variability in neural tuning. We trained a model neural network using reward-dependent Hebbian learning (Williams, 1992) to perform the task, and examined whether initial choice preferences (rates of licking right and left), and the resulting reward statistics, are related

to the learned neural representations. We found that higher rates of reward in trials with non-center frequencies early in learning lead to larger magnitude responses to the center frequency, a relationship which was confirmed in the data. Overall, our results suggest that, through its effects on reward statistics and consequent synaptic plasticity, choice preference during early auditory perceptual learning may play a causal role in producing across-animal variability in learned representations.

Disclosures: K.A. Martin: None. **C.J. Bredenberg:** None. **J. Lei:** None. **E.P. Simoncelli:** None. **C. Savin:** None. **R.C. Froemke:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.13

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant R01DC018621

Title: Mechanisms of rapid intrinsic plasticity in the zebra finch auditory cortex

Authors: *Y. LU[1], F. SCIACCOTTA[1], C. D. MELIZA[1,2];
[1]Dept. of Psychology, [2]Neurosci. Grad. Program, Univ. of Virginia, Charlottesville, VA

Abstract: Decoding acoustic signals for communication is a hallmark function of the auditory system in songbirds and humans alike. In songbirds, early acoustic experience has a profound impact on auditory perception and vocal learning, making them a powerful model to study the underlying neural mechanisms of experience-dependent development. Although information processing and storage in the brain is thought to be primarily orchestrated by synaptic plasticity, intrinsic plasticity—changes in voltage-gated channels and their dynamics—also contributes. In the zebra finch caudal mesopallium (CM), a cortical-level auditory area implicated in discriminating and learning species-specific vocalizations, we previously observed that auditory experience modulates intrinsic excitability along with expression of Kv1.1, a low-threshold potassium channel. Specifically, neurons in birds reared in a complex acoustic environment are more likely to be phasic, a dynamical behavior that can enhance the reliability and selectivity of neural responses to complex acoustic stimuli. Here, we investigated the cellular mechanism of this plasticity using whole-cell recording techniques. We found that Kv1.1 is necessary but not sufficient for phasic spiking, indicating that some neurons express Kv1.1 but do not localize it to the plasma membrane. In a subset of non-phasic (tonic) neurons, spiking became phasic after a few minutes of stimulation with depolarizing step currents. This rapid intrinsic plasticity was attenuated by blocking intracellular calcium, and it did not occur in quiet-reared birds, who have much lower expression of Kv1.1. These results suggest that exposure to a complex acoustic environment causes the accumulation of a ready-to-use pool of Kv1.1 that can be rapidly

mobilized to the membrane to dynamically regulate intrinsic excitability during the peak of the critical period for song memorization.

Disclosures: Y. Lu: None. **F. Sciaccotta:** None. **C.D. Meliza:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.14

Topic: D.05. Auditory & Vestibular Systems

Support: Wellcome Trust Principle Research Fellowship WT108369/Z/2015/Z

Title: The influence of sound statistics on auditory decisions in ferrets

Authors: *K. S. BOCHTLER[1], F. DICK[2], L. L. HOLT[4,3], A. J. KING[1], K. M. M. WALKER[1,5]; [1]Physiology, Anat. and Genet., Univ. of Oxford, OXFORD, United Kingdom; [2]Psychological Sci., [3]Birbeck, Univ. of London, London, United Kingdom; [4]Psychology, Neurosci. Inst., [5]Carnegie Melon Univ., Pittsburgh, PA

Abstract: The ability to direct our attention towards a single sound source such as a friend's voice in a crowded room is necessary in our acoustical world. This process is thought to rely, in part, on directing attention to different sound dimensions, such as frequency. Previous investigations have shown task-dependent changes in the frequency tuning of auditory cortical neurons when ferrets actively detect or discriminate a particular frequency of sound (e.g. Fritz et al. 2010). However, questions remain about how attentional gain can arise based on sound statistics. Specifically, to what extent can this modulation occur even if frequency is not a necessary component of the task demands? Mondor & Bregman (1994) demonstrated that human listeners' reaction times on a tone duration task were slower when the presented tone frequency was unexpected (i.e. low probability). Here, we test the hypothesis that the statistical likelihood of sound frequencies alone can also affect animals' behavioural decisions on orthogonal dimensions of sounds. We trained ferrets on a 2-alternative forced choice tone duration discrimination task in which we manipulated the statistical likelihood of tone frequencies. Our results show that, similar to humans, ferrets' reaction times on this duration judgement task increased for low-probability frequencies, while their accuracy remained stable across other frequencies. These results suggest that attentional filters are employed during listening, even for an acoustical dimension (frequency) that is orthogonal to the task demands (duration). Our future experiments will use this task in combination with microelectrode recordings to investigate the neurophysiological basis of statistical-based attentional filtering in the auditory cortex.

Disclosures: K.S. Bochtler: None. **F. Dick:** None. **L.L. Holt:** None. **A.J. King:** None. **K.M.M. Walker:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.15

Topic: D.05. Auditory & Vestibular Systems

Title: Speech motor representation in improving the perception of spectrally degraded speech

Authors: *S. A. MURAI[1,2], H. NAGAMURA[1], K. I. KOBAYASHI[1], H. RIQUIMAROUX[3]; [1]Grad. Sch. of Life and Med. Sci., Doshisha Univ., Kyoto, Japan; [2]Intl. Res. Ctr. for Neurointelligence (IRCN), Univ. of Tokyo, Tokyo, Japan; [3]The Inst. for Res. in Human Informatics, Tohoku Gakuin Univ., Sendai, Japan

Abstract: When listeners are trained with acoustically degraded speech forms, such as inputs from cochlear implants, their perceptual performance of the speech can be improved and maintained for long periods. Some studies have suggested that improvements in speech perception for novel speech forms are associated with plastic changes in speech motor regions, such as the premotor cortex (PM). However, little is known about how the motor regions represent speech motor information (i.e., articulation) during the perceptual learning of degraded speech. To elucidate the representational nature of the speech motor information in retuning the speech perceptual system, we conducted perceptual training of degraded speech sounds and measured the brain activity with the functional magnetic resonance imaging (fMRI). This experiment revealed how speech motor information is encoded and changes across the learning process in the frontotemporal speech network, including the motor regions. Participants were trained to noise-vocoded speech sounds, which are a cochlear implant stimulation with reduced spectral detail and low intelligibility. To focus on the sensorimotor processing of speech among multiple linguistic processing, we used the pre-lexical sounds like consonant-vowel monosyllables (e.g., /ko/, /to/, /po/) rather than words or sentences. Under the fMRI scanning, participants were trained only on some monosyllables that have differences in a motor gesture (i.e., place of articulation), not the other monosyllables. We found improvements in the behavioral performance of speech recognition after short-term training, particularly for trained monosyllables. Furthermore, the representational similarity analysis of the fMRI data during listening to degraded speech revealed that place of articulation information was increasingly represented in the motor speech regions, including the PM and the supplementary motor area. These results provide the first evidence that perceptual training changes neural representations of speech motor information during degraded speech perception and highlight the engagement of the sensorimotor processing as a facilitatory role in perceptual learning under difficult listening conditions.

Disclosures: S.A. Murai: None. **H. Nagamura:** None. **K.I. Kobayashi:** None. **H. Riquimaroux:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.16

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant MH120117

Title: Decoding tone frequency from echoic memory in primary auditory cortex of the Macaque

Authors: *J. ORCZYK, T. TEICHERT;
Psychiatry, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Echoic memory (EM) is a pre-categorical form of auditory short-term memory, important for the temporal integration of complex sounds such as conspecific vocalizations. Yet, the neural substrate of EM is unclear. The most promising hypothesis holds that EM stores information about past sounds in the absence of delay-period activity in the form of a 'negative trace' of depleted vesicles. However, it remains unclear if and how the information can be read out of such an activity silent state. Here we test if an activity silent negative trace resides in A1, and whether it can be reactivated by the subsequent presentation of a non-informative sound. To test these hypotheses, we performed single-trial decoding of sound-evoked local field potentials and multi-unit activity that were recorded from a semi-chronically implanted 96-channel electrode array, which covered the entire tonotopic map in primary auditory cortex in one macaque monkey. Each trial consisted of one of 21 pure tone pips of different frequency, spanning about 7.5 octaves, and followed by an identical white noise burst 350-450 ms after the tone. To validate the approach, we first decoded tone identity from the tone-evoked responses themselves using a support vector machine classifier with a linear kernel applied to data in a 10-ms sliding window. As expected, decoding accuracy was high, reaching an accuracy of over 35% correct responses in the time window 25-35 ms after tone onset. A second period of heightened decoding accuracy was observed at 180-300 ms after stimulus onset, peaking at about 13% at 215-225 ms. Decoding accuracy increased to over 50% (M = 53.4%, SD = 7.7%) when combining activity from multiple time windows, and almost 90% of trials were classified within ±0.75 octaves of the correct tone. We then tested if it is possible to decode the identity of the previous tone using responses evoked by the subsequent white noise burst. A similar bi-phasic pattern of decoding also emerged. However, while still significantly above chance, decoding was (i) overall much lower, and (ii) relatively stronger in the later (6.4%) compared to the earlier period (5.2%). When combining multiple time windows, decoding accuracy further improved to 7% (M=7.0 %, SD = 0.8%), significantly greater than shuffled controls (M = 4.4%, SD = 0.5%), as determined by a one-tailed paired t-test, $t(15) = 8.98$, $p < 0.01$ This finding suggests that information in A1 about prior stimuli persists in an activity silent state that can readily be reactivated by a subsequent non-informative broad-band stimulus. We speculate that EM may be activated in a similar manner by non-informative top-down activation of A1.

Disclosures: J. Orczyk: None. **T. Teichert:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.17

Topic: D.05. Auditory & Vestibular Systems

Support: NIH Grant DC017163

Title: Auditory discrimination learning in animals with and without developmental hearing loss is accompanied by behaviorally relevant changes to striatal auditory and motor neural activity

Authors: *J. D. GAY, T. M. MOWERY;
Dept. of Otolaryngology, Rutgers Univ., New Brunswick, NJ

Abstract: Background: The way in which naïve and well-trained animals perform in the same task is dependent on differences in the evoked sensory and motor neural response to behaviorally relevant stimuli. Through many decades of research, we know that developmental hearing loss leads to permanent differences in the physiology of the auditory neuraxis. Recent work from my lab has shown that these changes extend to the auditory striatum, and that hearing loss induced deficits briefly compensate during learning. In this study, we asked how neuronal response properties in the motor and auditory regions of the dorsal striatum change during the learning of an auditory associative conditioning task as a function of developmental hearing status.**Methods:** Adult Mongolian gerbils were implanted with a 64-channel electrode array that spanned the sensorimotor and auditory/visual regions of the striatum. Each animal was trained on an amplitude-modulation (AM) rate discrimination task using an appetitive reinforcement operant conditioning procedure. The freely moving animals were trained to initiate each trial by a nose poke, and an AM stimulus that indicated the availability of a food pellet (Go stimulus), versus an AM stimulus which signaled the absence of a reward (Nogo stimulus). Task difficulty is increased by moving modulation rates closer together. A sensitivity measure, d-prime (d' = z(hit rate) - z(false alarm rate)), was computed for each animal across training days. Cells were spike sorted offline (Plexon) and analyzed with Neuroexplorer and JMP software. **Results:** Putative single-unit recordings from medium spiny neurons were carried out for each day of training and compared to behavioral state across trial type (Go, NoGo), response (Hit, False Alarm, Correct Rejection), learning state (naïve, acquisition, mastery), and striatal region (motor, auditory). Significant changes within groups were compared between animals with and without a history of developmental hearing loss during the physiological emergence of the novel behavior (NoGo-CR, repoke) that is associated with increased d-prime and learning. **Conclusions**: These findings provide exciting correlations between epochs of learning and the neurophysiological processes from which novel behaviors progressively emerge. Our data show how the physiology before and after learning is significantly altered through sensory motor integration at the level of the striatum. Further, it provides novel insight into how hearing loss induced changes to underlying neurophysiology can simultaneously lead to both propagation of and compensation for learning impairments.

Disclosures: J.D. Gay: None. **T.M. Mowery:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.18

Topic: D.05. Auditory & Vestibular Systems

Support: NIDCD Grant DC-00046

Title: Task-dependent modulation of subcortical areas during auditory perceptual learning

Authors: *R. YING, D. STOLZBERG, M. CARAS;
Univ. of Maryland, College Park, MD

Abstract: Training can improve one's ability to discriminate among near-threshold stimuli, a process called perceptual learning. Understanding the neural basis of perceptual learning has translational implications for both nonclinical and clinical use. Previous research has shown training on a perceptual learning task strengthens task-dependent modulations of auditory cortical activity. However, it is unclear whether these changes emerge in the ascending auditory processing pathway and are inherited by the auditory cortex, or arise in the cortex de novo. To address this, we implanted Mongolian gerbils (Meriones unguiculatus) with chronic 64-channel microelectrode arrays in either the central nucleus of the inferior colliculus (CIC) or the ventral medial geniculate nucleus (vMGN). We recorded single- and multi-unit activity as animals trained and improved on an aversive go/no-go amplitude modulation (AM) detection task, and during passive exposure to the same AM sounds. Firing rates and cycle-by-cycle vector strengths (Yin et al., 2011) were calculated for individual neuronal responses to AM stimuli and transformed into the signal detection metric d'. Neural thresholds were obtained for each training day by fitting a logistic curve to d' values across AM depths and determining the depth at which d' = 1. As expected, neurons in the CIC encoded AM using a temporal strategy. Neural thresholds were similar during task and passive conditions, suggesting an absence of task-dependent modulation in the CIC. However, both task and passive neural thresholds improved, suggesting that the CIC does display learning-related plasticity independent of task engagement. In the vMGN, neurons used both temporal and rate strategies to encode AM. As in the CIC, neural thresholds recorded during task performance improved, suggesting that learning-based plasticity is also present in the vMGN. However, unlike in the CIC, rate-based neural thresholds in the vMGN were better during task performance compared to passive exposure, suggesting that the vMGN is subject to task-dependent modulation. Notably, the magnitude of task dependent modulation increased over the course of training, similar to what has been reported in the auditory cortex (Caras & Sanes, 2017). These findings suggest that training may improve neural sensitivity within the ascending auditory pathway, at or below the level of the auditory midbrain, and simultaneously strengthen non-sensory modulations of auditory thalamus. Our results contribute to a deeper understanding of the circuits supporting perceptual learning, and may ultimately inform strategies for improving sound perception in the hearing-impaired.

Disclosures: **R. Ying:** None. **D. Stolzberg:** None. **M. Caras:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.19

Topic: D.05. Auditory & Vestibular Systems

Support: BSF-NSF Grant 2016688, ANR Grant 17-NEUC-0005-01a France-Israel Center for Neural Computation
NSF grant IIS-1724421
NSF Next Generation Networks for Neuroscience Program (Award 2014217)
NIH grants U19NS112959 and P30AG068635

Title: The balance state explains the emergence and propagation of spatial heterogeneities in the ascending auditory pathway.

Authors: *A. BERROU[1], M. HARPAZ[1], M. JANKOWSKI[1], D. HANSEL[2], I. NELKEN[1];
[1]Edmond and Lily Safra Ctr. For Brain Sciences, Hebrew Univ. of Jerusalem, Jerusalem, Israel;
[2]Integrative Neurosci. & Cognition Center, CNRS- UMR8002, Paris, France

Abstract: The mammalian auditory pathway is tonotopically organized, so that nearby neurons tend to respond to similar best sound frequencies (BFs). However, recent results suggest that nearby neurons may also show quite different BFs.
We investigated experimentally the degree of disorder in the tone representation in three subsequent stations of the ascending auditory system: the inferior colliculus, medial geniculate body of the thalamus and auditory cortex in anesthetized rats, recording responses using Neuropixels electrodes and carefully histologically reconstructing each penetration. After removing the local BF changes induced by the tonotopic gradient and the inter-trial variability on the BF estimation, we found that spatial fluctuations in BF increase along the auditory pathway. More generally, we showed that the signal correlations between receptive fields to pure tone stimuli are locally higher and decrease more slowly with the distance in the inferior colliculus than in the thalamus and cortex.
To determine the mechanism of the progressive spatial mixing of BFs, we constructed a three-layer network model of the auditory pathway. Each layer consists of a recurrent local network comprising excitatory and inhibitory neurons. The feedforward as well as recurrent connectivities are random, with a distance-dependent probability of connection. We found that the increasing level of BF spatial heterogeneity can be accounted for only when the network operates in the “balanced regime” (van Vreeswijk C, Sompolinsky H. Chaos in neuronal networks with balanced excitatory and inhibitory activity. Science. 1996 Dec 6;274(5293):1724-6.)

Disclosures: **A. Berrou:** None. **M. Harpaz:** None. **M. Jankowski:** None. **D. Hansel:** None. **I. Nelken:** None.

Poster

544. Auditory: Learning and Adaptation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 544.20

Topic: D.05. Auditory & Vestibular Systems

Support: NIH R21DC018327
Klingenstein-Simons Fellowship Award in Neuroscience
Hearing Health Foundation Emerging Research Grant

Title: Cell-type specific contributions to the acquisition and performance of an auditory categorization task

Authors: *R. F. KRALL, M. P. ARNOLD, C. N. CHAMBERS, H. B. KING, H. J. MORFORD, J. M. WIEMANN, R. S. WILLIAMSON;
Otolaryngology, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Senses connect our brain with the environment, allowing us to perceive the world around us. Auditory information enters the brain via a feed forward hierarchal pathway that canonically terminates in the auditory cortex (ACtx). An open question is how this information is processed then routed brain-wide to influence behavior. Excitatory projection neurons in the ACtx are broadly comprised of three groups; intratelencephalic (IT), extratelencephalic (ET), and corticothalamic (CT). These distinct populations send vast projections where they target nodes of the ascending pathway as well as downstream regions classically associated with decision making, action, and reward. These organizational principles allow for the broadcasting of behaviorally-relevant information and the shaping of auditory representations across brain-wide neural networks. We hypothesize that ACtx projection neurons provide a critical link between auditory input and behavioral output, necessary for acquisition and performance of auditory-guided behaviors. To investigate this, we trained head-fixed mice to categorize noise based the rate of amplitude modulation (AM) and bilaterally silenced distinct neural populations during stimulus presentations using GtACR2 (on 20% of trials). To ensure that ACtx was necessary for successful performance of this task, we silenced all excitatory neurons *en masse* and found that inhibition biased decisions towards one spout, ultimately leading to a significant reduction in categorization accuracy. Unexpectedly, silencing of any of the 3 projection neuron classes had little effect on mice's ability to categorize the rate of AM noise, indicating that no single projection is necessary for task performance and suggesting that multiple projections may work synergistically. This apparent disconnect between cell-specific and global inhibition led us to examine other consequences of targeted inhibition across learning. We found that longitudinally inhibiting either ET or IT neurons led to a significant reduction in learning rate, evidenced by increased number of trials and sessions to achieve task proficiency. Furthermore,

ET and IT mice trained to expert level had lower accuracy and higher variability across sessions compared to wild-type controls. Using dynamic psychophysical modeling we were able to infer differential learning strategies based on the factors influencing choice for each projection neuron class. Our current efforts are focused on using the GLM-HMM framework to investigate dynamic switching between these distinct learning strategies to further probe the critical link between neocortical output and behavioral outcomes.

Disclosures: **R.F. Krall:** None. **M.P. Arnold:** None. **C.N. Chambers:** None. **H.B. King:** None. **H.J. Morford:** None. **J.M. Wiemann:** None. **R.S. Williamson:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.01

Topic: D.06. Vision

Support: NWO VICI grant 016.Vici.185.050 to S.O. Dumoulin

Title: Population receptive field models in MEG: mapping the temporal domain

Authors: *K. EICKHOFF[1,2,3], A. HILLEBRAND[4], M. C. DE JONG[1,2,5], S. O. DUMOULIN[1,2,3,6];
[1]Spinoza Ctr. for Neuroimaging, Amsterdam, Netherlands; [2]Computat. Cognitive Neurosci. & Neuroimaging, Netherlands Inst. for Neurosci., Amsterdam, Netherlands; [3]Vrije Univ. Amsterdam, Amsterdam, Netherlands; [4]Dept. of Clin. Neurophysiol. ad Magnetoencephalography Ctr., Amsterdam UMC location Vrije Univ. Amsterdam, Amsterdam, Netherlands; [5]Univ. of Amsterdam, Amsterdam, Netherlands; [6]Utrecht Univ., Utrecht, Netherlands

Abstract: Receptive fields are the building blocks of vision. They describe what part of the visual field is processed by groups of neurons (population receptive fields, pRFs), and can be estimated with functional Magnetic Resonance Imaging (fMRI). Surprisingly, pRFs are observed to be dynamic, i.e., they respond to different parts of the visual field at different times. To obtain a method to measure pRF dynamics in the healthy human brain on the neuronal timescale (ms), we combine pRF measured by fMRI with Magnetoencephalography (MEG), a neurophysiological technique with high temporal resolution. We replicate recent findings that pRF models estimated from fMRI predict MEG responses and adopt a new ERP design that informs us about the temporal dynamics of pRFs with millisecond resolution. We first estimate pRFs with fMRI using contrast-defined bar apertures that travel across the visual field. We then predict the MEG responses based on the pRF models using forward modeling. The predicted responses are compared to MEG signals that were recorded when showing a stimulus that closely resembled the fMRI design and a similar stimulus devised to generate evoked responses (ERP design). We show that pRF models are able to capture MEG responses (maximum R-squared =

71%), with best fits for occipital sensors. Perturbing pRF parameters (size and position) decreases the R-squared values, confirming that the fits are influenced by the pRF parameters. Predicting the MEG data from pRFs located in different visual regions suggests that pRFs located in V1 explain the MEG signal the most. Next, we will model the ERP responses with millisecond resolution. Taken together, the current results pave the way to study temporal dynamics of pRFs using MEG.

Disclosures: K. Eickhoff: None. **A. Hillebrand:** None. **M.C. de Jong:** None. **S.O. Dumoulin:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.02

Topic: D.06. Vision

Support: KNAW Onderzoeksfonds 2018

Title: Human population receptive field properties across cortical depth using ultrahigh spatiotemporal resolution fMRI

Authors: *J. HEIJ[1,2], L. RAIMONDO[1,2], J. SIERO[3], W. VAN DER ZWAAG[1,2], S. DUMOULIN[1,2,4,5], T. KNAPEN[1,2,4];
[1]Spinoza Ctr. for Neuroimaging, Amsterdam, Netherlands; [2]Computat. Cognitive Neurosci. and Neuroimaging, Netherlands Inst. for Neurosci., Amsterdam, Netherlands; [3]Radiology, Univ. Med. Ctr., Utrecht, Netherlands; [4]Exptl. and Applied Psychology, VU Univ., Amsterdam, Netherlands; [5]Exptl. Psychology, Utrecht Univ., Utrecht, Netherlands

Abstract: Laminar fMRI holds great promise for unraveling the computational interactions between feedforward and feedback information processing. Laminar fMRI is, however, hampered by the low-resolution relative to the cortical thickness, and that the most-used sequences suffer from large draining-vein biases. Here we use human line-scanning fMRI to mitigate these problems (Raimondo et al 2021), by sampling at both high spatial (200μm) and temporal (100ms) resolution. To position the line through the brain accurately, we devised a framework where we identified a location on a flat piece of cortex (to limit signals from layers mixing together) based on a separate session using a standard pRF-mapping experiment (Dumoulin et al 2008). The target location is chosen by both the desired functional and anatomical characteristics, to obtain a cortical location that represents para-foveal functional processing with minimal cortical folding. This prior knowledge uniquely allowed us to tailor the experimental design to this target location specifically. We performed pRFs experiments centered around the target location using contrast-defined stimuli optimized for the pRF location. Additionally, this setup allowed us to probe both pRF and hemodynamic properties across depth, which showed that pRF size and variance explained varied systematically across the cortical

surface. The results demonstrate that we can effectively wield line-scanning fMRI's spatial and temporal precision in human cognitive neuroscience experiments. Such a framework could bridge canonical fMRI experiments and more closely link to electrophysiological experiments, which in turn allows novel avenues for studying human physiology non-invasively.

Disclosures: **J. Heij:** None. **L. Raimondo:** None. **J. Siero:** None. **W. van der Zwaag:** None. **S. Dumoulin:** None. **T. Knapen:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.03

Topic: D.06. Vision

Support: BMF, Computational Life Sciences, project BINDA (031L0167)

Title: Oscillations emerge when neural population vectors lie at the edge of the neuronal manifold

Authors: *J. ESFANDIARI[1,2], C. URAN[1,2], A. PETER[1], G. SPYROPOULOS[1], M. VINCK[1,2]; [1]Ernst Strüngmann Inst. for Neurosci. in Cooperation with Max Planck Society, Frankfurt am Main, Germany; [2]Dept. of Neuroinformatics, Donders Ctr. for Neurosci., Nijmegen, Netherlands

Abstract: Neural stimulus responses represent a small subspace of all possible activity patterns. It has been shown that neural activity recorded during different tasks such as object recognition or movement lies on a low-dimensional manifold and can exhibit oscillatory behavior. Characterizing the structure of the response-subspace and its relation to oscillatory dynamics is crucial to decipher the information processing in neuronal networks. However, the role of these oscillatory dynamics in shaping or organizing neuronal manifolds remains unknown. Here, we recorded 64 channel multi-unit activity and local field potential (LFP) from two cortical visual areas (V1 and V4) of a single macaque monkey in a fixation task. Different types of images including gratings with different orientations, and natural scene images were presented as visual stimuli. Using nonlinear dimensionality reduction methods including t-distributed Stochastic Neighbor Embedding (t-SNE) and Uniform Manifold Approximation and Projection (UMAP), we clustered spiking responses to each stimulus in V1 and V4. Through the ventral pathway from V1 to V4, we observed that the neural manifold untangled in two main ways: the centers of the V4 clusters became more distant, and their diameters were smaller. Importantly, clusters corresponding to visual stimuli which induce gamma oscillations, such as gratings and a subset of natural scene images, lie on the edge of the neural manifold. We suggest that this extreme position is a result of oscillatory states bordering criticality and thus represent extremes of neural population coding and dynamics. Our work therefore makes a connection between encoding and dynamics in recurrent neural network.

Disclosures: **J. Esfandiari:** None. **C. Uran:** None. **A. Peter:** None. **G. Spyropoulos:** None. **M. Vinck:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.04

Topic: D.06. Vision

Title: Cell-type specific and state-dependent gamma-rhythmic synchronization between visual areas in macaques and mice

Authors: *G. SPYROPOULOS[1], M. SCHNEIDER[1], J. VAN KEMPEN[2], M. A. GIESELMANN[2], A. THIELE[2], M. A. VINCK[1];
[1]Ernst Struengmann Inst., Ernst Struengmann Inst., Frankfurt Am Main, Germany; [2]Newcastle Univ., Newcastle upon Tyne, United Kingdom

Abstract: Past experimental and theoretical work has posited that feedforward gamma rhythmic synchronization mediates communication between visual cortical areas and attentional selection. However, the physiological plausibility of this claim has been vigorously debated. In order to directly assess the proposed role of feedforward gamma rhythmic synchronization, we recorded LFPs and single unit activity in awake macaques and mice. In macaques, we simultaneously recorded activity in areas V1 and V4, while the subjects performed a visual attention task, whereas in mice, we recorded simultaneously from the LGN and areas V1 and V2, under conditions of visual stimulation. In both species, cortical activity was recorded in a laminar fashion. We observed that visual stimulation elicits robust inter-areal LFP-LFP phase locking in the gamma range, consistent with previous findings, but this phase locking was accompanied by very weak locking of spikes in the downstream area (e.g. V4) to LFPs in the upstream area (e.g. V1). Importantly, this weakly locked population of neurons predominantly comprised putative FS interneurons in feedforward input layers and not putative excitatory cells. Optogenetic tagging in mice revealed that the majority of gamma-locked FS interneurons in V1 expressed parvalbumin, whereas in V2, interareal gamma equally involved cells expressing either parvalbumin or somatostatin. Selective attention modulated V4 spiking similarly in, both, putative excitatory cells and FS interneurons, especially in superficial layers, but it increased the strength of inter-areal gamma locking only for FS interneurons. Lastly, population-based decoding analyses showed that the spiking of even individual neurons is more accurate at predicting the attentional state compared to population-wide locking of gamma LFPs. The accuracy of decoding based on spiking activity increased as a function of the number of simultaneously recorded neurons. Taken together, our findings cast serious doubt on the potential role of feedforward gamma synchronization in inter-areal communication and attentional selection, and instead suggest alternative roles such as normalization.

Disclosures: **G. Spyropoulos:** None. **M. Schneider:** None. **J. van Kempen:** None. **M.A. Gieselmann:** None. **A. Thiele:** None. **M.A. Vinck:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.05

Topic: D.06. Vision

Title: Compositionality of 3D orientation coding in the mouse visual cortex

Authors: *J. HOELLER, M. PACHITARIU, S. ROMANI;
Janelia Res. Campus, Howard Hughes Med. Inst., Ashburn, VA

Abstract: Different views of the same 3D visual scene are related by complex but predictable changes in luminance patterns at the level of the retina. To facilitate reasoning about visual scenes, e.g. planning spatial routes in an environment, the brain may convert these changes to simpler representations that are scene-independent. We hypothesize that the neural representations of 3D scenes have a property called “compositionality”; for example, the change in neural activity to a 3D rotation of a visual scene is the same as the change in neural activity to half the rotation plus the change to another half rotation. Here we study how the 3D orientation of a planar surface is encoded by populations of cortical neurons. Our hypothesis inspires a geometric model, in which we input the change in neural activity to a 3D rotation of a planar surface and then predict the change in neural activity to multiples of the rotation. To test our hypothesis, we imaged the activity of more than 60,000 neurons in the visual cortex while a mouse is passively viewing planar textures at different 3D orientations projected to a monitor. Even though our textures differ widely from each other (e.g. in spatial frequency, “natural-ness” etc.), we find that 3D orientation is responsible for a large fraction of the variance in neural activity. In a dimensionally reduced subspace of neural activity that contains most of the variance, 3D orientation can be decoded linearly within the range of values that we tested. Finally, the geometric model is consistent with our data across textures. This suggests that the encoding of 3D orientations of planar surfaces in the mouse visual cortex is compositional and texture independent. In conclusion, our findings provide a step towards understanding how visual information relevant for visual reasoning is encoded in a mouse brain.

Disclosures: **J. Hoeller:** None. **M. Pachitariu:** None. **S. Romani:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.06

Topic: D.06. Vision

Title: Contribution of different sources of variability changes along the mouse visual hierarchy

Authors: *X. JIA[1], R. IYER[2], D. DENMAN[3], J. SIEGLE[2], S. AKELLA[2], S. R. OLSEN[2], C. KOCH[2];
[1]Sch. of Life Sci., Tsinghua Univ., Beijing, China; [2]Allen Inst., Seattle, WA; [3]Dept. of Physiol. and Biophysics, Univ. of Colorado, Denver, CO

Abstract: Visually-guided behaviors require a stable representation of the world. However, neuronal responses to the same sensory stimulus are often variable. It is thus critical to understand what factors contribute to neuronal variability and how they change across different cortical areas to influence information representation and propagation. Previous work has proposed a range of statistical models to explain neuronal variability, ranging from simple regression to latent dynamics models. Here, we partitioned variability of individual neurons using a previously proposed modulated Poisson model, which assumes spikes are generated from a Poisson process whose rate is influenced by both sensory drive and a fluctuating gain. We simplified the model and evaluated the fraction of partitioned variability from independent Poisson process, stimulus drive and gain modulation in individual neurons along the mouse visual hierarchy. We find that the fraction of stimulus induced variance consistently decreases across the visual hierarchy, and its relative contribution depends on the stimulus complexity. The variance of stimulus independent gain, which reflects fluctuations of gain across time, gradually increases along the mouse visual hierarchy, consistent with the trend observed in non-human primates. The fraction of independent Poisson process doesn't show a clear trend across areas. Putative inhibitory neurons showed stronger gain modulation consistently across areas, while putative excitatory neurons were more strongly influenced by stimulus drive. Together, our work systematically partitioned variability across the mouse visual hierarchy and can potentially support future work for evaluating the influence of shared gain on information processing.

Disclosures: X. Jia: None. **R. Iyer:** None. **D. Denman:** None. **J. Siegle:** None. **S. Akella:** None. **S.R. Olsen:** None. **C. Koch:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.07

Topic: D.06. Vision

Support: ERC Starting Grant (SPATEMP, EU)
BMBF (Germany)

Title: Cell-type specific entrainment during rhythmic visual flicker stimulation

Authors: *M. SCHNEIDER, A. TZANOU, C. URAN, M. VINCK;
Ernst Strüngmann Inst., Frankfurt am Main, Germany

Abstract: Rhythmic synchronization has been proposed as a mechanism for communication between a sender and a receiver area. Synchronization between spikes is known to enhance the impact of spikes on post-synaptic targets, however, it is unknown whether this holds true for rhythmic visual drive in the narrow, high-frequency gamma-range (30-100 Hz). We hypothesized that due to the low-pass filtering properties of excitatory neurons, feedforward gamma-rhythmic inputs should preferentially activate fast-spiking interneurons in a receiving area. We tested our hypothesis by presenting visual flicker stimuli at different frequencies (ranging from 10-80 Hz) to head-fixed Ai32-Sst and Ai32-PV mice while simultaneously recording from LGN, V1 and the hippocampus, and complemented our recordings with detailed multi-compartmental models of different cell types in the mouse visual cortex. As predicted, we found that visual stimulation at faster frequencies preferentially entrained fast-spiking interneurons (identified through waveform characteristics) in V1. By contrast, slower visual flicker stimulation recruited both excitatory and inhibitory neurons in V1, which allows the imposed rhythm to propagate through the different layers of V1. We showed that 40 Hz visual rhythmic stimulation mostly drives PV, as well as a small fraction of Sst interneurons. If the frequency of the rhythmic visual stimulation increases into the gamma frequency range (30-100 Hz) the imposed visual rhythm doesn't propagate beyond the input layer of V1. In contrast to recent studies on the reduction of Alzheimer's disease-related amyloid plaques by 40 Hz flicker stimulation (Adaikkan et al. 2019), we find no evidence of entrainment of cells in higher brain regions such as the hippocampus. Lastly, we confirmed our empirical observations by synaptically stimulating computational models of PV and Pyramidal cells in the visual cortex of mice with inhomogeneous Poisson spike trains in the corresponding frequency ranges. In total, our results indicate that gamma-rhythmic synchronization is not a mechanism for inter-areal feedforward communication, but rather that it serves as a mechanism to suppress inter-areal communication via the recruitment of fast-spiking inhibitory interneurons in the receiver.

Disclosures: M. Schneider: None. **A. Tzanou:** None. **C. Uran:** None. **M. Vinck:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.08

Topic: D.06. Vision

Support: DIRP, NIMH, USA, ZIAMH002797
BRAIN initiative Grant U19 NS107464-01

Title: Highly variable, scale-free cortical network responses in the face of precise holographic single neuron perturbation

Authors: *T. RIBEIRO, A. VAKILI, S. PAJEVIC, D. PLENZ;
Section on Critical Brain Dynamics, Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: Trial-by-trial variability quantifies the highly variable spike count found in single neurons when sensory stimuli are presented repeatedly (Heggelund & Albus, 1978; Shadlen & Newsome, 1998; Tolhurst et al., 1983; Vogels et al., 1989). The variability is much higher than expected for simple Poisson processes, potentially involves many sources, and poses fundamental constraints to ideas on cortical coding. In this work, we studied the fundamental aspect of such network variability by employing holographic stimulation of single neurons. This will allow us to identify fundamental aspects of network response variability to well-controlled unitary perturbations such as single cell firing, which have recently become an essential tool in neuroscience. We co-expressed GCaMP7s and the opsin ChrimsonR in layer II/III pyramidal neurons of widefield-identified primary visual cortex (V1). The activity of 150 - 300 neurons was recorded using 2PI in a ~450 μm x 450 μm area of V1 (100 - 200 μm depth) at ~45 Hz framerate while holographically stimulating single pyramidal neurons expressing the opsin (100 ms; ~5 - 10 mW, ~100 - 150 trials; low-repetition Light Conversion laser) using a spatial light modulator (Meadowlark). Images were denoised using a machine-learning-based algorithm (DeepInterpolation). Denoised calcium traces were then deconvolved for spike extraction using MLspike. During stimulation of the target cell, a subset of non-stimulated cells responded significantly to the perturbation (> 92% baseline spike count). Laser-stimulated pyramidal cells (target cells) and non-target cells exhibited similar spontaneous (baseline) spiking variability that was well above Poisson expectation, which was maintained for evoked spike count variability. We hypothesized this response variability to be independent from the variable number of spikes induced by holographic stimulation in target cells. We separated target cell responses into low vs high spike count categories, which by definition reduces variability for each category. We show that non-target cells maintain high spike count variability for either category that is comparable to baseline. Size of stimulus-triggered neuronal avalanches distributed as a power law over 4 orders of magnitude. We relate this scale-invariance to unitary branching tree responses expected in cortical networks that display criticality. We conclude that the highly variable, yet scale-free activity in response to precise holographic single neuron perturbation is an intrinsic property of a critical cortical network and does not originate from the variability of holographic stimulation.

Disclosures: T. Ribeiro: None. **A. Vakili:** None. **S. Pajevic:** None. **D. Plenz:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.09

Topic: D.06. Vision

Support: BMF (Bundesministerium fuer Bildung und Forschung), Computational Life Sciences, project BINDA (031L0167)

Title: Multi-neuron temporal spiking patterns yield stable encoding of natural movies

Authors: *B. SOTOMAYOR[1,2], F. P. BATTAGLIA[3], M. VINCK[1,2];
[1]Ernst Struengmann Inst. For Neurosci. in Cooperation with Max Planck Society, Frankfurt am Main, Germany; [2]Dept. of Neuroinformatics, Donders Ctr. for Neurosci., Nijmegen, Netherlands; [3]Radboud Univ. Nijmegen, Donders Inst. for Brain, Cognition and Behaviour, Nijmegen, Netherlands

Abstract: The current dogma in neuroscience is that neurons primarily convey stimulus information through their firing rate. However, recent studies suggest a remarkable speed of sensory processing which may be incompatible with traditional rate-coding schemes, and it has been hypothesized that sensory encoding may rely on the spike timing relationships among neurons. To study multi-neuron spiking patterns, we developed SpikeShip, an unsupervised, linear, geometry-based dissimilarity measure that aligns spikes across pairs of epochs based on optimal transport cost. This method has linear computation cost and is sensitive to higher-order correlations in spike trains. We used both rate and timing codes to find clusters across N > 8000 neurons from six visual areas during natural video presentations in 32 mice. We split the video into 30 sub-videos of one second each as in previous studies, and compared information content in firing rate population codes and multi-neuron temporal spiking patterns. Using SpikeShip, we show that (1) multi-neuron temporal patterns convey substantially more information about natural movies than population firing rates; (2) multi-neuron temporal patterns show high reliability across presentations, in contrast to firing rate codes; (3) firing rate codes exhibit memory across frames, whereas temporal patterns form a discrete and discontinuous manifold separating different movie frames; (4) the advantage of temporal information becomes larger as the number of neurons grows. These findings reveal the importance of temporal spiking patterns in the encoding of natural visual inputs.

Disclosures: B. Sotomayor: None. **F.P. Battaglia:** None. **M. Vinck:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.10

Topic: D.06. Vision

Support: NIH Grant EY027023

Title: Understanding the geometry of neuronal population representations in macaque areas V1 and V2.

Authors: *A. PAVULURI, A. KOHN;
Dominick P. Purpura Dept. of Neurosci., Albert Einstein Col. of Med., Bronx, NY

Abstract: Cortical representations of visual information have usually been studied by estimating the selectivity of individual neurons in different areas. This approach has been most successful in early visual areas; in higher cortex, it has proved challenging to understand representations in this manner. A related but distinct approach is to consider the geometry of neuronal population representations. Though derived from single neuron selectivity, the geometric view can provide insight difficult to infer from measurements in individual neurons. For instance, the geometric view can reveal whether representations emphasize separability, by ensuring that populations responses to different images occupy a high dimensional space, or generalizability, by requiring responses occupy a low dimensional manifold with meaningful axes. We sought to understand differences in representations of cortical areas V1 and V2 of macaque monkeys. We used multiple Neuropixel probes to record responses of neuronal populations to a rich ensemble of naturalistic textures. V2 neurons have been shown to be selective for higher-order statistics present in textures, whereas V1 responses are determined by the spectral content of those images. Our stimulus ensemble included different textures and, for each texture, different samples, which were distinct images with identical texture statistics. We define the trial-averaged population responses to different samples of a particular texture as an 'object manifold'. We found that V2 population responses provided better discriminability between object manifolds of different textures than V1 populations did. Better performance was achieved by greater center-to-center distances between the object manifolds in population response space. To understand the geometry of the representations, we assessed how object manifolds were oriented relative to the axis of discriminability, by determining how shuffling the trial-averaged responses to different samples affected performance. Shuffling improved discriminability in both areas, but the effect was clearly stronger in V2. This result suggests that object manifolds for different textures are arranged more consistently in V2 than V1. We confirmed this by comparing the alignment of the principal components of object manifolds for different textures. This alignment was higher in V2 than V1. Our results reveal consistent differences in the geometry of V1 and V2 neuronal population responses to textures. V2 representations afford better discriminability, but also bear hallmarks of a low-dimensional representation of texture statistics that should allow better generalizability.

Disclosures: **A. Pavuluri:** None. **A. Kohn:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.11

Topic: D.06. Vision

Support: Fonds KNAW I 2020

Title: Predicting position changes in population receptive fields following simulated scotomas using a divisive normalisation model

Authors: *M. DAGHLIAN[1], M. BITTENCOURT[2], R. J. RENKEN[2], S. O. DUMOULIN[3], F. W. CORNELISSEN[2];
[1]Spinoza Ctr. for Neuroimaging, Amsterdam, Netherlands; [2]Univ. Med. Cntr Groningen, Groningen, Netherlands; [3]Spinoza Ctr. For Neuroimaging, Amsterdam, Netherlands

Abstract: Patients are often unaware of retinal lesions and do not perceive anything "missing" in the visual field, a phenomenon termed "filling-in". Retinal lesions are also accompanied by apparent remapping within the visual cortex; (population) receptive fields ((p)RF) around the lesion projection zone seem to shift locations. It has been suggested that remapping is driven by plasticity and represents the neural basis for perceptual filling-in. However, simulated scotomas in healthy participants also cause receptive field locations to remap. This implies that remapping is not necessarily driven by plasticity or related to perceptual filling-in. We propose that remapping occurs because introducing scotomas changes the responses of neurons nonlinearly. A divisive normalisation (p)RF model can account for a variety of nonlinearities including surround suppression, spatial compression, and spatial oversaturation. Our simulations show that this model can explain apparent (p)RF shifts, with scotomas changing the balance of excitation and inhibition shifting the response centre-of-mass of the (p)RF. Linear (p)RF models cannot capture these responses without shifting (p)RF locations, leading to apparent remapping. Here we test this account using ultra-high-field (7T) fMRI, standard retinotopic mapping stimuli (a drifting bar) and simulated scotomas. Two scotomas (small and large) are simulated with circular patches on the display which are set to mean luminance and will occlude the stimulus bar. The divisive normalisation model predicts a different pattern of shifts depending on the relative position and size of the scotomas and pRFs. This study in healthy controls helps determine the extent to which remapping can be explained by stimulus driven nonlinearities and provides a baseline for future studies on patients, allowing us to identify evidence for or against plasticity in the adult visual cortex.

Disclosures: M. Daghlian: None. **M. Bittencourt:** None. **R.J. Renken:** None. **S.O. Dumoulin:** None. **F.W. Cornelissen:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.12

Topic: D.06. Vision

Support: Simons Collaboration on the Global Brain 542999
NIH Grant EY028626

Title: Neuronal population signal flow between macaque visual cortical areas under different temporal contexts

Authors: *A. KRISHNA[1], B. M. YU[4,5], C. K. MACHENS[6], A. KOHN[1,2,3];
[1]Dominick Purpura Dept. of Neurosci., [2]Opthamology and Visual Sci. Dept., [3]Systems and Computat. Biol. Dept., Albert Einstein Col. of Med., Bronx, NY; [4]Electrical and Computer Engin. Dept., [5]Biomed. Engin. Dept., Carnegie Mellon Univ., Pittsburgh, PA; [6]Champalimaud Neurosci. programme, Champalimaud Fndn., Lisbon, Portugal

Abstract: The sensory systems of humans and other primates are comprised of discrete cortical areas which are arranged hierarchically, with extensive bottom-up (feedforward) and top-down (feedback) connections between areas. Whereas feedforward processing has been studied extensively, relatively little is known about the role feedback signaling plays in sensory processing. To assess feedforward and feedback signaling between cortical areas, we used multiple Neuropixel probes to record spiking activity from hundreds of neurons simultaneously, in macaque visual cortical areas V1 and V2. We then applied multivariate analyses to the measured neuronal population responses to track signal flow between areas. Inspired by predictive coding hypotheses, we tested whether the balance between feedforward and feedback signaling would be altered by stimulus 'expectation'. To define expectation, we used an adaptation paradigm that involved exposure to a drifting grating stimulus (adapter), followed by a stimulus (test) that had either the same orientation or was orthogonal.
Inter-areal (V1-V2) population interactions, estimated using canonical correlation analysis, differed when the test was matched or mismatched to the adapter. The difference in population correlations occurred first for positive time delays between V1 and V2, indicating altered feedforward interactions. This was followed by a change in correlation for negative time delays - indicative of altered feedback. The population activity patterns that were most correlated between V1-V2 were distinct for these different temporal contexts (i.e., responses to stimuli matched or unmatched to the adapter). We also observed diverse responses in single neurons in both areas, ranging from response suppression to facilitation following adaptation. There was no evident laminar clustering of these different response properties.
Our observations indicate that differences in temporal context result in altered inter-areal interactions and detectable changes in the balance of feedforward and feedback signaling between cortical areas.

Disclosures: A. Krishna: None. **B.M. Yu:** None. **C.K. Machens:** None. **A. Kohn:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.13

Topic: D.06. Vision

Support: Max Planck Society
DFG SFB870

Title: Orientation preference of V1 neurons in mice shows experience-dependent drift

Authors: *J. BAUER[1], U. LEWIN[1], T. ROSE[2], C. E. SCHOONOVER[3], A. J. P. FINK[3], T. BONHOEFFER[1], M. HÜBENER[1];
[1]Max Planck Inst. for Biol. Intelligence, Martinsried, Germany; [2]Circuit Mechanisms of Behavior, Univ. of Bonn Med. Ctr., Bonn, Germany; [3]Columbia Univ., New York, NY

Abstract: Recent studies have shown that responses to sensory stimuli in adult mouse visual cortex are less stable over time than previously thought. Chronic recordings of neuronal responses to complex stimuli such as natural movies and - to a lesser extent - drifting gratings have revealed slow correlation decay. This so-called representational drift has also been observed in higher cortical areas, for instance, in the representation of task variables in posterior parietal cortex, and has profound implications for the implementation of robust coding strategies by biological neural networks.
However, to date, it remains unknown which neuronal tuning features are susceptible to representational drift in the visual cortex. Using repeated 2-photon calcium imaging of V1 neurons in awake mice during presentation of drifting gratings, we show that the preferred orientation of individual neurons is not entirely stable. We observe a time-dependent drift in orientation preference of individual neurons across days to weeks.
Experience has been hypothesized to stabilize representations. To investigate the impact of continuous visual experience on neuronal tuning in V1, we manipulated the statistics of the visual environment using cylinder lens goggles mounted in front of the eyes for four weeks, thus limiting visual experience to a narrow range of orientations. We still found neurons tuned to all orientations after this intervention, but on average single-cell orientation preference drifted towards the experienced orientation. This indicates that continuous experience of visual features is not necessary for their representation in the visual cortex, but that the absence of specific features destabilizes their representation by influencing the direction of tuning drift.
Together, these results demonstrate that the preferred orientation of individual neurons undergoes limited representational drift. Importantly, we show that the statistics of the visual environment are a key determinant of the direction of this drift.

Disclosures: J. Bauer: None. **U. Lewin:** None. **T. Rose:** None. **C.E. Schoonover:** None. **A.J.P. Fink:** None. **T. Bonhoeffer:** None. **M. Hübener:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.14

Topic: D.06. Vision

Title: More complex and less clustered orientation tuning in macaque V4

Authors: *D. JIANG[1], X. ZHAO[1], S. ZHANG[1], T. WANG[1], R. JIANG[2], S. TANG[1,2,3], C. YU[1,4,3];

[1]Ctr. for Life Sci., [2]Sch. of Life Sci., [3]IDG-McGovern Inst. for Brain Res., [4]Sch. of Psychology and Cognitive Sci., Peking Univ., Beijing, China

Abstract: Although V4 neurons tend to have complex feature tuning properties, some of them are still tuned to stimulus orientation. Here we used single- and two-photon imaging to sample a large number of V4 neurons in awake, fixating macaques, in order to have a comprehensive description of their orientation tuning and functional organization properties. Using two-photon imaging to record neuronal responses to a drifting grating ($\sigma = 0.53^o$, contrast = 90%, SF = 1.6 cpd, speed = 2 cycle/s), we identified a total of 16,178 V4 neurons in fourteen 800-μm FOVs of five monkeys. Overall, about 21% of identified V4 neurons were orientation selective (Freidman test, $p < 0.01$), in contrast to around 90% in V1. A von Mises function was used to fit orientation tuning curves. Fitting results showed that about 61% of orientation-tuned V4 neurons had a single preferred orientation, while 34% had two peaks in their orientation tuning functions. The corresponding percentages are approximately 90% and 10% in V1. Unlike V1 neurons that are organized in orientation clusters and columns, V4 neurons showed no clear or weak orientation clustering. When much larger cortical surfaces of V4 were studied with single-photon imaging at a coarser resolution, the vector sums of responses to various orientations with individual pixels of the images tended to have very large circular variances (>0.85), indicating often similar responses to different orientations and thus very weak orientation clustering on a more global scale, consistent with two-photon imaging results. Twin-peak orientation-tuned V4 neurons may help V4 respond to two-dimensional stimulus patterns, such as cross patterns, T-junctions, angles, and curvatures, which are more complex than bars and edges that V1 neurons respond to. Meanwhile, neurons with a wide range of orientation tuning in the same place may facilitate the emergence of these complex responses involving multiple orientations.

Disclosures: D. Jiang: None. **X. Zhao:** None. **S. Zhang:** None. **T. Wang:** None. **R. Jiang:** None. **S. Tang:** None. **C. Yu:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.15

Topic: D.06. Vision

Support: Simons Collaboration on the Global Brain 542999

Title: Early and midlevel visual areas interact via distinct communication subspaces

Authors: *A. I. JASPER[1], A. XU[1], C. K. MACHENS[4], B. M. YU[5,6], A. KOHN[1,2,3];
[1]Dominick Purpura Dept. of Neurosci., [2]Ophthalmology and Visual Sci. Dept., [3]Systems and Computat. Biol. Dept., Albert Einstein Col. of Med., Bronx, NY; [4]Champalimaud Neurosci. Progamme, Champalimaud Fndn., Lisbon, Portugal; [5]Electrical and Computer Engin. Dept, [6]Biomed. Engin. Dept.,, Carnegie Mellon Univ., Pittsburgh, PA

Abstract: Most brain functions involve interactions between different brain areas. Recent work has shown that early visual areas V1 and V2 interact through a communication subspace. The communication subspace defines which population activity patterns are relayed between areas and which remain private within a source area. We sought to leverage this new framework to investigate how population activity propagates across multiple stages of early and midlevel visual cortex. Specifically, we aimed to test (1) whether a source area communicates to different target areas with distinct or similar communication subspaces; and (2) whether communication between layers of a cortical area also occurs through a communication subspace.
To this end, we recorded large neuronal populations using multiple Neuropixel probes, in areas V1, V2 and V3B of anesthetized macaque monkeys. Sampled populations consisted of up to 200 neurons in each area, distributed across cortical layers. Neurons in different cortical areas had partially overlapping spatial receptive fields. We analyzed the relationship of trial-to-trial fluctuations in population activity in V1, V2 and V3B and found activity in each area could be used to predict activity in the remaining areas. These predictions involved a low-dimensional mapping between source and target populations—evidence for communication subspaces between each pairing of areas. Within V1, interactions between different cortical layers also showed evidence of occurring through a communication subspace, but the dimensionality of those subspaces were generally higher than for inter-areal interactions. Next, we analyzed whether inter-areal subspaces to different downstream targets—for example V1 to V2 compared to V1 to V3B—were similar or distinct. We found subspaces to different downstream areas were overlapping but not identical.
Our results suggest that communication subspaces are used to relay activity both across cortical layers of a cortical area and between areas. Communication subspaces to different target areas are distinct, suggesting that different patterns of activity may be used to communicate selectively with different target networks.

Disclosures: **A.I. Jasper:** None. **A. Xu:** None. **C.K. Machens:** None. **B.M. Yu:** None. **A. Kohn:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.16

Topic: D.06. Vision

Support: NESTOR
INTENSE

Title: Semi-automatic recovery of visual maps from neural data via high-channel cortical implants

Authors: *A. LOZANO[1], M. LA GROUW[2], X. CHEN[3], B. LI[2], F. WANG[2], P. R. ROELFSEMA[2];

[1]Netherlands Inst. of Neurosci., Amsterdam, Netherlands; [2]Netherlands Inst. for Neurosci., Amsterdam, Netherlands; [3]Netherlands Inst. For Neurosci., Amsterdam Zuidoost, Netherlands

Abstract: Disclosures P. R. Roelfsema and Xing Chen are cofounders of the startup company Phosphoenix BV

Abstract

Electrical stimulation of the visual cortex via an electrode induces the perception of a dot of light, known as a 'phosphene.' Multiple phosphenes could be combined to create the perception of shape and the development of a high-channel-count visual cortical prostheses based on this principle could one day restore functional vision in blind people. However, researchers and clinicians face the challenging task of mapping the perceived location of each phosphene within the visual field. Previous methods for phosphene mapping required prosthesis user to manually report the location of the phosphene induced on each individual electrode, e.g. using a touchscreen, which requires tens of seconds per electrode. These methods become prohibitively time-consuming when carried out on hundreds to thousands of electrodes. Additionally, many blind people experience spontaneous visual phenomena that can interfere with perception of electrically induced phosphenes or have disorganized eye movements that make phosphene localization challenging. In this work, we developed an automated and scalable computational method* for phosphene mapping and tested it on a dataset comprising signals from several hundred electrodes in the visual cortex of monkeys. Our methodology allowed to reliably recover the relative locations of the receptive fields of neurons at up to ~800 electrodes using a few seconds ($t > 10$ s) of neuronal data from area V1, consistently yielding a low error (RMS ~3 mm), and high correlation (~0.9) against ground truth retinotopic mapping data in two monkeys. In addition, the method was robust in case of a non-continuous coverage of the cortical tissue with the brain implant and can be used to map electrodes that are 25 mm apart. We also examined which frequency bands yield information about the phosphene maps at global (inter-array) and local (intra-array) scale. We found that global information is prominent at lower LFP frequencies, and high LFP frequencies give insight in the local relative retinotopic positions. In conclusion, our results demonstrate a new method to recover reliable phosphene maps, with unprecedented scale, precision, and time efficiency. *our methodology is involved within a current patent application procedure and will be fully disclosed within the SfN poster publication.

Disclosures: **A. Lozano:** None. **M. La Grouw:** None. **X. Chen:** Other; Phosphoenix. **B. Li:** None. **F. Wang:** None. **P.R. Roelfsema:** Other; Phosphoenix.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.17

Topic: D.06. Vision

Support: National Natural Science Foundation of China Grant U20A20221, 81961128029, 32100802
National Key R&D Program of China Grant 2018YFA0701400
Key Research and Development Program of Zhejiang Province Grant 2020C03004
China Postdoctoral Science Foundation Grant 2020M681829

Title: Spatial frequency representation in V2 and V4 of macaque monkey

Authors: Y. ZHANG[1,2], K. SCHRIVER[1,2,3,4], ***J. HU**[1,2,3], A. ROE[1,2,3,4];
[1]Zhejiang Univ., Hangzhou, China; [2]Dept. of Neurosurg. of the Second Affiliated Hospital, Interdisciplinary Inst. of Neurosci. and Technology, Sch. of Med., Hangzhou, China; [3]MOE Frontier Sci. Ctr. for Brain Sci. and Brain-machine Integration, Sch. of Brain Sci. and Brain Med., Hangzhou, China; [4]Key Lab. of Biomed. Engin. of Ministry of Education, Col. of Biomed. Engin. and Instrument Sci., Hangzhou, China

Abstract: Spatial frequency representation in V2 and V4 of macaque monkey
Authors: Ying Zhang[1,3], Kenneth E. Schriver[1,2,3,*], Jia Ming Hu[1,2,3,*], Anna Wang Roe[1,2,3,*]
Affiliations:[1]Department of Neurosurgery of the Second Affiliated Hospital, Interdisciplinary Institute of Neuroscience and Technology, School of Medicine, Zhejiang University, Hangzhou, 310027, China.[2]MOE Frontier Science Center for Brain Science and Brain-machine Integration, School of Brain Science and Brain Medicine, Zhejiang University, Hangzhou, 310012, China.[3]Key Laboratory of Biomedical Engineering of Ministry of Education, College of Biomedical Engineering and Instrument Science, Zhejiang University, Hangzhou, 310027, China.*****Correspondence: annawang@zju.edu.cn; hujiaming@zju.edu.cn; kenschriver@zju.edu.cn
Abstract: Spatial frequency (SF) is an important attribute in the visual scene, however there remains many unsolved questions about how primate visual cortex codes this fundamental information. Many studies have examined SF preference in V1 but few have focused on the organization of SF in extrastriate areas. In particular, there is little known about the relationship between SF preference and the functional organizations in V2 and V4. Using intrinsic signal optical imaging, we find that V4 functional domains in the foveal region are tuned to a surprisingly broad range of SFs. In both V2 and V4, similar to V1, SF preference maps and orientation preference maps exhibit orthogonally aligned gradients. We also find that color domains in V2 and V4 avoid overlap with high SF domains, and, moreover, that, at least in V2, the periodicity of SF preference parallels the periodicity of functional stripe cycles; this resolves a long-standing controversy based on cytochrome oxidase (CO) stripes. These findings of the spatial arrangement of SF preference in V2 and V4 provide us new understanding about "hypercolumn" architecture and suggest SF is an inherent aspect of fundamental units of representation across early visual cortical areas.

Disclosures: Y. Zhang: None. **K. Schriver:** None. **J. Hu:** None. **A. Roe:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.18

Topic: D.06. Vision

Support: NIH grant ZIAMH002956
NIH BRAIN grant U19NS107464

Title: Filtering of extraneous input patterns by visual cortex measured with cell-specific holographic stimulation

Authors: P. K. LAFOSSE, Z. ZHOU, V. M. SCOTT, Y. DENG, *M. H. HISTED;
NIH / NIMH, Bethesda, MD

Abstract: Transforming patterns of inputs into output firing is a central aspect of neural computation, but how networks of many recurrently-connected cells impact these transformations remains challenging to study *in vivo*. One proposed mechanism is recurrent cortical networks can amplify desired inputs or filter out extraneous inputs, a computation that can benefit perceptual behavior by favoring certain stimuli. Here, we investigate whether mouse visual cortex (V1) amplifies or attenuates responses to visual input via an adjustment of the input-output relationship of its component neurons. Using holographic stimulation methods, we precisely target individual cells based on their visually-evoked activity, and directly provide a fixed optogenetic input. To test if the local network might change these neurons' input-output functions, we stimulated V1 pyramidal cells (n=342 cells; n=5 animals) both during and without visual stimulation to measure changes in optogenetically-evoked output. For moderately visually-driven cells ($\Delta F/F > 10$ % and < 80%; n=47 cells), we find no evidence for changes in input-output relationships (p=0.239, K-S two-sample test). However, visually-suppressed cells ($\Delta F/F < -10$%, n=48 cells) show significantly weaker outputs to fixed input when the visual stimulus is present compared to without ($p<1*10^{-5}$, K-S two-sample test). These results indicate V1 neurons are pulled into a sublinear input-output regime when suppressed by the network, but largely remain in a linear regime when driven. This supports the stabilized supralinear network model of cortical function (Rubin et al., 2015), specifically, suggesting that in vivo V1 transfer functions have a supralinear and then linear regime (as predicted by theory; Sanzeni et al., 2020; Miller and Troyer, 2001; Hansel and van Vreeswijk, 2002). Moreover, we show that during spontaneous activity neurons operate just above the supralinearity. In sum, our data suggest that due to the shape of individual cells' transfer functions, visual cortical neurons can filter selected input patterns — not by amplifying responses but by attenuating inputs unrelated to the current visual stimulus.

Disclosures: P.K. LaFosse: None. **Z. Zhou:** None. **V.M. Scott:** None. **Y. Deng:** None. **M.H. Histed:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.19

Topic: D.06. Vision

Support: r01gm13363

Title: Population spike-synchrony contributes to the spectral exponent of aperiodic neural activity

Authors: *M. PRESTON[1], T. FEI[2], B. VOYTEK[3];
[1]Neurosciences Grad. Program, Univ. of California San Diego, La Jolla, CA; [2]Univ. of California San Diego, La Jolla, CA; [3]Cognitive Sci., Univ. Of California, San Diego, La Jolla, CA

Abstract: It has long been appreciated that neural oscillations support memory and cognition; however, recent research highlights the significance of non-oscillatory, aperiodic brain activity in these processes. Aperiodic activity changes during development and aging, varies across cortical depth and between regions, and is dynamically modulated in a task-related manner. Despite the growing evidence for the functional significance of aperiodic activity, the neural mechanisms underlying this activity have not been fully characterized. A recent computational model links the local field potential (LFP) aperiodic exponent to the balance of excitation and inhibition (Gao, 2017). In this model, the LFP is simulated as the linear summation of postsynaptic currents, driven by an excitatory and an inhibitory population of Poisson spike trains. While neural firing is commonly modeled as a Poisson process, cortical firing can exhibit highly-correlated activity, especially in the absence of stimulation. In the present study, we extend this LFP model to investigate the relationship between population synchrony and aperiodic activity. We hypothesize that shifts in correlated spiking activity are reflected in the LFP spectral exponent. We first introduce temporally correlated spike trains into the LFP model and compare spectral signatures to those of the Poisson population. We show that correlated spiking activity is associated with a steepening of the LFP power spectrum i.e., an increase in the aperiodic exponent. Time-resolved spectral parameterization reveals that decreases in correlated spiking activity recapitulate event-related shifts in aperiodic activity, similar to those observed in response to visual stimulation. We follow up these findings by simulating surrogate LFPs using empirical spike trains recorded from the primary visual cortex of macaques (Kohn & Smith, 2016). These simulations reveal a negative correlation between population spiking variability and the LFP aperiodic exponent during spontaneous activity (linear regression: n=6; r=-0.874; p=0.023). Together, these findings suggest that stimulus-evoked shifts in global population synchrony may be an additional neural mechanism reflected in the spectral exponent.

Disclosures: **M. Preston:** None. **T. Fei:** None. **B. Voytek:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.20

Topic: D.06. Vision

Title: Stimulus-specific omission responses in mouse primary visual cortex

Authors: *S. MIRBAGHERI[1], L. P. JIANG[2], A. FISHER[2], R. P. N. RAO[2], N. A. STEINMETZ[1];
[1]Biol. Structure, [2]Computer Sci., Univ. of Washington, Seattle, WA

Abstract: Background:
Predictive coding theory argues that each sensory area computes the difference between the local representation and a prediction of that representation made by higher areas of the brain, i.e., a prediction error. Based on this idea, if a predicted stimulus is omitted, a rise in neural activity should be detected as an indication of prediction error. Such omission responses have been reported in multiple studies. However, omission responses could either reflect a general surprise signal without specific content or, in accordance with the predictive coding theory, could have content related to the expected stimulus. We show that omission responses arise after learning a visual stimulus sequence and contain stimulus-specific content.
Method:
We trained mice for five days by exposing them each day to 1000 trials of a sequence of 5 natural images, presented in the order ABBCC, where A, B, and C refer to three unique images. Each image appeared for 50 ms and was separated from the subsequent one by 100 ms of a gray screen. Animals were water restricted during the training, and each sequence was followed by a water reward 500 ms after the last image. A random inter-trial period separated each sequence. After training, we recorded the neural activity in the primary visual cortex of the mice using a 4-shank Neuropixels 2.0 probe. During the recording, the animal was exposed to 400 repeats of the learned sequence, ABBCC, randomly interleaved with 40 trials of other probe conditions, e.g. omitting different elements of the sequence.
Results:
We observed that omission responses are stronger following training and arise throughout the first session of exposure in untrained subjects. Moreover, this rise in neural activity was not observed when the sequence began with a novel stimulus. We additionally show that responses when B and C are omitted become more distinguishable in the trained animal compared to the naive animal using linear discriminant analysis. This result indicates that omission responses are stimulus-specific.
Conclusion:
We established a task paradigm and training regimen that allows us to study the content of omissions in learned sequences. Using this design, we have demonstrated that such responses have distinct content, providing the opportunity to investigate the source and computational mechanisms of predictive processing in the brain going forward.

Disclosures: S. Mirbagheri: None. **L.P. Jiang:** None. **A. Fisher:** None. **R.P.N. Rao:** None. **N.A. Steinmetz:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.21

Topic: D.06. Vision

Title: A two-photon imaging study on the roles of macaque V1 and V4 in texture-based figure-ground segregation

Authors: *X. ZHAO[1], X. DONG[1], S. TANG[2], C. YU[3];
[1]Acad. for Advanced Interdisciplinary Studies, AAIS, [2]Sch. of Life Sci., [3]Sch. of psychological and cognitive sciences, Peking university, Beijing, China

Abstract: Where and how the brain segregate texture-based figure and ground remains a challenge to the understanding of primate vision. Earlier evidence has been inconsistent on whether V1 neurons could signal texture-based figure from ground (Lamme, 1995), or simply detect local orientation discontinuities through surround modulation (Rossi et al., 2001). We studied this classical issue by recording V1 and V4 neuronal responses to texture-based figure-ground stimuli with two-photon calcium imaging (GCaMP5) in awake, fixating, macaques.A total of 2015 orientation-tuned V1 neurons in three macaques and 2820 orientation-tuned V4 neurons in another three macaques on the basis of calcium responses to a single oriented bar. The optimal V1 neuronal responses at the preferred orientation were suppressed by a figure-ground texture, which was a 4° x 4° figure consisting of iso-oriented short lines superimposed on a 32° x 32° background consisting of orthogonal lines (ground). The suppression was lessened when the figure-ground boundary, but not the figure, fell on the pRF, regardless of individual neuron's orientation preference. The boundary effect was consistent with Rossi et al. (2001) in that V1 neurons detect orientation discontinuities between figure and ground line elements, not the figure per se. No consistent figure-ground effects were not observed in individual V4 neurons. We then performed population coding analysis by training linear SVM to decode the figure-ground boundary and the figure using PCA-transformed neural responses in V1 and V4. Both V1 and V4 could decode the figure-ground boundary and the figure, reaching decoding accuracies above 85% with sufficient number of principal components. However, V1 neurons could detect the boundary with considerably higher efficiency than V4 neurons, requiring a few (<10 vs. 20-30) principal components. In contrast, V4 neurons are substantially more efficient than V1 to perform figure-ground segregation (10-20 vs. >30 PCs). Moreover, when linear SVM was trained with original neuronal responses, both V1 and V4 were found to assign more weights to neurons preferring boundary orientations for boundary detection and figure-ground segregation. Our results suggest that V1 and V4 neurons respond to different aspects of figure-ground stimuli formed by line textures. V4's roles in figure-ground segregation are mostly manifested through changes of response patterns that can be revealed with population decoding, rather than through changes of response amplitude.

Disclosures: X. Zhao: None. **X. Dong:** None. **S. Tang:** None. **C. Yu:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.22

Topic: D.06. Vision

Support: Howard Hughes Medical Institute

Title: Invariant texture recognition in mouse visual cortex

Authors: *F. DU[1,2], M. PACHITARIU[1], C. STRINGER[1];
[1]Janelia Res. Campus, Ashburn, VA; [2]The Solomon H. Snyder Dept. of Neurosci., The Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: Visual textures such as leaves or rocks patterns provide important information about the environment for mice in various visual tasks. For example, in object recognition and object segmentation, invariant texture recognition is required to identify the same class of texture regardless of rotation, scale, and viewpoint. To study the neural basis of such invariant texture recognition in mice, we recorded ~40,000 neurons simultaneously in the mouse visual cortex while presenting ~14,000 visual stimuli sampled from 32 texture classes via random rotation, scaling and cropping. The performance of a linear classifier trained on the neural data achieved a test accuracy of 81.67% ± 2.74% (stddev, n=4 recordings, chance=0.03), and also achieved a test accuracy of 61.59% ± 2.90% (stddev, n=3 recordings, chance=0.03) on the neural data in response to textures with normalized spatial frequency, suggesting a robust representation of texture class in the mouse visual cortex which requires features in addition to spatial frequency. Furthermore, the patterns of errors made by the classifier were highly-consistent across mice, and classification accuracy improved slightly in higher-order visual areas. We found that such computations were mainly supported by a subset of texture-coding neurons which formed 10% of the population and were spread throughout visual areas, with the performance of the classifier trained on the texture-coding neurons being similar to that of the entire population. Next, we compared the texture representation in the mouse visual cortex and in the convolutional neural network (CNN). We found that artificial neurons from a pre-trained AlexNet model performed well in the texture classification task, but their pattern of errors did not match the patterns from the neural data. To further explore the underlying computational operations leading to texture invariance in the neural data, we fit a CNN encoding model from textures directly to the responses of the coding neurons. The model explained some of the neural patterns in response to texture images, however, it still could not fully capture the pattern of errors in the mouse visual cortex during the texture classification task. In summary, visual textures are invariantly encoded in both the artificial neural network and the mouse visual cortex, but the texture representations appear substantially different between real and artificial neurons.

Disclosures: F. Du: None. **M. Pachitariu:** None. **C. Stringer:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.23

Topic: D.06. Vision

Title: Neuronal population activity in macaque V5/MT reflects and predicts visual perceptual strategy.

Authors: *C. GAILLARD[1], S. BEN HADJ HASSEN[1], A. J. PARKER[1,2], K. KRUG[1,2];
[1]Inst. of Biol., Otto-von-Guericke-University, Magdeburg, Germany; [2]Dept Physiology, Anat. & Genet., Univ. of Oxford, Oxford, United Kingdom

Abstract: Changes in cognitive state often influence the bias and accuracy of judgments during human decision-making. Here we probed the neural events associated with changes of state by recording from sensory neurons in the macaque visual area V5/MT, during the performance of a perceptual decision task. One example of cognitive state changes are switches in strategy when a previously rewarded choice influences a decision about the upcoming choice. These are manifest behaviorally as win-stay or win-switch strategies. Recording single and multi-unit neuronal activity as strategy changes reveals the neural events that underly integration of state changes with incoming visual information. A perceptually-rotating cylinder was formed from moving dots. A difference in binocular depth of dots forming the front and rear surfaces determines the direction of rotation of the cylinder, but otherwise rotation direction is ambiguous. V5/MT neurons sensitive to both motion direction and binocular depth have a clear preference for the direction of rotation of these cylinders. We measured the preference of V5/MT neurons for rotation direction and recorded these neurons in 3 macaque monkeys as they reported the rotation direction of the cylinder. We tested whether neural activity could predict subjects' cognitive strategy across consecutive choices, by examining cases where an unambiguous and rewarded trial T(N) is followed by an ambiguous trial T(N+1). Regularized Linear Decoding applied to single neuron recordings on trial T(N) successfully predicts the cognitive strategy adopted by the subjects on trial T(N+1) (59.6% correct), with changes identifiable within 200ms of stimulus onset. Changes of cognitive strategy also affect population measures of neuronal activity on the ambiguous trials T(N+1). Adoption of a win-switch strategy is associated with a 26% decrease in noise correlations of V5/MT recorded neurons on trial T(N+1). Consistent with the proposal that conjoint neuronal activity is affected by cognitive strategy, continuous wavelet analysis reveals different patterns of oscillatory signals in V5/MT activity depending on strategy. Specifically, win-switch strategy was associated with an enhanced oscillatory activity in the alpha (8-12 Hz) and low-beta (14-17 Hz) oscillatory activity compared to win-stay strategy, independently of subjects' previous choice. Overall, we demonstrate strong neuronal correlates of cognitive strategy implementation in the V5/MT neuronal population, thus providing promising insights into the neuronal mechanisms at the core of human perceptual decisions.

Disclosures: C. Gaillard: None. **S. Ben Hadj Hassen:** None. **A.J. Parker:** None. **K. Krug:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.24

Topic: D.06. Vision

Support: NIH DA036657

Title: Off-manifold coding in visual cortex revealed by sleep

Authors: *E. F. DE OLIVEIRA[1], S. KIM[1], T. QIU[1], A. PEYRACHE[3], R. BATISTA-BRITO[1], L. L. SJULSON[2,1];
[1]Dominick P. Purpura Dept. of Neurosci., [2]Neuroscience, Psychiatry, Albert Einstein Col. of Med., Bronx, NY; [3]Montreal Neurolog. Inst., McGill Univ., Montreal, QC, Canada

Abstract: Low-dimensional population dynamics and movement-related activity are found throughout the brain. However, primary visual cortex contains high-dimensional sensory representations, raising the question of how low-dimensional dynamics and high-dimensional representations coexist. Here we approached this question by analyzing neuronal activity during slow-wave sleep, which provided reliable estimates of low-dimensional, internally-generated manifold structure. We found that movements and visual scenes were both encoded in the on-manifold subspace, which accounts for more variance than chance during sleep, but visual scenes were also encoded in the off-manifold subspace, which accounts for less variance than chance during sleep. This off-manifold subspace contains sparse activity in the neurons with the strongest low-dimensional modulation by movement, which paradoxically prevents movement-evoked activity from interfering with high-dimensional stimulus representations. These results reveal an unexpected link between low-dimensional dynamics and sparse coding, suggesting that these phenomena play a role in creating and accessing an off-manifold coding space for high-dimensional representations.

Disclosures: E.F. de Oliveira: None. **S. Kim:** None. **T. Qiu:** None. **A. Peyrache:** None. **R. Batista-Brito:** None. **L.L. Sjulson:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.25

Topic: D.06. Vision

Support: Natural Sciences and Engineering Research Council Discovery Grant
NSERC Discovery Grant RGPIN-2019-06741

Title: Fast fMRI can dissociate entrained oscillatory neural activity to simultaneously presented visual stimuli

Authors: *R. W. RAFEH[1], L. E. MULLER[2], A. R. KHAN[3], M. MUR[4,5], T. W. SCHMITZ[6]; [1]Neurosci. Grad. Program, [2]Dept. of Mathematics, [3]Dept. of Med. Biophysics, [4]Dept. of Psychology, [5]Dept. of Computer Sci., Western Univ., London, ON, Canada; [6]Dept. of Physiol. and Pharmacol., Univ. of Western Ontario, London, ON, Canada

Abstract: Brain oscillations come about via the rhythmic firing of neural populations. Oscillations play a pivotal role in neural communication and cognition (Lewis et al., 2012; Fries, 2015). Owing to its high temporal resolution, electroencephalography (EEG) has been commonly employed to study oscillatory neural dynamics in humans. However, EEG is unable to spatially resolve oscillatory neural activity on a scale that allows for the examination of oscillatory dynamics in fine grained maps. Unlike EEG, functional magnetic resonance imaging (fMRI) allows the precise localization of neural population activity on the millimeter scale. Recent studies show that hemodynamic oscillations driven by an entrained neural signal can be captured using fMRI (Lewis et al., 2016, 2018). However, these studies only demonstrate the feasibility of fMRI entrainment paradigms using a single stimulus, limiting the potential applications of this technique. Here we provide preliminary evidence for the entrainment of the hemodynamic signal in the human visual cortex to multiple frequencies simultaneously.
In this high-field (7T) fMRI experiment, three participants detected target color changes in one visual field quadrant while two gratings were presented in the opposite quadrant. In alternating trials, these gratings either oscillated at 0.2 and 0.5 Hz, respectively (entrainment), or did not oscillate (control). Data were rapidly sampled (TR=225 ms) from a slab centered on the occipital lobe.
Consistent with prior work (Kay et al., 2013, 2015), population receptive field (pRF) mapping enabled the definition of the visuospatial preferences of individual voxels across the visual cortex. We selected voxels whose preferred visual location overlapped with the spatial location of the entraining grating stimuli. We observed that oscillatory dynamics in pRF isolated voxels were modulated at the frequencies of the entraining stimuli (0.2 Hz and 0.5 Hz) during the entrainment relative to the control condition.
These results open the door to a new class of fMRI experimental paradigms in which oscillatory dynamics can be measured in fine-grained cortical maps. Entrainment paradigms in combination combined with accelerated fMRI sequences may thus facilitate the examination of how internal cognitive states interact with entrained oscillatory dynamics in human sensory cortices.

Disclosures: R.W. Rafeh: None. **L.E. Muller:** None. **A.R. Khan:** None. **M. Mur:** None. **T.W. Schmitz:** None.

Poster

545. Population Dynamics in Visual Cortical Networks

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 545.26

Topic: D.06. Vision

Support: National Key R&D Program of China, Grant, 2018YFA0701400
National Natural Science Foundation of China, Grant, U20A20221
Key Research and Development Program of Zhejiang Province, Grant, 2020C03004

Title: Submillimeter mesoscale representation in macaque visual dorsal pathway revealed by 7 T fMRI

Authors: *J. WANG[1], X. DU[1], S. YAO[1,2], L. LI[1,2], A. PING[3], A. ROE[1,4];
[1]Dept. of Neurosurg. of the Second Affiliated Hospital, Interdisciplinary Inst. of Neurosci. and Technology, Sch. of Medicine, Zhejiang Univ., Hangzhou, China; [2]Key Lab. for Biomed. Engin. of Ministry of Education, Col. of Biomed. Engin. and Instrument Science, Zhejiang Univ., Hangzhou, China; [3]Dept. of Neurosurgery, The Second Affiliated Hosp. of Zhejiang Univ. Sch. of Med., Hangzhou, China; [4]Key Lab. for Biomed. Engin. of Ministry of Education, Zhejiang Univ., Hangzhou, China

Abstract: In the visual system, the cerebral cortex is organized into two distinct pathways: the ventral pathway (concerned with object identification) and the dorsal pathway (concerned with information for the purpose of self-motion). In contrast to the ventral pathway, little is known about mesoscale organization in the dorsal pathway. Here, we have applied ultra-high field 7T MRI in macaque monkeys to examine response to motion across a wide swath of the dorsal visual cortex. Using visual motion stimuli (8 Hz drift grating), we mapped BOLD activity by GE-EPI in anesthetized macaques scanned in 7T MRI at submillimeter resolution (0.6 mm) with a custom RF coil. We find: (1) BOLD fMRI at 7T is sufficiently stable, robust, and reproducible for detecting mesoscale functional domains in anesthetized monkeys. (2) Motion direction domains and systematic motion direction maps were observed across multiple areas in the dorsal pathway, including areas V2, V3a, MT, MST. (3) For the first time, orientation columns in MT/MST and V3a are detected at whole brain scale of macaque monkey. (4) In some areas, motion domains are systematically spatially interleaved with color domains. (5) A focal superior part of LIPd and LIPv is sensitive to visual sensory motion information. We show that using ultra-high field 7T MRI is capable of achieving largescale mapping of fine-scale functional domains, something difficult to achieve with 2-3 mm voxel resolution conventional fMRI. Our findings show, for the first time, that dorsal pathways (including area such as MT/MST, V3a and LIP) are comprised of functionally specific mesoscale domains, and suggest common processing architecture in ventral and dorsal information processing streams.

Disclosures: **J. Wang:** None. **X. Du:** None. **S. Yao:** None. **L. Li:** None. **A. Ping:** None. **A. Roe:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.01

Topic: D.06. Vision

Support: NEI 5R01EY022122-09
NIGMS 1T32GM132498

Title: Co-organization of responses to direction, speed and temporal frequency in ferret visual cortex

Authors: *V. SUÁREZ CASANOVA[1], N. LASKY-NIELSON[1], K. CHENG[1], R. RODRIGUEZ[1], J. TOUBOUL[2], J. RIBOT[3], S. D. VAN HOOSER[1];
[1]Biol., [2]Mathematics, Brandeis Univ., Waltham, MA; [3]Col. de France, Paris, France

Abstract: To perceive a moving object in an animal's visual environment, it is necessary for the visual system to interpret the direction and speed of the object as well as its spatial properties: orientation and spatial frequency. Decoding the spatial characteristics of a stimulus is well understood - tuning responses of the primary visual cortex (V1) for specific properties does not change when other parameters of stimulus are modulated. But how the visual cortex might encode the temporal properties - direction, temporal frequency (TF), and speed - is only partially understood. In particular, it has been reported that some neurons can reverse their preferred direction as TF is increased (Moore et al. 2004), and recent unpublished data from author Ribot shows that these changes also occur at the level of cortical maps. Therefore, a one-dimensional population read-out of stimulus direction is not possible for this population of cells . Here we report progress on the functional organization underlying temporal processing of both individual and neighboring neurons, namely the relationship between spatial frequency, TF and direction processing. For this purpose, we used in-vivo electrophysiology and two-photon calcium imaging in the ferret primary visual cortex and characterized temporal characteristics of neurons across layers. We hypothesized that the activity of V1 neurons contains the information necessary to decode stimulus direction, speed, and TF in a non-linear manner. We examined whether the columnar organization of the temporal tuning is maintained across visual cortical layers, or whether variations in temporal tuning characteristics emerge across the layers. Our preliminary results suggest that speed-sensitive cells exist in different layers of cortex but whether these cells exist within specific functional columns remains to be described. These results will elucidate the structure of the receptive field of these cells across layers and may provide answers to functional architecture underlying these properties and whether they share similar computing principles.

Disclosures: V. Suárez Casanova: None. **N. Lasky-Nielson:** None. **K. Cheng:** None. **R. Rodriguez:** None. **J. Touboul:** None. **J. Ribot:** None. **S.D. Van Hooser:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.02

Topic: D.06. Vision

Support: Mousetrap Fund, USC

Title: Double bouquet cells, by randomly targeting pyramidal neuron dendrites, may facilitate the learning of arbitrary nonlinear functions at the single neuron level

Authors: F. C. MEL DE FONTENAY[1], *B. W. MEL[2];
[2]Biomed. Engin., [1]USC, Los Angeles, CA

Abstract: The cerebral cortex consists of a single basic architecture that is largely conserved across cortical areas and species, and yet performs a bewildering variety of functions. What are the architectural biases and modifiable parameters of the cortical circuit that underlie this remarkable flexibility?
Our approach in this work has been to try to connect two types of information, about: (1) a specific area of cortex, including experimental and modeling data that tell us what cells and local circuits are capable of computing; and (2) a specific behaviorally-relevant task that certain cortical neurons are assumed to perform. We focus on an important natural vision problem - how to optimally combine color and luminance cues for object boundary detection. "Color-luminance" cells are common in V1 (Johnson et al. 2008; Li et al. 2015), but the particular mathematical function that these neurons use to combine their color (parvo) and luminance (magno) inputs is not known.
To directly study this cue-combination problem, we labeled ~25,000 image patches that contained natural object boundaries, and spanned the 2-D space of light-dark and red-green contrast levels. We then trained a conventional “deep network” (DN) with the human-labeled data using backpropagation, and visualized the 2-input color combination rule. The learned function had a peculiar nonlinear form, raising the question as to how the cortex might compute such a function.
A previous biophysical result, combined with an intriguing architectural feature of human and monkey (but not rodent) visual cortex could provide an answer: a quasi-regular lattice of double bouquet cell axon bundles courses vertically through the layers of visual cortex on ~30 um centers, making contact with pyramidal neuron (PN) basal and oblique dendrites at random locations (DeFelipe et al. 2006). Given that focal inhibition, depending on its location, can alter the threshold and/or gain of a dendrite's sigmoidal input-output curve (Jadi et al. 2012), this arrangement of inhibitory columns intersecting radially-oriented dendrites seems ideally suited to create a diverse set of nonlinear “basis functions” within each PN’s dendritic arbor. Modification of a single layer of excitatory weights onto a PN’s dendrites might then permit that cell to learn an arbitrary nonlinear (low-dimensional, monotonic) function of its inputs.
In support of this idea, we developed a simple 2-layer model of a PN whose dendrites receive random focal inhibition, and confirmed that only a single layer of modifiable synapses is needed to almost perfectly reproduce the nonlinear 2-input combination rule learned by the DN.

Disclosures: F.C. Mel de Fontenay: None. **B.W. Mel:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.03

Topic: D.06. Vision

Support: CIHR
NSERC

Title: Spike Mutual Information of Contrast Gain Control

Authors: *N. CORTES[1], L. LAPLANTE[2], L. IKAN[2], V. CHOUINARD[3], M. VANNI[2], C. F. CASANOVA[4];
[1]Univ. De Montreal, Montreal, QC, Canada; [3]École d'optométrie, [2]Univ. de Montréal, Montreal, QC, Canada; [4]Sch. of Optometry, Univ. de Montéal, Montreal, QC, Canada

Abstract: The contrast response function (CRF) characterizes the response of visual neurons to stimuli that vary in contrast. CRF can be modified mainly in two manners: contrast and response gain. While contrast gain changes the position of CRFs along the y-axis, as a linear change, response gain modifies the dynamic ranges of the CRF, changing the curve in a nonlinear manner. Both mechanisms have been postulated to drive (contrast gain) or modulate (response gain) neuronal responses. Little is known about the spike train mutual information that these two gain control mechanisms have when a visual neuron is stimulated with random contrast levels. We tested this assumption theoretically by measuring spike train mutual information for contrast and response gain processing. Neuronal responses were simulated with a mathematical spike model that integrates visual responses nonlinearly as an instantaneous rate, then used to generate time points with the inhomogeneous Poisson process. The visual stimulus consisted of parallel bars that randomly and dynamically vary in luminance. The results show that mutual information changes were low when the contrast curve's position was moved, simulating contrast gain-type responses (~20% variation). Conversely, the mutual information was modulated more than twice when the response to high visual contrasts was saturated, representing response gain changes. Such results may be consistent with the driver and modulatory framework of visual gain control. If driver-like mechanisms are associated with gain-contrast regulation, the messages encoded in the temporality of the spikes would change little to internal variations, keeping the message transmitted intact. On the other hand, in a response gain control, modulatory effects would regulate temporal attributes robustly to adjust visual information according to the intrinsic neural demands of the visual system. Preliminary experimental data in mice's visual cortex support our assumptions.

Disclosures: **N. Cortes:** None. **L. Laplante:** None. **L. Ikan:** None. **V. Chouinard:** None. **M. Vanni:** None. **C.F. Casanova:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.04

Topic: D.06. Vision

Support: NIH NEI R01 EY025219

Title: Evaluating the resolution of functional ultrasound for imaging orientation domains in ferret primary visual cortex

Authors: *W. HU[1], S. ZHU[2], M. DOYLEY[1], F. BRIGGS[2];
[1]Electrical and Computer Engin., Univ. of Rochester, Rochester, NY; [2]Dept. of Neurosci. and Brain and Cognitive Sci., Univ. of Rochester Sch. of Med., Rochester, NY

Abstract: Functional ultrasound imaging (fUS) emerged in 2011 [Macé et al, 2011] and has become an important tool for studying structure and function in the brain. Compared to traditional imaging methods like functional magnetic resonance imaging (fMRI), fUS provides high spatial and temporal resolution (<0.1mm, >10Hz) with good penetration depth (>15mm). However, most applications of fUS to date have identified large structures in the cortex (e.g., retinotopic maps) rather than functional organization at a finer scale. In this work, we test whether fUS can resolve orientation domains in primary visual cortex (V1) of ferrets, which have been well characterized previously using optical imaging and electrophysiological methods [Müller et al, 2000]. fUS was performed through a craniotomy, allowing imaging access to V1, using a Vantage 128 scanner (Verasonics, Inc., Kirkland, WA, USA) with an 18 MHz L22-14vX transducer. Responses to drifting vertical gratings placed within the field of view were acquired in a series of parallel para-sagittal imaging planes. We observed retinotopic organization of fUS-activated pixels as well as clusters of pixels that often appeared to form columns perpendicular to the pial surface. To measure the structure of these activated pixel clusters, we projected the activated pixels from individual clusters onto the pial surface across each para-sagittal imaged plane. Then we stacked the projection from each plane to form a 3D surface map where pixel position was quantified with a value representing the penetration depth from the pial surface into the cortical layers to the white matter. As the pixels per cluster went from superficial cortical layers to deep cortical layers, the number of pixels, or activation area, within each layer decreased. Thus, the activated orientation domains formed cones rather than columns. The distance between neighboring cones activated by the vertical grating at a depth corresponding to layer 4 was 0.62± 0.36 mm, which is quite consistent with the same measurement based on optical imaging data (0.70±0.10mm). These results demonstrate that fUS has the resolution required to image orientation domains in ferret V1.

Disclosures: W. Hu: None. **S. Zhu:** None. **M. Doyley:** None. **F. Briggs:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.05

Topic: D.06. Vision

Support: Max Planck Society

Title: Ocular dominance columns in mouse visual cortex

Authors: *P. M. GOLTSTEIN, D. LAUBENDER, T. BONHOEFFER, M. HÜBENER;
Max Planck Inst. for Biol. Intelligence, Martinsried, Germany

Abstract: The columnar organization of neuronal response properties is a fundamental feature of the neocortex. The primary visual cortex (V1) of higher mammals such as cats and monkeys shows columns for stimulus features like orientation preference and ocular dominance. Models suggest that, for instance V1's size relative to the retina, could be a key factor determining whether a columnar organization is established or not. Accordingly, in rodent V1, which is – in comparison to other animals – relatively small in relation to the retina, there appear to be no orientation columns as found in higher mammals. However, the situation is less clear for ocular dominance. So far, *in vivo* imaging studies in mice have not reported an obvious functional clustering for ocular dominance, while experiments employing activity mapping with immediate early genes revealed eye-specific patches in rat V1.
Here we use a mouse line with widespread expression of a genetically encoded calcium indicator (GCaMP6s.Niell) to test whether mice show a functional organization for ocular dominance. We performed wide field-of-view, cellular-resolution two-photon calcium imaging throughout cortical layers 2/3, 4 and 5 of binocular V1. In most animals, we observed a patchy organization of eye preference within layer 4, with clear clusters of neurons that preferentially responded to the ipsilateral eye, surrounded by a contiguous region of contra-eye dominance. In several mice, the clusters of ipsilateral preferring cells extended vertically from layer 4 into layer 5 and layer 2/3, thus forming ocular dominance columns. The degree of functional clustering and columnar organization varied considerably across mice, reminiscent of the variability observed in other animals, like squirrel monkeys. The observation of ocular dominance columns in the minute binocular visual cortex of mice sets a new boundary condition for computational models explaining the emergence of a columnar organization in the brain. Our finding could also help refining ideas on how ethological and evolutionary factors relate to the presence of ocular dominance columns.

Disclosures: P.M. Goltstein: None. **D. Laubender:** None. **T. Bonhoeffer:** None. **M. Hübener:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.06

Topic: D.06. Vision

Title: Differing proportions and laminar patterns of calbindin-, calretinin-, parvalbumin-immunoreactive neurons across early and mid-level visual cortical areas in macaque monkeys

Authors: *J. KRUEGER[1], C. C. PARK[2], A. A. DISNEY[1];
[1]Dept. of Neurobio., [2]Duke Univ., Durham, NC

Abstract: Calcium-binding protein (CBP) expression has been used as a tool to classify subtypes of interneurons. Identifying subpopulations, whether it is based on morphology or chemical marker, is clearly a crucial step in understanding how neuronal networks are built but utilizing CBP expression simply as a classification method may represent a missed opportunity for understanding circuit function. The three most studied CBPs - parvalbumin (PV), calbindin (CB), and calretinin (CR) - have very different properties as calcium buffers and, in the case of CB and CR, probably as sensors as well. These distinct functional properties may offer insight into functional differences between the cells by which they are expressed. We know from a set of nonhuman primate studies that these three CBPs have unique density profiles that vary between primary visual cortex (V1) where PV-immunoreactive (-ir) neurons are most abundant and the frontal cortex where CB-ir and CR-ir make up the majority. We also know that, unlike in rodents, these three CBPs are rarely co-expressed in primates, at least in the occipital lobe. What is not known is whether there is a gradual or an abrupt shift in the distribution profiles along the anterior-posterior axis. We sought to address this by quantifying PV-ir, CB-ir, and CR-ir neurons in six visual areas: V1, secondary visual cortex (V2), tertiary visual cortex (V3), visual regions V3a and V4, as well as in the medial temporal visual area (MT). Triple immunofluorescence from three macaques revealed that (1) PV-, CB-, and CR-ir neurons belong to separate subpopulations (i.e., no co-expression) in all cortical areas; (2) Proportionally, PV-ir neurons constitute the majority in all areas but decrease from ~70% of CBP-ir neurons in V1, V2, and V3 to 50% in V3a, V4, and MT. The proportion of CB-ir neurons remains relatively stable across all areas while CR-ir neurons increase to 25% in the same regions where PV declines; and (3) Despite overall proportions between PV-ir, CB-ir, and CR-ir neurons appearing similar in V1, V2, and V3 as well as in V3a, V4, and MT, respectively, individual profiles differ across laminae in each area, yielding a distinct motif for each cortical area. Given the impact calcium buffering can have on short term synaptic plasticity and dendritic integration, it seems likely that these anatomical differences have consequences for visual processing in each cortical area.

Disclosures: J. Krueger: None. **C.C. Park:** None. **A.A. Disney:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.07

Topic: D.06. Vision

Support: NIH R01EY028657
NIH Grant 5T32EY021462-09

Title: V1 noise correlations are invariant to changing noise at the periphery

Authors: *R. O'SHEA, N. J. PRIEBE, I. M. NAUHAUS, X. WEI;
Univ. of Texas at Austin, Austin, TX

Abstract: Sensory neurons show considerable variability in their response to repeated presentations of a stimulus. In particular, retinal ganglion cells (RGCs) show correlated response fluctuations for visual stimuli (noise correlations). The magnitude of these RGC noise correlations depends on the ambient light conditions: scotopic light levels induce larger noise correlations than photopic light levels (Ruda et al., Nat. Comm. 2020). The impact of these changes at the periphery on downstream sensory processing are yet to be determined. We examine the impact of changing light levels by recording from V1 populations under scotopic and photopic conditions in awake mice using calcium imaging and electrophysiology. Our imaging data shows that the pattern of noise correlations over cortical space in V1 in excitatory and inhibitory populations differs- with parvalbumin-positive interneurons (PV cells) showing larger and more spatially extensive noise correlations than their excitatory counterparts. Intriguingly, these patterns remain constant across levels of light adaptation, despite the known changes in afferent inputs. To understand our experimental observations, we constructed a two-layer neural network model of the thalamocortical circuit in which the V1 receptive fields and all V1 noise correlations emerge from pooling of units in input layer. We find that varying the number of effective inputs to each V1 cell significantly alters the comparative noise structures under different light conditions. In a regime of sparse thalamocortical convergence, there is an increase in the magnitude of V1 noise correlations under scotopic relative to photopic conditions. As the number of effective inputs to each V1 neuron increases, these differences in noise correlations across light levels in V1 are reduced, consistent with our experimental results. It may also be the case that the distinct correlations observed in PV cells play a role in maintaining an invariant cortical representation of the visual input as the structure of feedforward noise changes—a hypothesis we are currently investigating. Our results suggest that changes in noise correlations at the periphery do not necessarily affect the structure of noise downstream. We propose two potential mechanisms behind this finding based on dense thalamocortical excitation and cortical normalization via PV cell-mediated inhibition.

Disclosures: **R. O'Shea:** None. **N.J. Priebe:** None. **I.M. Nauhaus:** A. Employment/Salary (full or part-time):; Wispr AI. **X. Wei:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.08

Topic: D.06. Vision

Support: NSERC Discovery Grant RGPIN-2020-05105
NSERC Discovery Accelerator Supplement: RGPAS-2020-00031
Healthy Brains, Healthy Lives New Investigator Award 2b-NISU-8
CIFAR Canada AI Chair
CIFAR Learning in Machine and Brains Fellowship

Title: Neuronal optimal stimuli synthesized with deep learning reveal functional segregations in the mouse visual cortex

Authors: *D. LIN[1], R. DA SILVA[2], A. GHOSH[3], R. TONG[3], S. TRENHOLM[3], B. A. RICHARDS[4];
[1]McGill Univ. Integrated Program in Neurosci., McGill Univ. Integrated Program in Neurosci., Montreal, QC, Canada; [2]Montreal Neurolog. Inst., Montreal, QC, Canada; [4]Neurol. and Neurosurg., [3]McGill Univ., Montreal, QC, Canada

Abstract: In recent years, advances in deep learning have helped neuroscientists grain deeper understanding about the functional organization of the sensory cortex, especially the brain areas responsible for visual processing. Artificial neural networks (ANN) have been extensively used as models for the ventral stream of primate visual cortex due to the hierarchical correspondence between layers of ANN and brain areas along the ventral visual pathway. However, little is known about whether such an hierarchical organization exists among the visual areas in rodents, or whether rodent visual areas also correspond to different layers of ANNs. Here, we approach this question from a novel angle: we ask, what type of visual stimuli excites neurons in the mouse visual cortex most? And, are the most exciting visual stimuli different for different visual regions? To answer these questions, we recorded the responses of thousands of neurons from six different regions in the mouse visual cortex (V1, LM, AL, LI, POR, RL) to natural images, and trained convolutional neural networks to predict the responses from these neurons using the activities read out from different layers. We then synthesized the optimal stimuli for each neuron using gradient ascent and analyzed their spatial features. We found that, while the middle-to-late layers of VGG16 pretrained on object recognition predicted neuron responses best, there existed little evidence that suggested a hierarchical correspondence between layers of VGG16 and mouse visual areas. However, clustering analysis on the spectral characteristics of optimal stimuli suggested that mouse visual areas were functionally separated, and that the optimal stimuli for one region maximally drove the best model of that region as compared to other regions. Then, with a closed-loop paradigm, we experimentally confirmed in mice that (1) optimal stimuli indeed drove neurons better than natural stimuli and (2) optimal stimuli of one region maximally activated that region. Our results suggested that, unlike primates, mouse visual cortex may not possess a hierarchical organization amongst the visual regions; instead, these regions may be parallely organized but functionally separated based on their preferred sensory stimuli. Combining large-scale neurophysiological recordings with deep learning approaches, our study pushes the frontiers of the synergy between neuroscience and artificial intelligence.

Disclosures: D. Lin: None. **R. Da Silva:** None. **A. Ghosh:** None. **R. Tong:** None. **S. Trenholm:** None. **B.A. Richards:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.09

Topic: D.06. Vision

Support: Fond de recherche du Québec Health: 2022-2023 - BF2 - 312876
Interdisciplinary Center for Brain and Learning
The Natural Sciences and Engineering Research Council of CanadaThe Natural Sciences and Engineering Research Council of Canada

Title: Topographic integration of widefield patterned optogenetic stimulation in the mouse visual cortex

Authors: *I. DJEROUROU[1], E. MORGAN[2], V. CHOUINARD[2], V. DAIGNEAULT[2], M. PTITO[1], M. VANNI[1];
[1]École d'Optométrie, [2]Dept. de neurosciences, Univ. de Montréal, Montréal, QC, Canada

Abstract: Optogenetics could be a promising alternative to electrical stimulation in cortical visual neuroprostheses approaches. Already revolutionary in neuroscience, optogenetics allows a high spatial, temporal and neuronal specificity. Previous studies showed that optogenetics evoked functional visual perception in mice, using a neural-resolved holographic stimulation strategy. Although very effective for studying neural circuits, this strategy is restricted to a small region. A more widefield approach using spatially defined pattern could be used to cover the whole visual cortex. The aim of this study is to explore the feasibility to use widefield optogenetic stimulation to evoke functional vision in the mouse by characterizing with calcium imaging the integration of the optogenetic-triggered signal that should ideally replicate the one from natural visual stimulation. We used mice expressing the calcium indicator jrGECO1a in the excitatory neurons and the photosensitive ion channel ChR2. They were implanted with a head bar and a chronic imaging chamber covering the entire dorsal cortex including the entire visual cortex. We compared the calcium responses evoked by the visual stimulation of a rectangle in the visual field with the optogenetics stimulation of their cortical representation: On each mouse, we obtained the retinotopic mapping of the visual areas. The altitude and azimuth phase maps of V1 were extracted to generate photostimulation patterns scanning V1 and feed digital micromirrors device with a sequence of cortical stimulation. In parallel, the evoked calcium activity was measured, and sign maps were computed. Preliminary results suggest that most of the calcium activity observed stayed at the location of the optogenetic stimulation site. However, notable activity was observed at distance in the higher visual areas (HVA) using short laser pulses (pulse duration = 3ms, frequency = 10Hz, power = 10mW/mm^2). Unfortunately, this signal was not strong enough to observe the limits between V1 and HVA. This project exploring the basis of artificial perception using optogenetics could lead to a new method of cortical mapping using optogenetics without sensory stimulation.

Disclosures: I. Djerourou: None. **E. Morgan:** None. **V. Chouinard:** None. **V. Daigneault:** None. **M. Ptito:** None. **M. Vanni:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.10

Topic: D.06. Vision

Support: NIH R01. EY011488-23

Title: Recurrent interactions drive spatially organized stimulus selectivity in supra-granular layers of visual cortex before the developmental emergence of a columnar architecture

Authors: *A. A. LEMPEL, D. FITZPATRICK;
Max Planck Florida Inst., Jupiter, FL

Abstract: The cerebral cortex develops a model of the world by learning from sensory experience. The representation of edge orientation in the ferret's primary visual cortex (V1) offers a great model to study this as it matures with experience after eye-opening. Orientation preference in V1 is organized vertically, forming columns where neurons share stimulus preference across layers, and horizontally, forming a map where preference changes smoothly across columns. The mechanisms that coordinate tuning development across neurons to produce such organized representation remain unknown. Mature tuning derives from selective convergence of feedforward inputs from the lateral geniculate nucleus onto layer 4 (L4) neurons which then relay tuned signals to other layers within a column. Based on this, previous studies suggested that orientation maps could emerge from an initial organization of thalamic inputs to L4 before experience. This hypothesis then predicts that responses to oriented edges will show a columnar organization and some degree of tuning in L4 in naïve animals. Here, we combined electrophysiology and calcium imaging to examine the columnar organization of responses to oriented gratings in visually naïve and experienced animals. In naïve animals, responses in Layers 2/3 (L2/3) exhibited robust modular spatial patterns with high variability and weak, though significant, selectivity. Surprisingly, the responses of neurons in L4 of naïve animals were not selective and were not correlated with the modular patterns present in L2/3. This demonstrates that early orientation selectivity and modular responses are not driven by feedforward inputs to L4 neurons. Instead, they may emerge from recurrent interactions in L2/3. Consistent with this, responses in L2/3 of naïve animals exhibited selectivity only after long latencies, in sharp contrast to the short-latency selectivity seen in older, experienced animals. Next, we aimed to resolve whether L2/3 selectivity in naïve animals depends on local interactions within a module or interactions across modules. We combined whole-cell electrophysiology and optogenetics to measure response tuning while silencing activity locally within the module corresponding to the recorded neuron or on neighboring modules. Altogether, our data indicate that recurrent interactions across modules generate modular, tuned responses in L2/3 of naïve animals. Such modular, selective responses could serve as an initial scaffold for the experience-driven development of feedforward selectivity mechanisms that would increase response reliability and drive strong tuning with a columnar organization.

Disclosures: A.A. Lempel: None. **D. Fitzpatrick:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.11

Topic: D.06. Vision

Support: NIH R00 EY030840

Title: Stimulus-driven network complexity of neuronal activities

Authors: D. TANG[1,2], X. JIA[3,4], J. ZYLBERBERG[2], *H. CHOI[1];
[1]Georgia Inst. of Technol., Atlanta, GA; [2]York Univ., Toronto, ON, Canada; [3]Tsinghua Univ., Beijing, China; [4]Allen Inst., Seattle, WA

Abstract: Different visual inputs are dynamically routed even though the anatomical structure is relatively fixed. Elucidating the relationship between the dynamic functional network and the visual stimulus is critical to our understanding of visual processing. With a large Neuropixel dataset of mouse brain from the Allen Institute, we investigate the varying patterns of functional connectivity associated with different stimuli. We focus on neural activity in six visual cortical regions, recorded while mice observed stimuli of varying degrees of complexity: flashes, drifting gratings, static gratings, natural scenes and movies, and gray screen (approximation for spontaneous activity). From the responses to each stimulus, we evaluated the directed functional connectivity using spike cross-correlograms (CCG) between pairs of neurons. For significant causal connections, we examined 'sharp intervals' within a short positive latency by detecting extreme values that lie outside the confidence interval. A causal connection is defined as excitatory or inhibitory if its correlation strength has a positive or a negative sign respectively. Interestingly, we found that more natural and complex stimuli tend to evoke fewer functional connections among neurons, and their correlation strengths are among the weakest. On the other hand, flashes evoke the strongest causal connections, but its connectivity pattern on the regional level is incredibly close to resting-state, both with random and abundant between-region connections. All stimuli except for flashes, evoke more within-region connections, especially in primary visual area (V1). Both excitatory and inhibitory networks follow heavy-tailed in-degree and out-degree distributions but are not strictly power law, indicating that a few neurons acting as hubs in the functional network play a crucial part in stimulus-driven interactions. The density of inhibitory connections shows a distinct trend against stimulus from the excitatory network; however, their absolute connection strengths are correlated ($r=0.95$, $p=7.8\times10^{-29}$). Network transitivity (fraction of triangles) of excitatory links reaches the highest for both moving and static gratings, showing that neurons tend to interact in triplets given those stimuli. Thus, distinct visual stimulus types lead to qualitatively different topological properties of functional connectivity.

Disclosures: D. Tang: None. **X. Jia:** None. **J. Zylberberg:** None. **H. Choi:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.12

Topic: D.06. Vision

Support: Charles University Primus Research Program 20/MED/006

Title: Unified spiking model of structured activity and travelling sparse waves in the resting state of the primary visual cortex

Authors: T. RÓZSA, R. CAGNOL, *J. ANTOLIK;
Charles Univ., Prague, Czech Republic

Abstract: In the primary visual cortex (V1) of higher mammals, spontaneous activity is modular and reflects the underlying structure - co-active regions are highly correlated to orientation maps[1]. This activity travels in sparse waves, in line with the propagation speed of unmyelinated lateral connections [2]. These two intertwined phenomena have so far only been studied separately, both experimentally [1,4] and computationally [5,2]. Furthermore, there is a lack of theoretical and mechanistic understanding of how these two phenomena interact with evoked activity.
In this study we offer a unifying computational theory of structured spontaneous activity and traveling waves, by presenting a single large-scale spiking model of cat V1 that exhibits both phenomena. Our model exhibits spontaneous activity, travelling in sparse waves matching the conduction speed of V1 layer 2/3 horizontal connections, and is simultaneously correlated with the underlying orientation map. From the outset [3], our model demonstrates a wide variety of previously reported visually evoked properties, which shows the compatibility of the identified mechanisms of spontaneous travelling waves and structured activity with the mechanisms required by the evoked regime.
1. Smith, G.B. et al., M. Nat Neurosci 21, 1600-1608 (2018)
2. Davis, Z.W. et al. Nat Commun 12, (2021)
3. Antolík, J. et al., (2018) doi:10.1101/416156.
4. Muller, L. et al., Nat Commun 5, (2014).
5. Cai, D. et al., Proceedings of the National Academy of Sciences 102, 5868-5873 (2005)

Disclosures: T. Rózsa: None. **R. Cagnol:** None. **J. Antolik:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.13

Topic: D.06. Vision

Support: DARPA Grant HR001120C0021
Templeton World Charity Foundation
Weill Neurohub Investigator Grant
CJ and Elizabeth Hwang Professorship

Title: Dynamic and Active Predictive Coding: New Approaches to Understanding Cortical Function

Authors: *R. P. N. RAO, L. P. JIANG, D. C. GKLEZAKOS, A. FISHER, V. SATHISH; Univ. of Washington, Seattle, WA

Abstract: Background: The predictive coding model of Rao and Ballard ascribed the role of spatial predictions to cortical feedback connections but did not elaborate on how the cortex predicts future events at multiple time scales and how self-initiated actions influence predictions. Recent experiments have demonstrated both temporal response hierarchies and action-conditioned predictions in the cortex. A normative framework for understanding the neural and computational basis of these cortical properties has remained elusive. **Methods:** We first investigate a new predictive coding model called dynamic predictive coding that learns hierarchical temporal dynamics via hypernetworks. Hypernetworks allow higher level neurons to modulate the dynamics of lower-level neurons, enabling the learning of temporal response hierarchies. We then extend this model to incorporate actions, resulting in a "canonical cortical module" consisting of a state-prediction network and an action-prediction network which feed into each other. These modules can in turn be assembled to form a hierarchical network model of multiple interacting cortical areas. **Results:** We show that when exposed to natural videos, a dynamic predictive coding network learns separable and inseparable (direction-selective) space-time receptive fields similar to those found in primary visual cortex. Moreover, the same network also explains the phenomenon of activity recall in the visual cortex. We then show that active predictive coding learns hierarchical sensory-motor models of the world, with applications to tasks ranging from parts-based parsing of objects and scenes using eye movements to solving hierarchical reinforcement learning and planning problems. **Conclusion:** Our results suggest that the cortex learns a hierarchical generative model of the world, with each cortical area learning sensory-motor representations whose dynamics are modulated by feedback from higher levels. Such an architecture may form the basis for learning rich compositional representations that underlie the human ability to generalize quickly in novel situations and solve complex problems through hierarchical task decompositions.

Disclosures: R.P.N. Rao: None. **L.P. Jiang:** None. **D.C. Gklezakos:** None. **A. Fisher:** None. **V. Sathish:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.14

Topic: D.06. Vision

Support: U19-NS107464

Title: Sequence selectivity in visual cortex due to pattern-specific recurrent suppression

Authors: *C. DEVEAU[1,2], Z. ZHOU[1], P. K. LAFOSSE[1], Y. DENG[1], M. HISTED[1];
[1]Natl. Inst. of Mental Hlth., NIH, Bethesda, MD; [2]Neurosci. Grad. Program, Brown Univ., Providence, RI

Abstract: As organisms move through the world they must process sequences of sensory information across time. The order of sensory sequences is important: a movie or speech played forward and backward is perceived differently. Cerebral cortical circuits seem to have the capacity to process inputs that vary in time, given their strong recurrent connectivity and the capabilities of artificial recurrent networks (Sussillo and Abbott, 2009). However, how the cortex amplifies or transforms temporal patterns of input has been largely unknown, and particularly in the visual cortex, past work suggests only weak sensitivity to temporal input patterns. Here we examine visual cortical transformations of temporally-patterned inputs using two-photon holographic stimulation. We find that the mouse visual cortex is sensitive to the sequential order of small (20 cells or more) ensembles or patterns of neural activity, which change every 30 milliseconds. The size of responses to the patterns is dependent on the order of the stimulation sequence. For example, we find that when cells are stimulated first in the sequence their activation is on average 17.3 +/- 2.9% larger (N=4 animals, N=7 experiments) than when they are stimulated last in the sequence. To investigate the mechanism of this tuning for temporal sequences, we examine responses to the component patterns. Early patterns of activity produce patterned suppression in other cells, likely due to a recurrent excitatory-inhibitory network mechanism. When cells in later stimulation patterns are suppressed by activation of patterns early in the sequence, the responses of the later-stimulated cells are reduced. A linear summation model of this mechanism fits the data well. In summary, we find that a pattern of input across V1 neurons produces suppression in a subset of other selected neurons, allowing certain sequences of patterned activation to be amplified and others to be suppressed.

Disclosures: C. Deveau: None. **Z. Zhou:** None. **P.K. LaFosse:** None. **Y. Deng:** None. **M. Histed:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.15

Topic: D.06. Vision

Support: HHMI

Title: A better model for vision research: mapping the tree shrew visual system using functional ultrasound imaging.

Authors: *J. B. WEKSELBLATT[1], R. NAYAK[2], F. LANFRANCHI[2,3], F. LUONGO[1], D. A. WAGENAAR[1], M. G. SHAPIRO[1], D. Y. TSAO[3];
[1]Caltech, [2]Caltech, Pasadena, CA; [3]Berkeley, Berkeley, CA

Abstract: The choice of model species is an important consideration in biomedical studies, particularly when such models are intended to generate knowledge that will translate to humans. The challenge of finding adequate models for translational research is particularly acute in neuroscience. Existing animal models often present methodological challenges: in primates, which allow the probing of complex cognition, it is difficult to record from large ensembles of neurons or manipulate genetically defined cell populations; in rodents, rudimentary cortical organization and behavioral repertoire limits the modeling of visual processing, and acuity in rodent vision is ~2-3 orders of magnitude worse than humans. For these reasons, we chose to study visual organization in the tree shrew, an animal with high visual acuity and considerable cognitive abilities.

A variety of brain regions are active during sensory perception and behavior. A high-resolution, brain-wide activity map could identify brain regions involved in specific behaviors, which is especially useful when studying a new species. The macaque has classically provided a key model for visual processing, due to its amenability to behavioral tasks and well-delineated visual hierarchy. More recently, the rodent has emerged as a model for visual processing, due to the availability of molecular and genetic tools. An ideal model system would satisfy both needs: amenability to complex psychophysical tasks and tractability for molecularly-based recording and perturbation techniques.

Towards this goal, we have embarked on an effort to establish the tree shrew as a model organism for study of high-level vision and cognition. A diurnal animal, the tree shrew has a cone-dominant retina, and a columnarly-organized visual cortex. Due to its small size and relatively short reproductive and developmental cycles, the tree shrew offers experimental and genetic accessibility similar to rodents.

We have applied functional ultrasound imaging to record activity in the tree shrew brain at a resolution of ~100 um. This work identifies functional modules involved in the perception of various stimuli and reveals organizing principles of the tree shrew visual cortex. This technique provides an experimental approach to monitor whole brain activity in normal and disease states. We believe the tree shrew will serve as an important species for the study of visual perception and computation given its unique combination of experimental tractability and impressive visual capabilities. Moreover, we have advanced the techniques for awake, headfixed ultrasound imaging of whole-brain activity and describe surgical and technical advances.

Disclosures: J.B. Wekselblatt: None. **R. Nayak:** None. **F. Lanfranchi:** None. **F. Luongo:** None. **D.A. Wagenaar:** None. **M.G. Shapiro:** None. **D.Y. Tsao:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.16

Topic: D.06. Vision

Support: MVLS DTP Studentship 00544838
BBSRC BB/S006605/1
Bial Foundation Grants Programme Grant id: A-29315, number: 203/2020, grant edition: G-15516

Title: Opposing trend in population receptive field size across upper and lower visual fields in the superior colliculus and early visual cortex

Authors: *K. MORAVKOVA[1], A. FRACASSO[2];
[1]Univ. of Glasgow, Sch. of Psychology and Neurosci., Glasgow, United Kingdom; [2]Sch. of Psychology and Neuroscience, Univ. of Glasgow, Glasgow, United Kingdom

Abstract: In primate superior colliculus (SC), the sizes of receptive fields representing the upper visual field (UVF) are smaller than those of the lower visual field (LVF). This anisotropy is potentially driven by visual exploration of near and far regions and it has not yet been characterised in the human brain, due to the lack of spatial resolution of previously available non-invasive techniques. To investigate this anisotropy in the human visual cortex and subcortex, we analysed 1) Human Connectome Project (HCP) retinotopy dataset published by Benson et al., 2018 containing cortical and subcortical 7 Tesla (7T) fMRI data from 181 participants collected using standard retinotopic stimuli and 2) 7T data of 18 participants from early visual cortex collected at the University of Glasgow using the moving bar retinotopic paradigm. Both datasets were analysed using the population receptive field (pRF) modelling framework, a method conventionally used to characterise visually-evoked responses in the human brain. We quantified the differences in pRF size above and below the horizontal meridian for both datasets for the early visual areas V1, V2 and V3 (HCP and Glasgow data) and the superior colliculus (HCP data only). Compatible with findings from primates, we observed an increase in pRF size in the LVF compared to the UVF in the superior colliculus (Figure 1A). Moreover, the difference in pRF size became more prominent with greater distance from the fovea (Figure 1B). Interestingly, this trend is reversed in early visual cortical areas with pRFs being larger in the UVF than in the LVF. The observed effect was present in both analysed datasets (Figure 1C and D). The discrepancy between the two trends might hint towards different processing of visual information in the superior colliculus and the early visual cortex, prompting a debate about potential principles by which visual information is inherited from the subcortex to the cortex and whether known organisational principles remain the same or differ across the visual system hierarchy.

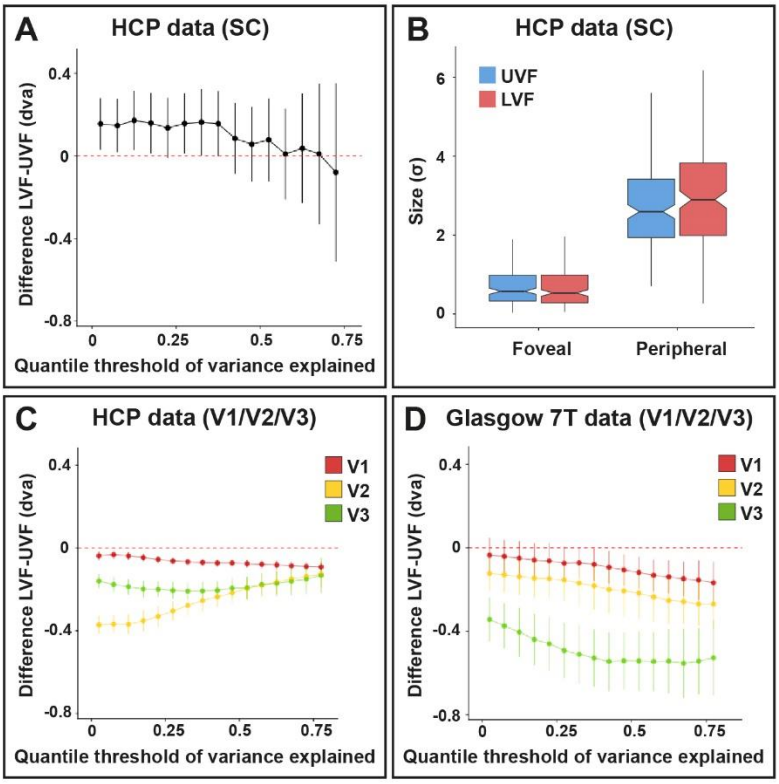


Figure 1: Characterising trends in population receptive field (pRF) size across upper and lower visual fields (UVF, LVF) in the superior colliculus and early visual cortex.
A: Difference between pRF size of LVF and UVF over varying quantile thresholds of variance explained for superior colliculus (SC) data. A similar trend compared to findings of monkey neurophysiology can be observed, with pRFs in LVF being larger than in the UVF. The trend becomes more prominent with more considerate threshold levels due to the low number of remaining voxels used for analysis. The lines represent bootstrapped confidence intervals.
B: The difference between pRF size across the UVF and LVF for the SC when split according to the proximity of pRFs towards the fovea. For pRFs nearer the fovea, the difference appears to be negligible, while for more peripheral pRFs the difference approaches significance.
C/D: Same as A, but now showing data for areas of the early visual cortex (V1, V2, V3). Results in C represent data of 181 participants obtained from the HCP retinotopy dataset (Benson et al., 2018). Results in D represent data of 18 participants obtained in Glasgow using the moving bar retinotpic paradigm at the resolution of 7T fMRI. The trend appears to go into the opposing direction of the results observed in the SC data, with pRFs being larger in the UVF than in the LVF. The trend appears to be present for all three analysed early visual areas. Bootstrapped confidence intervals for D are larger, possibly due to the decerased size of the dataset compared to C.

Disclosures: K. Moravkova: None. **A. Fracasso:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.17

Topic: D.06. Vision

Support: CRCNS: NSF 2011542 (GS) and BMBF 01GQ2002 (MK)
MnDrive Graduate Fellowship in Neuromodulation

Title: Intracortical network interactions in the developing ferret visual cortex

Authors: *H. N. MULHOLLAND[1], D. KONG[2], M. KASCHUBE[2], G. B. SMITH[1];
[1]Neurosci., Univ. of Minnesota-Twin Cities, Minneapolis, MN; [2]Frankfurt Inst. For Advanced Studies, Frankfurt Am Main, Germany

Abstract: Prior to eye opening, spontaneous activity is already highly organized into modular, distributed patterns, revealing large-scale correlated networks that are predictive of future functional networks and exist in the absence of long-range horizontal projections and patterned feedforward activity. Evidence from computational modeling predicts that these distributed

patterns could emerge from a self-organizing network comprised of purely short-range intracortical interactions involving lateral inhibition. If large-scale correlation structures are an emergent property of the network arising from local activity propagating throughout the cortex, then inactivating small, localized circuits should result in global disruptions in network function. Using a custom designed microscope, we can simultaneously image wide-field epifluorescent calcium activity and optogenetically stimulate specific neural populations with temporal and spatial precision, allowing us to test specific predictions of the role of lateral interactions in developing networks in vivo. Here, we virally expressed GCaMP6s in excitatory neurons and the red-shifted channelrhodopsin Chrimson-ST in inhibitory neurons in layer 2/3 of young ferret visual cortex. Prior to eye opening (postnatal day 24-29), we imaged both baseline spontaneous activity and visually evoked ON/OFF responses to a full-field change in luminance. We then repeated the experiments while optogenetically stimulated small (~300 μm) regions of inhibitory cells to silence local activity. We found that local inhibition not only effectively dampened both visually evoked and spontaneous activity at the site of stimulation, but that regions up to 2mm away showed disrupted activity. These disruptions were evident in spatial rearrangements of the large-scale correlation maps, with significant decreases in similarity between baseline and opto-inhibited correlation networks. Importantly, spontaneous correlation patterns returned to baseline after optogenetic inhibition, allowing us to reversibly interrogate the degree to which different parts of the cortex integrate into the network. Additionally, principal component analysis of the opto-inhibited events reveals that disruptions to the underlying network may lead to the suppression of prominent components and the enhancement of others. Together, this is strong evidence that millimeter-scale patterns of distributed activity arise from a self-organizing network of propagating short-range intracortical interactions.

Disclosures: **H.N. Mulholland:** None. **D. Kong:** None. **M. Kaschube:** None. **G.B. Smith:** None.

Poster

546. Functional Architecture and Circuits in the Visual Cortex

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 546.18

Topic: D.06. Vision

Support: NSF 2011542
BMBF 01GQ2002

Title: Universality of modular correlated networks across the developing neocortex

Authors: *N. J. POWELL[1], B. HEIN[2], D. KONG[3], J. ELPELT[3], H. N. MULHOLLAND[1], M. KASCHUBE[3], G. B. SMITH[4];
[1]Neurosci., Univ. of Minnesota-Twin Cities, Minneapolis, MN; [2]Columbia Univ., New York, NY; [3]Frankfurt Inst. For Advanced Studies, Frankfurt Am Main, Germany; [4]Neurosci., Univ. of Minnesota, Minneapolis, MN

Abstract: In order to deal with a complex environment, animals form a diverse range of neural representations that vary across cortical areas, ranging from unimodal sensory input to higher-order representations of goals, outcomes and motivation. The diversity of these representations suggests a high degree of specialization in functional organization across cortical areas, however, the developmental origin of this diversity remains unclear. Diverse representations may be rooted in an area-specific functional organization established early in development by endogenous mechanisms. Alternatively, a common representational architecture may exist across the early neocortex, established by common rules of dynamic network interactions, while functional specification arises primarily through area-specific inputs. Here we address this fundamental question by examining spontaneous activity across the developing ferret cortex. We show that spontaneous activity in both sensory (visual—V1, auditory—A1, and somatosensory—S1) and association cortices (posterior parietal—PPC and prefrontal—PFC) is highly modular and exhibits millimeter scale correlations. Across all areas in animals 7-10 days prior to eye opening, modular patterns of spontaneous activity were nearly indistinguishable from those seen previously in V1, which reflect the columnar representation of visual features. Over the subsequent 3 weeks, a period spanning both eye opening and ear canal opening, we find that while both the degree of modularity and the strength of long-range correlations decline with age, all cortical areas retained significant modular structure. In all areas examined, spontaneous activity became increasingly sparse and higher dimensional over this period, suggesting an improved representational capacity with increasing maturity. Furthermore, similar to published reports in V1, sensory evoked activity in A1 exhibits strongly modular responses with significant statistical similarity to spontaneous activity, suggesting that early spontaneous networks seed developing cortical representations in sensory areas and raising the possibility of a similar relationship in higher association areas such as PFC. Together, our results demonstrate that modular networks with long-range correlations in spontaneous activity are not unique to columnar V1, but rather are a universal feature during development. These findings suggest that the diverse representations found across neocortex may arise from a common developmental origin.

Disclosures: **N.J. Powell:** None. **B. Hein:** None. **D. Kong:** None. **J. Elpelt:** None. **H.N. Mulholland:** None. **M. Kaschube:** None. **G.B. Smith:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.01

Topic: D.08. Multisensory Integration

Support: Medical Research Council (MRC)
Wellcome Trust
Cancer Research UK
The Alan Turing Institute

Title: Multisensory integration in the mouse visual cortex

Authors: *A. EGEA WEISS[1], A. DOMANSKI[1], A. VIDUOLYTE[2], X. CANO-FERRER[1], G. KONSTANTINOU[1], M. F. IACARUSO[1];
[1]The Francis Crick Inst., London, United Kingdom; [2]Univ. Col. London, London, United Kingdom

Abstract: The animal brain continuously receives a wealth of sensory information, and uses this information to guide behaviour. However, the process by which the different sensory streams are integrated to form a unified percept remains poorly understood. Interestingly, the mammalian sensory cortices, where much of the processing of sensory information is thought to take place, feature direct connections between areas primarily dedicated to different sensory modalities. The auditory cortex, for instance, sends numerous projections to the visual cortex in mice. These connections may be crucial to the integration and binding of sensory inputs at early stages of processing. Identifying what information is carried by these cortico-cortical projections is a necessary step towards understanding this integration. Furthermore, elucidating how these cross-modal inputs are integrated with the processing of other sensory and non-sensory variables within their target neural populations is crucial to understanding their role.
We used dual-colour two-photon calcium imaging in head-fixed mice to simultaneously record the activity of axons from auditory cortex and cell populations in visual cortex while mice were presented with a series of auditory and visual stimuli, including pure tones, sounds originating from different spatial locations, and naturalistic movies. As expected, auditory cortex axons showed tuning to tone frequency. Interestingly, these axons also showed tuning to sound location. We examined how their activity and feature selectivity relate to response properties in the surrounding visual cortex. Finally, we investigated the encoding of the animal's uninstructed body movements in these auditory and visual populations, and observed how it interacts with their encoding of sensory variables.

Disclosures: A. Egea Weiss: None. **A. Domanski:** None. **A. Viduolyte:** None. **X. Cano-Ferrer:** None. **G. Konstantinou:** None. **M.F. Iacaruso:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.02

Topic: D.08. Multisensory Integration

Support: SWC PhD Programme - The Gatsby Charitable Foundation (GAT3755) and The Wellcome Trust (219627/Z/19/Z)
Wellcome Trust 223144
BBSRC T016639

Title: Probing the integration of auditory, visual and motor signals in mouse superior colliculus

Authors: *F. TAKACS, P. COEN, M. ROBACHA, C. BIMBARD, T. SIT, K. D. HARRIS, M. CARANDINI;
Univ. Col. London, London, United Kingdom

Abstract: [Introduction] The superior colliculus (SC) contains visual, auditory and motor maps that are thought to support sensorimotor transformations. However, it is not clear how SC neurons integrate stimuli of different modalities, whether this integration depends on brain state, and whether it relates to motor activity.[Methods] We used chronically implanted Neuropixels 1.0 and 2.0 probes to record SC responses (~900 neurons) while head-fixed mice performed a spatial audiovisual decision task. After each behavioral session, we also recorded responses from the same neurons during passive presentation of the stimuli (checkerboards images and pink noise bursts played in multiple azimuthal positions). We presented the same stimuli to a separate cohort of untrained naïve mice while recording acutely (~2,600 neurons).[Results] In both naïve and trained mice, we observed neurons responding to visual and auditory stimuli, but neurons responding to both stimuli were rare (~5%). In those neurons, audiovisual integration was predominantly additive. Most audiovisual neurons responded to all auditory stimuli, regardless of azimuthal location, and these non-spatial auditory neurons were the most correlated with uninstructed stimulus-evoked movements. During the task, visual signals predominantly appeared in the superficial SC, while auditory and motor signals appeared in a mixed population of neurons in deeper layers. [Conclusions] Our results suggest that SC neurons integrating auditory and visual spatial location exist but are rare. This integration is additive, echoing the behavior of the mice (Coen et al bioRxiv 2021). Visual signals are confined to the superficial layers and undergo little contextual modulation, while auditory signals are predominantly in deep layers and are modulated by body movements, both uninstructed and task-related. We are currently investigating whether auditory and visual stimuli are represented differently in naïve and trained mice.

Disclosures: F. Takacs: None. **P. Coen:** None. **M. Robacha:** None. **C. Bimbard:** None. **T. Sit:** None. **K.D. Harris:** None. **M. Carandini:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.03

Topic: D.08. Multisensory Integration

Support: Medical Research Council (MRC)
Wellcome Trust
Cancer Research UK

Title: Temporal integration of audio-visual stimuli in the mouse superior colliculus

Authors: *G. BIANCHINI, X. CANO-FERRER, G. KONSTANTINOU, M. F. IACARUSO;
The Francis Crick Inst., London, United Kingdom

Abstract: Our sensory systems continuously process inputs from different modalities and organise these streams of information such that our subjective representation of the outside world is a unified experience. The integration of multimodal information is strongly dependent on the relative timing of sensory inputs. When multiple forms of sensory stimuli arise from a single source, the synchrony of these stimuli can contribute to perceptual binding and provide distance cues. The difference between the velocities of light and sound introduces lag for auditory signals with respect to visual information, this delay scales with distance and therefore could carry information about the distance to the stimulus source.
Here we investigated how audio-visual signals are integrated in the mouse superior colliculus (SC), a midbrain area that represents the location of visual and auditory targets topographically. Neuronal activity was recorded with Neuropixels probes in awake animals presented with visual and auditory stimuli with staggered onset times (ranged from 0 to 300ms). The location of the recording site was varied along the antero-posterior and latero-medial axes of the SC and confirmed by combining electrophysiological features with histological reconstructions of fluorescently-labelled probe tracks. Around 24% of recorded neurones were modulated by both visual and auditory input. Multi-modal neurons were located across the entire depth of the SC. SC neurons showed a median latency of 28 ms (IQR: 27 ms) for auditory and 77 ms (IQR: 36 ms) for visual responses and exhibited a broad range of audio-visual delay preferences.
These results indicate a variable integration window for audio-visual stimuli that could allow SC neurons to encode a broad range of distances. Finally, we characterised the anatomical organisation of audio-visual delay encoding in the SC.

Disclosures: G. Bianchini: None. **X. Cano-Ferrer:** None. **G. Konstantinou:** None. **M.F. Iacaruso:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.04

Topic: D.08. Multisensory Integration

Support: NSERC Discovery Grant

Title: Past and present experience alters audiovisual temporal and synchrony perception in rats

Authors: *M. AL-YOUZBAKI, P. ARORA, A. L. SCHORMANS, B. L. ALLMAN;
Anat. & Cell Biol., Western Univ., London, ON, Canada

Abstract: In providing us with a complete perceptual experience, our brain naturally merges information from different sensory modalities (e.g., vision and hearing). This ability to integrate audiovisual stimuli is not static but adapts to conditions within our environment. For example,

human studies have shown that past and present experiences can significantly affect both the accuracy and sensitivity of audiovisual perception. Interestingly, some clinical populations (e.g., ASD and schizophrenia) experience deficits in the perceptual binding of audiovisual stimuli, often observed through the widening of the temporal window of integration. These perceptual deficits imply disruption of the neural circuitry underlying audiovisual binding. While extensive research has been conducted in this field using human participants, the neural mechanisms underlying audiovisual perception remain elusive, due in part to a lack of translational studies using suitable animal models. To that end, we set out to develop and validate rat models of audiovisual temporal and synchrony perception; important first steps toward conducting mechanistic studies. To investigate audiovisual perception, rats were trained to report whether an auditory or visual stimulus was presented first (i.e., temporal order judgment; TOJ task), or trained to perform a synchrony judgement (SJ) task, in which they reported whether they perceived an auditory and a visual stimulus pair to be presented at the same moment in time (synchronous) or at different times (asynchronous). Once trained using operant conditioning, rats were tested under different protocols to reveal the suitability of the tasks to model audiovisual perception in humans, including: (1) rapid recalibration to asynchronous stimuli (2) sensory adaption to prolonged stimulus exposure (3) performance changes based on altered testing conditions, and (4) disruption of glutamatergic neurotransmission. Overall, rats were able to adapt to prolonged stimulus exposures in both tasks, but only showed evidence of rapid recalibration to asynchronous stimuli in the TOJ task. Similarly, altering the testing conditions through the introduction of a background noise affected performance on the TOJ but not the SJ task. Finally, disrupting glutamatergic neurotransmission through systemic injections of MK-801 altered audiovisual perception in both tasks. Overall, the rat TOJ task appears to be the better choice for the mechanistic studies that we are planning to conduct; however, the SJ task still warrants further consideration due to its relevance to clinical populations.

Disclosures: **M. Al-youzbaki:** None. **P. Arora:** None. **A.L. Schormans:** None. **B.L. Allman:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.05

Topic: D.08. Multisensory Integration

Support: DFG CRC/TRR-135, project number 222641018
HMWK Clusterproject TAM

Title: Visual-tactile integration during simulated self-motion in macaque area VIP

Authors: *S. DOWIASCH[1,2], J. CHURAN[1,2,3], A. KAMINIARZ[1,2], F. BREMMER[1,2]; [1]Philipps-Universität Marburg, Marburg, Germany; [2]Ctr. for Mind, Brain and Behavior, Marburg, Germany; [3]German Primate Ctr., Göttingen, Germany

Abstract: Self-motion through an environment induces various sensory signals, i.e., visual, vestibular, auditory, and tactile. In contrast to visual and vestibular stimulation, the interaction of visual and tactile self-motion stimuli has been rarely considered. In a previous behavioral study in humans, we could show an influence of tactile flow on bimodal (visuo-tactile) heading perception. The neural basis of this behavioral effect is currently unknown. Neurons in the macaque ventral intraparietal area (area VIP) have been shown to respond to visually simulated (optic flow) and real (vestibularly driven) self-motion but also to tactile (and auditory) stimulation. Remarkably, spatial locations of visual and tactile receptive fields are spatially congruent and preferred directions for motion are coaligned. This suggests an involvement of area VIP in the processing of visuo-tactile self-motion information.
In our current study, we presented visual and tactile self-motion stimuli to a macaque monkey while recording neural activity in area VIP. Visual stimuli simulated self-motion through a 3D-cloud of dots in three out of eight directions, covering the full azimuthal stimulus space (centered on the neuron's preferred heading direction). Tactile stimuli were presented via one of eight nozzles, with an angular separation of 45°. Each neuron was tested in 9 (8 tactile directions + no tactile flow) times 4 (3 visual directions + no optic flow) conditions.
Overall, 63% of the neurons responded to visually simulated self-motion, 59% responded to tactile stimulation and 34% responded to both modalities. We found that, on average, purely visually selective neurons showed lower baseline activities and higher stimulus response activities as compared to purely tactilely selective neurons. Interestingly, bimodal neuron responses formed a mixture, i.e. their baseline activity corresponded to those of purely tactile neurons, while their stimulus response to each individual modality resembled those of the respective selective neurons. However, during bimodal stimulation, the visual response was highly dominant. The response latency for both modalities was almost similar.
In conclusion, our data show that macaque area VIP encodes visual-tactile self-motion information. Remarkably, responses do not reflect simple superposition of visual and tactile information, but more complex interaction terms. A functional equivalent of macaque area VIP has been identified in humans. This suggest that our results on visual-tactile interaction at the neural level can be transferred to human self-motion perception.

Disclosures: **S. Dowiasch:** None. **J. Churan:** None. **A. Kaminiarz:** None. **F. Bremmer:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.06

Topic: D.08. Multisensory Integration

Support: R01DC017532
R01DC020363

Title: Physical properties of eye movement-related eardrum oscillations (EMREOs) in rhesus monkeys

Authors: *S. N. LOVICH[1], D. M. KAYLIE[2], C. KING[3], C. SHERA[4], J. M. GROH[1];
[1]Dept. of Psychology and Neurosci., [2]Dept. of Head and Neck Surgery & Communication Sci., [3]Ctr. for Cognitive Neurosci., Duke Univ., Durham, NC; [4]Caruso Dept. of Otolaryngology, USC, Los Angeles, CA

Abstract: The auditory, visual, and oculomotor systems work together to aid spatial perception. We have recently reported an oscillation of the eardrum that is time-locked to the onset of an eye movement in the absence of sounds or visual stimuli. These eye movement-related eardrum oscillations (EMREOs) suggest that interactions between auditory, visual, and oculomotor systems may begin as early as the ear itself. Much is still unknown about this phenomenon. Open questions include: 1) Which motor systems of the inner and middle ear contribute to this eardrum oscillation? Potential candidates include the stapedius muscle, tensor tympani muscle, and/or outer hair cells. 2) What neural circuits drive this oscillation? 3) What are the cognitive or perceptual effects of this oscillation, especially with respect to sound localization? To study the anatomical and neural circuits, we use the rhesus monkey as a model to perform controlled invasive surgical and pharmacological manipulations. The rhesus monkey can perform saccadic eye movements on similar time scales to human participants, and we are able to record ear canal changes in the same manner as with human participants. Monkeys have a highly-reproducible oscillation in both ears, comparable to humans, including alternating phase of the oscillation between the ears and separable horizontal and vertical components related to the horizontal and vertical components of the eye movement. Finally, monkeys allow for a single, specific surgical or pharmacological intervention after baseline data collection, data collection almost immediately after the procedure, and data collection on the order of thousands of trials.

Disclosures: S.N. Lovich: None. **D.M. Kaylie:** None. **C. King:** None. **C. Shera:** None. **J.M. Groh:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.07

Title: WITHDRAWN

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.08

Topic: D.08. Multisensory Integration

Support: NIH Grant MH019929

Title: Comparison of Retro- and Orthonasal Olfaction in Rat Cortical Networks

Authors: *T. GRAY[1], I. GOLDSTEIN[1], D. B. KATZ[2];
[1]Volen Natl. Ctr. for Complex Systems, [2]Dept Psychol, Brandeis Univ., Waltham, MA

Abstract: Smell and taste are highly interdependent sensory modalities, but how activity in olfactory and gustatory circuits impact one another in real-time remains unclear. Previous work has demonstrated that activity in the gustatory cortex (GC) can influence odor encoding in the piriform cortex (PC) and that GC is required for retronasal odor discrimination (the smell of food in the mouth, which generally accompanies eating). The aim of this study is to identify the temporal dynamics of information transmitted from GC to PC in a dynamic manner to mediate retronasal odor perception. By comparing the temporal response profiles of retronasal odor responses in the two areas, we can determine if there is a unique relationship between each area's sensory responses in the context of retronasal olfaction. We have developed and successfully tested an easily produced 3D printed drivable electrode array that targets both PC and GC to look at these two regions simultaneously in freely moving animals. These electrophysiological recordings and manipulations provide insights to the characteristics and relationships between GC and PC retronasal odor responses. We will further dissect these relationships by comparing different ways in which GC processing specifically influences odor discrimination in the piriform cortex during consumptive behaviors.

Disclosures: T. Gray: None. **I. Goldstein:** None. **D.B. Katz:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.09

Topic: D.08. Multisensory Integration

Support: SNUBH Research Grant 06-2021-0355
SNUBH Research Grant 06-2021-0356

Title: Feasibility study of immersive virtual reality based post-stroke unilateral spatial neglect evaluation system

Authors: *W. CHANG[1], H.-M. CHO[1], S. CHA[1], S. CHO[2], S.-H. PARK[2], S. BAEK[2], W.-S. KIM[1], N.-J. PAIK[1];
[1]Seoul Natl. Univ. Bundang Hosp., Seongnam-si, Korea, Republic of; [2]Delvine Inc., Seoul, Korea, Republic of

Abstract: Background Unilateral spatial neglect (USN), a syndrome with reduced attention and awareness of the contralesional side of space, is a common impairment of right hemisphere

stroke. Recently, attempts were made to adopt immersive-virtual reality (VR) in the assessment of USN. In this study, we developed a novel Neglect Syndrome Assessment Tool (NeSAT) using immersive-VR and tested in the stroke patients and healthy subjects. **Method** We enrolled 12 right-handed healthy subjects (57.7±14.0 years old; 9 women and 3 men), and 14 right-handed patients with unilateral right hemispheric stroke (53.3±14.6 years old; 4 women and 10 men). NeSAT was developed using Pico Neo 3 Pro Eye (Pico Interactive Ltd., California, USA) to construct virtual environment. Subjects can move the pointer in the VR using a controller connected to VR and perform various tasks by pressing the button on the controller. There are two types of test in NeSAT; visual tracking and visual scanning. During visual tracking, the angle at which subjects identify a moving object (from the left to the center or the right to the center) on the screen and press the button was measured. The visual tracking angle difference (AD) was defined as the difference between the angle for an object from the left to the center and that from the right to the center. Visual scanning is similar to the apple test, pumpkins of various shapes are shown on the screen at regular intervals. Subjects were instructed to select normal pumpkins without any defects. The number of normal pumpkins correctly selected (CPS) and the difference between the number of pumpkins with defects on the right and left (defected pumpkin difference (DPD)) were measured. All stroke patients were evaluated with conventional Behavioral Inattention Test (BIT-C) and score below 129 were diagnosed as having USN. NeSAT results were compared between stroke patient with/without USN (USN+, USN- respectively) and the healthy subjects (HS) with Kruskal-Wallis test. Correlation between results of NeSAT and BIT-C in the stroke patients were analyzed with Spearman correlation. **Result** Of 14 stroke patients, 6 were diagnosed with USN. There were significant difference in AD(p=0.031) and CPS(p=0.006) between USN+, USN- and HS. Post-hoc analysis revealed significant difference between USN+ and HS in both AD (p=0.048) and CPS (p=0.004). In the stroke patients, BIT-C showed significant correlation with AD (r=-0.542, p=0.045), DPD (r=-0.675, p=0.008) and CPS (r=0.736, p=0.045). **Conclusion** This study revealed NeSAT as a valid, reliable tool for assessing USN in stroke. Results show the feasibility of developing a neglect battery test for the comprehensive evaluation of USN using VR.

Disclosures: **W. Chang:** None. **H. Cho:** None. **S. Cha:** None. **S. Cho:** A. Employment/Salary (full or part-time):; CEO of Delvine Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Has stock in Delvine Inc.. **S. Park:** None. **S. Baek:** None. **W. Kim:** A. Employment/Salary (full or part-time):; CMO of Delvine Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Has stock in Delvine Inc.. **N. Paik:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.10

Topic: D.08. Multisensory Integration

Support: Swedish Research Council

Title: Alpha oscillations bind visuotactile information responsible for body ownership

Authors: *M. D'ANGELO, H. EHRSSON;
Dept. of Neurosci., Karolinska Inst., Solna, Sweden

Abstract: Multisensory temporal integration of visual and visuotactile stimuli is thought to occur within the alpha frequency: stimuli are integrated when they fall within the same cortical cycle and segregated for different cycles. However, it is unknown if alpha oscillations similarly bind also bodily sensory information to generate the sense of body ownership, i.e., the experience of one's own body as one's own. Here, we clarify if the individual temporal resolution of perception correlates with the temporal resolution of multisensory integration involved in body ownership (Exp 1). Then, we showed that alpha oscillations temporally integrate visuotactile signals to originate the sense of body ownership (Exp 2). In Exp 1, 30 adults performed (i) a simultaneity judgment task (SJT) to measure their multisensory temporal resolution, i.e., participants judged the perceived synchronicity of visual and tactile stimuli separated by different delays, and (ii) a body ownership task, where we induced illusory body ownership over a fake hand. To induce the illusion, two robot arms applied taps to a fake hand and to the participants' hidden hand, either synchronously or at different asynchronies. After a short period of visuotactile stimulation, participants reported if they perceived the fake as their own hand or not. In Exp 2, we used transcranial alternating current stimulation (tACS) to modulate cortical oscillations at low (8Hz) or high alpha (13Hz) and also included a control condition with no brain stimulation (Sham). Participants (n=30) received tACS over the posterior parietal cortex while they performed (i) the ownership task and (ii) a modified SJT with the fake hand, i.e., participants judged if the taps on the fake and real hands were synchronous or not. Exp 1 showed that participants with a higher synchronicity threshold tolerated higher levels of asynchronies in the bodily illusion. Exp 2 found that tACS modulates multisensory integration in both tasks: Low alpha tACS, as compared to Sham, enlarges the synchronicity threshold and the level of asynchronies tolerated in the bodily illusion. High alpha tACS reduced the synchronicity threshold and the level of asynchronies tolerated in the bodily illusion. Thus, driving alpha frequencies toward faster vs. slower oscillations modulates multisensory temporal resolution involved in body ownership, just like in the SJT. These results indicate that alpha oscillations temporally integrate visuotactile information to generate the sense of body ownership, in addition to their role in multisensory temporal integration of external events.

Disclosures: M. D'angelo: None. **H. Ehrsson:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.11

Topic: D.08. Multisensory Integration

Support: NSF Grant 1707398 (Neuronex)
Gatsby Charitable Foundation Grant GAT3708
Boehringer Ingelheim Fonds

Title: Active electrolocation insights from artificial neural networks

Authors: *D. TURCU, A. ZADINA, L. F. ABBOTT, N. SAWTELL;
Dept. of Neuroscience, The Mortimer B. Zuckerman Mind, Brain and Behavior Inst., Columbia Univ., New York, NY

Abstract: Mormyrid electric fish generate pulsed electric fields and identify the resistive, capacitive and spatial properties of nearby objects by detecting microvolt and microsecond distortions in these fields. They do this despite much larger external modulations caused by self-motion, boundary effects and changing water conductivity. Mormyromast electrosensors on the skin of the fish are highly sensitive to the minute field distortions produced by objects. The cerebellum-like circuits of the electrosensory lobe process the mormyromast responses and enable the fish to perceive their surroundings. Although it has been suggested that particular features associated with the peaks of the electric pulse waveform are critical for object detection, the precise features used have not previously been characterized. By combining experimental measurements, neural network modeling and an electric field model, we identified temporal filters that characterize the operation of mormyromast electrosensors and developed a novel adaptive model of their sensory processing. Physical properties of the fish's environment can be extracted from responses generated by the resulting mormyromast model. Our electric field model unifies previous work into a fast and scalable analytic framework that allows us to generate large simulated data sets for developing and analyzing neural network models. We characterized the mormyromasts as temporal convolutional filters optimized to extract behaviorally relevant stimuli. We also fit convolutional filters to experimentally measured responses of the mormyromasts and found results that agree well with those obtained by optimization. Our mormyromast model continually adapts to slowly varying input modulations, in line with experimental findings. We generated electrosensory data from thousands of model mormyromasts, distributed on the skin of a virtual fish, responding to various environmental conditions and nearby objects in the presence of many tail positions. We trained feed-forward neural networks to process inputs from our model mormyromasts and extract physical properties of the environment. Naïve neural networks only partially extract some of these properties, but specialized networks developed from fitting the data learn to accurately predict numerous physical parameters characterizing the fish's surroundings. This result emphasizes the importance of specialization in the electrosensory lateral lobe. Our active electrolocation framework provides a basis for modeling computation in the cerebellum-like circuits of the fish's electrosensory lobe and sheds light on mammalian cerebellum function and computation.

Disclosures: D. Turcu: None. **A. Zadina:** None. **L.F. Abbott:** None. **N. Sawtell:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.12

Topic: D.08. Multisensory Integration

Support: NRF Grant 2021R1A2C3012159
NRF Grant 2021R1A4A2001803
KAIST Global Singularity Program for 2020

Title: Flexible integration of audiovisual inputs in the parietal cortex mediates multisensory decisions under audiovisual conflicts

Authors: *I. CHOI, J.-H. KIM, Y.-H. SONG, S.-H. LEE;
Biol. Sci., KAIST, Daejeon, Korea, Republic of

Abstract: Multisensory integration is a fundamental process underlying optimal decision-making in mammals. However, the state-dependent flexibility in multisensory decisions remains elusive. Here, we found that the mouse posterior parietal cortex (PPC) mainly represents visual information of the audiovisual stimuli, which was suppressed when the mice make auditory-dominant decisions in audiovisual conflict. Interestingly, locomotion suppresses auditory inputs to the PPC and switches auditory-dominant decisions to visual-dominant ones. In the auditory cortex (AC), locomotion suppressed neurons projecting to the PPC (AC_{PPC}) but not those projecting to the striatum (AC_{STR}). By circuit-specific optogenetic manipulation, we double-dissociated that the AC_{PPC} mediates auditory-dominant decisions in audiovisual conflicts and the AC_{STR} mediates unisensory auditory decisions. Moreover, axons of the secondary motor cortex (M2) projected to and suppressed the AC_{PPC} neurons, and this M2 input to the AC was critical for locomotion-dependent switches in multisensory decisions. Collectively, our data demonstrate that locomotion suppresses auditory afferents to the PPC to enhance visual decisions under audiovisual conflicts without disrupting unisensory decisions. This modulation promotes flexible perceptual decisions in actively moving animals exposed to multisensory stimuli.

Disclosures: I. Choi: None. **J. Kim:** None. **Y. Song:** None. **S. Lee:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.13

Topic: D.08. Multisensory Integration

Support: NIMH (R00MH115082-04)
Whitehall Foundation (2019-05-44)
Georgia State University Brains & Behavior Fellowship

Title: Evidence of Multisensory Predictive Coding Ensemble in Mammalian Posterior Parietal Cortex

Authors: *A. B. VAN DERVEER[1], J. P. HAMM[1,2,3];
[1]Neurosci. Inst., Georgia State Univ., Atanta, GA; [2]Ctr. for Neuroinflam. and Cardiometabolic Dis., [3]Ctr. for Behavioral Neurosci., Georgia State Univ., Atlanta, GA

Abstract: Context significantly influences mammalian sensory processing. In the predictive coding model, incoming sensory inputs are compared against sensory predictions based on previously encountered stimuli. Cortical responses to stimuli that align with predictions at that level of processing are suppressed, while stimuli that break from those predictions drive neural responses to send the information to the next level of the cortical hierarchy. Such prediction error and suppression responses exist within the primary visual cortex (Hamm et al., 2021). Whether and how such processes occur in hierarchically higher cortical areas, like those involved in multisensory (MS) processing, is not known, despite most real-world sensory information coming in MS formats. The posterior parietal cortex (PPC, PTLp in mice) is a MS integration region, as it receives inputs from primary and secondary sensory areas. We measured neuronal activity in PPC with two-photon calcium imaging using cre-dependent GCaMP6s expression in Vglut-Cre (pyramidal neurons, PYRs; n=10; 5 female) and SST-Cre (Somatostatin neurons; SST; n=10; 5 female) mice, during either unisensory (US) "oddball" paradigms, involving full square-wave gratings or pure tones in a random order (many standards control), then an oddball sequence with one redundant (88% "standards") and one deviant stimulus (12% "oddballs"), or a MS oddball. For the MS "oddball" paradigm, visual (45º [Va] and 135º [Vb]) and auditory stimuli (2.0kHz [Aa] and 4.0kHz [Ab]) were paired. Mice were trained with sequences of consistent AaVa and AbVb combinations, then MS "oddball" sequences, with 87.5% trained pairs and 12.5% deviant pairs (6.25% each AbVa, AaVb). We hypothesized that PYRs and SSTs in PPC would display the predictive coding responses of deviance detection (DD; i.e., enhanced responses to deviant stimuli) and stimulus-specific adaptation (SSA; i.e., reduced responses to redundant stimuli), but mainly to complex, MS stimuli.Our results show that PYRs exhibit DD to MS audio-visual oddball stimuli ($t(356)=5.62$; $p<0.001$) but not to US audio ($t(262)=-1.16$; $p=0.248$) or visual oddball stimuli ($t(241)=0.2$; $p=0.838$). Intriguingly, SSTs in PPC show significant US SSA (vis: $t(45)=4.5$; $p<0.0001$; aud: $t(19)=-2.7$; $p<0.015$) and DD (vis: $t(45)=3.08$; $p<0.005$; aud: $t(19)=-2.11$; $p=0.04$) but not MS DD. Together, these results suggest a predictive coding ensemble in PPC for MS stimuli, involving only complex "prediction error" responses in PYRs and an active suppression of simpler "prediction errors" by SST neurons.
NIMH (R00MH115082-04), Whitehall Foundation (2019-05-44), Georgia State University Brains & Behavior Fellowship

Disclosures: A.B. Van Derveer: None. **J.P. Hamm:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.14

Topic: D.08. Multisensory Integration

Support: NIH Grant DC018580
NIH Grant EY032230

Title: Visual instruction on the development of auditory spatial topographic map in mouse superior colliculus

Authors: *Y. SI[1], A. M. LITKE[2], D. A. FELDHEIM[1];
[1]MCD Biol., [2]SCIPP, UC Santa Cruz, SANTA CRUZ, CA

Abstract: Localizing a novel object in a complex environment and instantaneously evaluating its saliency is a fundamental brain function critical for survival. To achieve this, the brain needs to receive, process, and integrate sensory information from various modalities. A model to study spatial sensory integration is the superior colliculus (SC), a midbrain structure that receives somatosensory and auditory inputs that both align topographically with visual inputs. This sensory input provides information crucial about the location of objects with respect to the body and space. The SC then integrates these inputs to promote an appropriate motor response. Previous studies have elucidated that the development of the retinotopic maps uses a combination of genetic and activity-dependent mechanisms; expression of specific gene(s) and activity patterns give neurons instructions about where to project and with whom to synapse, thus instructing visual spatial map creation. However, the incident direction of the sound source must also be taken into consideration. It remains unknown how the computed auditory map of space develops and becomes aligned with the retinotopic map. We hypothesize that the topographic map of the auditory space in the SC develops using the retinocollicular map as a template. To test this hypothesis, we have manipulated the retinocollicular map by using retinal enucleated, dark-reared, and EphA3 knock-in mouse models. Together with blind analysis and large-scale in vivo physiological recordings of SC neurons in response to virtual space auditory stimuli, we are able to assess the topographic map of the auditory space under conditions whereby the retinocollicular map is altered. Our data show that in the absence of post-natal light-induced visual experience, the auditory topographic map of space maintains its overall topological structure; however, the range of azimuthal coverage on the anterior-posterior axis of the SC is expanded, and the precise registration of topography between the visual and auditory maps is impaired. This suggests that light-induced visual activity is not required to form but to fine-tune the topographic map of auditory space and the alignment with the retinotopic map in the mouse SC.

Disclosures: Y. Si: None. **A.M. Litke:** None. **D.A. Feldheim:** None.

Poster

547. Cross-Modal Processing I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 547.15

Topic: D.08. Multisensory Integration

Support: KAKEN JP 18KK0286
KAKEN JP 20H04286

Title: Evaluation of horizontal, vertical, and torsional optokinetic responses in goldfish as potential indicators of spatial orientation

Authors: *S. TADOKORO[1,5], Y. SHINJI[2], R. BAKER[6], Y. HIRATA[1,3,4];
[1]Dept. Robotic Sci. and Technology, Grad. Sch. of Engin., [2]Dept. Computer Sci. and Technology, Grad. Sch. of Engin., Chubu Univ., Kasugai, Aichi, Japan; [3]Ctr. for Mathematical Sci. and Artificial Intelligence, [4]Acad. of Emerging Sci., Chubu Univ., Kasugai,Aichi, Japan; [5]Otolaryngology, Head and Neck Surgery, Natl. Def. Med. Col., Tokorozawa, Saitama, Japan; [6]Dept. Neurosci., Grossman Sch. of Med., New York Univ., New York, NY

Abstract: Spatial orientation (SO) is the perception of own postural and motor behavioral state. SO is formed by integrating multi-modal sensory information mainly from vestibular, visual, and somatosensory systems. The velocity storage mechanism (VSM) has been postulated to play an important role In integrating vestibular and visual sensory information (Laurens & Angelaki, 2017). Characteristics of the VSM are known to be reflected in the reflexive eye movements called the vestibuloocular reflex (VOR) and optokinetic response (OKR) whose functions are to stabilize vision during head motion. One of the animal species most thoroughly understood for neuronal mechanisms of these eye movements is goldfish. Their VOR is known to be robustly induced for all horizontal (H), vertical (V), and torsional (T) head rotations (yaw, roll, and pitch, respectively); however, their OKR has been evaluated only in the H direction. Currently, we measured V and TOKR in 13 goldfish to identify their frequency characteristics and learning capabilities. Full-field velocity step visual stimulus were given alternating in direction every 10 s for 90 min around either of the three axes. Naïve animals presented minimal V and TOKR in contrast to their highly compensatory HOKR. However, V and TOKR gradually manifested, reaching to gains (max. eye velocity / visual stimulus velocity) greater than 0.3 on average by the end of the visual training. Interestingly, while VOKR gain increased in both upward and downward directions, TOKR gain increased only for extorsion. Frequency characteristics were identified to be low-pass filter-like as has been previously noted for the HOKR (Marsh & Baker, 1997). During 90 min visual training, greater gain increases were observed at lower frequencies in both V and TOKR. These results suggest that because the probability of visual motions around roll and pitch axes without congruent vestibular signals were very unlikely for naïve goldfish, the inadequate eye movements resulted from inaccurate formation of SO. Prolonged exposure to the incongruent visual motions alone updated the prior as well as the likelihood of visual and vestibular information in the Bayesian inference framework and resulted in appropriate SO formation to produce a more compensatory OKR. The time course of V and TOKR gain changes during visual training may provide insights into how fast the VSM updates its multi-sensory fusion process to form SO. The asymmetrical change in TOKR may be associated with greater value for pitch-down head motion than pitch-up during feeding behavior, making extorsion more flexible than intorsion to stabilize vision.

Disclosures: S. Tadokoro: None. **Y. Shinji:** None. **R. Baker:** None. **Y. Hirata:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.01

Topic: E.04. Voluntary Movements

Support: NSF GRPF
NIH NINDS R01NS096083

Title: The role of effort in motor learning

Authors: *R. MARBAKER[1], A. A. AHMED[2];
[1]Univ. of Colorado Boulder, Boulder, CO; [2]Mechanical Engin., Univ. of Colorado, Boulder, CO

Abstract: Movement error has long been known to be a driver of motor learning. Inherent to all movements is another cost, an effort cost of making the movement. Interestingly, similar to error, effort costs also reduce with motor learning. Is it possible that a desire to reduce effort costs also contributes to motor learning? More broadly, does the effort cost of movement influence how much we are willing to correct? Here we modulate the movement effort of performing a visuomotor rotation task, and ask whether effort alters the learning process. We hypothesized that when effort is dependent on task error, effort will accelerate learning. Additionally we hypothesized that when task effort is independent of error, effort will impair learning.
Participants (n=59) performed a visuomotor rotation task, reaching to targets positioned at 45 degree increments along the perimeter of a 10cm in five experimental blocks: i) baseline, ii) learning where a 30-degree rotation is introduced, iii) retention where visual feedback is removed, iv) washout, and v) savings where the 30-degree rotation is reintroduced. Four effort conditions were differentiated by modifying velocity-based damping resisting, b, of the reaching movement. In the first group, effort was dependent on the error of the previous reach, with larger errors incurring higher damping on the subsequent trial. The remaining three groups experienced effort costs that were independent of error with low (b=5Ns/m, n=17), medium (b=20Ns/m, n= 17), and high (b=35Ns/m, n=8) damping. While not explicitly increasing with error, higher damping increases the cost of online corrections. We compared behavioral metrics between groups using two-way repeated-measures anovas. Additionally we fit exponential curves and state-space models to determine learning rate, remembering factor, and error-sensitivity.
All groups learned the task, reducing error during learning, and with a faster reduction during savings. There were no differences between effort groups for learning metrics including initial error, final error (learning: p=0.63; savings: p=42), error reduction (learning: p =0.71; savings: p=0.82), and error change in retention (p=0.48). Exponentially fitted learning rates were also similar between groups (learning p=0.94; savings: p=0.35). No differences were detected between groups in the remembering factor (learning: p =0.50; savings: p=0.84) or error sensitivity (learning: p=0.94; savings: p=0.61). Our results suggest that neither the error-dependent or error-independent effort costs presented impacted motor learning in this task. Future work will investigate mandatory online correction.

Disclosures: R. Marbaker: None. **A.A. Ahmed:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.02

Topic: E.04. Voluntary Movements

Title: Implicit sensorimotor adaptation in participants with severe acquired and congenital proprioceptive loss

Authors: *A. CHANDY[1], J. S. TSAY[2], C. MIALL[3], R. CHUA[4], F. R. SARLEGNA[5], R. IVRY[6];
[1]Univ. of California, Berkeley, Berkeley, CA; [2]Psychology, Univ. of California Berkeley, Albany, CA; [3]Sch. of Psychology, Univ. of Birmingham, Birmingham, United Kingdom; [4]Sch. of Kinesiology, The Univ. of British Columbia, Vancouver, BC, Canada; [5]CNRS and Aix-Marseille Univ., Marseille, France; [6]Univ. California, Berkeley, CA

Abstract: Models of implicit adaptation have typically focused on how this process is driven by visual sensory prediction errors, without consideration of the possible role of proprioception. We address the latter by examining implicit adaptation in individuals who are functionally deafferented. While there has been previous work showing little effect of deafferentation on motor adaptation, the methods conflate implicit and explicit processes (Ingram et al., 2000; Bernier et al., 2006; Sarlegna et al., 2010; Miall et al., 2018), making the results difficult to interpret. We used a visuomotor adaptation task known to isolate implicit adaptation in which participants were instructed to reach with an unseen hand directly to a visual target while ignoring a visual cursor that moved at a fixed 30° offset from the target. Periodically, the participants were asked to report their perceived limb position after the movement was terminated, using a wheel of visual landmarks. The deafferented participants and their matched controls exhibited robust implicit adaptation, with the heading angle of the movements shifting across trials in the direction opposite to the cursor. Strikingly, the degree of implicit adaptation did not significantly differ between groups, suggesting that implicit adaptation is preserved in deafferented participants. Moreover, in both groups, reports of perceived limb position were biased towards the visual cursor, with a trend towards a greater bias in the deafferented group. This mismatch between perceived and desired position of the hand may result in a kinesthetic error that drives implicit adaptation, even in participants without peripheral afferent input.

Disclosures: **A. Chandy:** None. **J.S. Tsay:** None. **C. Miall:** None. **R. Chua:** None. **F.R. Sarlegna:** None. **R. Ivry:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.03

Topic: E.04. Voluntary Movements

Support: Region Bourgogne Franche Comte ANER Robotself

Title: Human integration of spatial and temporal structure in pointing trajectory sequence learning through human-robot face-to-face imitation

Authors: *P. DOMINEY[1], V. VUILLEMOT[2], G. POIRIER[1], C. PAPAXANTHIS[1], J. GAVEAU[1];
[1]INSERM U1093 CAPS UB/UBFC, Dijon, France; [2]ESEO, Dijon, France

Abstract: Sequential movement can be characterized by its organization in space and in time. Research simulating human cortical dynamics using recurrent reservoir networks has demonstrated that the serial and temporal structure of sensorimotor sequences are encoded in a shared representation. Evidence for this shared representation has been provided by human sequence learning experiments using the serial reaction time task. The current research examines how spatial and temporal structure of full upper-arm pointing sequences are learned. We tested 9 human subjects (6 male) age 22.4 +/- 1.8) in a sensory motor sequence learning task approved by the UBFC ethics committee CERUBFC-2021-10-07-028. Subjects were seated face-to-face with the humanoid robot Pepper. The robot made pointing movement sequences with its left arm. Subjects were instructed to follow the pointing finger-tip of the robot with their own right finger-tip, in a mirror image of the robot in front of them. An experiment consisted of 9 blocks of pointing movement to four targets (ABCD) in the shared frontal space between the human and robot. Each block consisted of 80 pointing movements with a duration of 90 seconds. Blocks 1-4,6,7,9 all used a sequence that had a fixed sequential structure, and temporal structure. Temporal structure was created by systematically inserting 500ms pauses before two of the 4 targets. Test blocks 5 and 8 systematically varied the sequential structure, while leaving the temporal structure intact, or vice-versa. Performance was evaluated by measuring the cross-correlation lag between the robot and human movement trajectories recorded by a motion capture system (Vicon, 200Hz). We randomized the order of sequential and temporal structure perturbations in Blocks 5 and 8. We measured the effects of Block, and of the order of the Space and Time shift. There is a significant effect for Block ($F(8,40) = 10.8$, $p < 0.0001$). Post hoc (LSD) tests revealed that lag for test block 5 is significantly greater than that for surrounding training blocks 4 and 6. Likewise, lag for test block 8 is significantly greater than the lag for training blocks 7 and 9. There is no effect for the order of the test blocks ($F(1,5) = 0.6$, $p > 0.4$). Importantly there is no interaction between the order of presentation, and the block effect ($F(8,40) = 0.3$, $p > 0.8$). This allows us to conclude that the spatial and temporal structure are both learned. These results are consistent with the hypothesis of a shared and integrated representation of the sequential and temporal structure of these sensorimotor sequences in a common recurrent cortical network.

Disclosures: P. Dominey: None. **V. Vuillemot:** None. **G. Poirier:** None. **C. Papaxanthis:** None. **J. Gaveau:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.04

Topic: E.04. Voluntary Movements

Support: NIH NINDS NS116883
Foundation for Physical Therapy Research
NIH NINDS NS120448

Title: Understanding strategic movement re-aiming as a process of reinforcement learning

Authors: *J. S. TSAY[1], H. KIM[2], J. A. TAYLOR[3], S. MCDOUGLE[4], A. M. HAITH[5], J. W. KRAKAUER[5], R. IVRY[1], A. G. COLLINS[1];
[1]Univ. of California Berkeley, Berkeley, CA; [2]Univ. of Delaware, Delaware, DE; [3]Princeton Univ., Princeton Univ., Princeton, NJ; [4]Yale Univ., New Haven, CT; [5]Johns Hopkins Univ., Baltimore, MD

Abstract: Multiple processes contribute to successful goal-directed movements, including top-down decision processes for movement selection and planning and lower-level processes that ensure movements are properly calibrated. A large body of empirical work using visuomotor rotation tasks has inspired computational models that characterize these lower-level implicit recalibration processes. However, computational accounts of top-down movement selection processes have been lacking. Here, we propose that the emergence of a re-aiming strategy can be understood within a reinforcement learning framework, where an action-value space is explored to maximize reward. We considered three models of strategic re-aiming: 1) Updating of action-value associations occurs only for the selected action via trial-and-error ("local update" model); 2) Updating of action-value associations occurs for all possible actions through an inference about the true underlying rotational relationship between the cursor and the hand ("global update" model); 3) Updating a belief about the size of the rotational relationship between the cursor and hand ("rotational update" model). To arbitrate between the three models, we examined two visuomotor rotation learning datasets in which participants strategically re-aimed to counteract a rotation imposed between their unseen hand and a visual cursor. Within 30 reaches, participants learn to re-aim in the opposite direction of the rotation, bringing the rotated cursor to the target. For both datasets, the global update model provided a better account of both the group data and idiosyncratic individual re-aiming behaviors. These results indicate that re-aiming after unsuccessful movements is neither a trial-and-error process - one that could be implicit and insensitive to the underlying rotation - nor an inferential process that explicitly updates an estimate of the rotation size. Rather, the data point to an iterative process that globally updates action-value associations via an inference about the true size and direction of the rotation. These results provide a computational perspective on a flexible, strategic process that allows the sensorimotor system to respond to rapid changes in the environment.

Disclosures: **J.S. Tsay:** None. **H. Kim:** None. **J.A. Taylor:** None. **S. McDougle:** None. **A.M. Haith:** None. **J.W. Krakauer:** None. **R. Ivry:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); RBI is a co-founder of Magnetic Tides.. **A.G. Collins:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.05

Topic: E.04. Voluntary Movements

Support: LSU CHSE Dean's Faculty Research Grant

Title: A simple 100% oxygen treatment improves motor sequence learning processes

Authors: Z. WANG[1], J. RODRIGUE[1], G. SPIELMANN[1,2], N. JOHANNSEN[1,2], F. GREENWAY[2], *M. DALECKI[1];
[1]Louisiana State Univ., Louisiana State Univ., Baton Rouge, LA; [2]Pennington Biomed. Res. Ctr., Baton Rouge, LA

Abstract: Our previous work showed that a simple normal-baric 100% oxygen treatment (OxTr) can improve motor learning processes of visuomotor adaptation. Motor sequence learning (MSL) is critical for rehabilitation and training settings and our daily lives as well, as a large number of functional movements involves correctly ordered sub-movements. However, it is not known whether OxTr improves MSL. Thus, we investigated whether a similar OxTr improves MSL performance, focussing on the acquisition of motor components themselves and transitions between sub-movements.Our preliminary data set included 29 healthy young adults, randomly divided into a 100% oxygen treatment (OxTr; N=15, M=20.86 yrs.) and an air treatment (AirTr; N=14, M=20.71 yrs.) group. Participants performed a motor sequence task by pressing the spatial-compatible key on a keyboard according to four visual stimuli. Two pre-determined 8-item sequences free of trills (e.g., 2-4-2-4) and runs (e.g., 1-2-3-4) were randomly assigned to test whether there is an effect of OxTr on MSL training progress. The experiment started with a baseline session with 10 trials of random 8-item sequences. Then, gas treatment was turned on and 4 training session blocks were conducted after a 3 minutes' break. Each training block included 30 trials of one sequence (deep training, DT) and 10 trials of a different sequence (shallow training, ST). Gas treatment was provided via nasal cannula (5 l/min), with 100% oxygen flow in group OxTr or regular air flow in group AirTr. Gas flow was turned off at the end of the training session, and after a 3-min break followed a final testing session of two blocks. Each block included 5 trials for each sequence (DT, ST, random). Main performance variables were response time (RT; ms) to visual cues and correctness rate of the response (CR; %). Data was normalized by subtracting each participant's baseline performance.ANOVA revealed a positive effect of OxTr on MSL. RT improvement was 16.1% greater in the OxTr than AirTr group (242 ± 63 vs. 208 ± 43 ms) for DT ($p=0.051$) and 27.0% greater (191 ± 73 vs. 150 ± 54 ms) for ST in testing sessions ($p<0.05$). Final testing session also showed a similar pattern ($p<0.05$). There was no significant CR difference between groups ($p>0.05$).Our results show that 100% oxygen supply reduced RT during a MSL task. Short-term consolidation of this effect sustained after the removal of gas treatment as well. Our study findings suggest that 100% normal-baric oxygen treatment could possibly improve motor sequence learning performance.

Such a treatment may have the potential for a cost-effective and easily accessible add-on therapeutic strategy in rehabilitation or skill training scenarios.

Disclosures: Z. Wang: None. **J. Rodrigue:** None. **G. Spielmann:** None. **N. Johannsen:** None. **F. Greenway:** None. **M. Dalecki:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.06

Topic: E.04. Voluntary Movements

Support: ASU Global Sport Institute
ASU Graduate College Graduate Research Support Program

Title: Placebo Effects of tDCS on Motor Acquisition

Authors: *N. K. HAIKALIS, A. HOOYMAN, H. DZIENDZIEL, L. DUNGCA, A. LEWIS, K. KAJITANI, H. GURRAM, B. HILL, S. Y. SCHAEFER;
Arizona State Univ., Tempe, AZ

Abstract: Studies using transcranial direct current stimulation (tDCS) to enhance motor training are often irreproducible. This may be due in part to differences in stimulation parameters (e.g., stimulation site, intensity, or duration) across studies, but it is also plausible that uncontrolled placebo effects may interact with the true 'treatment' effect of tDCS. Thus, the purpose of this study was to test whether there was a placebo effect of tDCS on motor training and to identify possible mechanisms of such an effect. Fifty-one participants (age: 22.2 ± 4.16; 26 F) were randomly assigned to one of three groups: active anodal tDCS (n=18), sham tDCS (n=18), or no stimulation control (n=15). First, participant expectations about how much tDCS could enhance motor function and their general suggestibility were assessed. Participants then completed 30 trials of functional upper extremity motor training with or without online tDCS. Stimulation (20-min, 2mA) was applied to the right primary motor cortex (C4) in a double-blind, sham-controlled fashion, while the control group was unblinded and not exposed to any stimulation. Following motor training, expectations about how much tDCS could enhance motor function were assessed again for participants in the sham and anodal tDCS groups only, along with their self-report of which stimulation type (sham or active) they believed they received. Results showed no effect of anodal tDCS on motor training (p=.23). However, there was a significant placebo effect, such that the sham and anodal tDCS groups improved more during motor training than the control group (p=.01). More specifically, average improvement in motor learning varied by group (p=.0005), such that the control group improved by 5.74 seconds from the first five to last five training trials (95% CI [5.36, 6.11]), the sham tDCS group improved by 8.38 seconds (95% CI [7.83, 8.93]), and the anodal (active) tDCS group improved by 8.9 seconds (95% CI [8.41, 9.39]). This placebo effect was not due to participants' overall suggestibility (p=.99) or

expectations about tDCS (p=.43). Interestingly, regrouping participants by *perceived* tDCS group showed that those who *thought* they received active stimulation improved more during training than those who thought they received sham, regardless of whether they actually received active stimulation. Thus, this exploratory study showed that there is a measurable placebo effect of tDCS on motor training, likely driven by participants' perceptions of whether they received stimulation. Future studies should consider placebo effects of tDCS, and identify their underlying mechanisms in order to leverage them in clinical care.

Disclosures: N.K. Haikalis: None. **A. Hooyman:** None. **H. Dziendziel:** None. **L. Dungca:** None. **A. Lewis:** None. **K. Kajitani:** None. **H. Gurram:** None. **B. Hill:** None. **S.Y. Schaefer:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.07

Topic: E.04. Voluntary Movements

Support: NIH Grant K12 HD055931

Title: The impact of Parkinson's disease on motor planning and adaptation

Authors: *H. KIM, J. TANG;
Univ. of Delaware, Newark, DE

Abstract: The basal ganglia are thought to play an important role in controlling movement amplitude and adapting future motor commands in response to errors in movement extent and direction (i.e., gain and angular adaptation, respectively). As such, numerous studies have examined the impact of Parkinson's disease (PD), a neurodegenerative disease of the basal ganglia, on motor planning and adaptation, but with mixed results. Additionally, surprisingly little is known about how PD impacts the scaling of movement amplitudes across different target distances or its effects on gain adaptation. Here, we set out to examine these questions during goal-directed reaching, leveraging behavioral methods and analyses that specifically assay feedforward control. We hypothesized that participants with PD would show greater planning deficits with respect to movement extent versus direction, especially during larger movements, and deficits in gain as opposed to angular (i.e., visuomotor rotation) adaptation based on recent work suggesting the latter remains intact. In Experiment 1, participants (n=10 patients ON meds, 9 age-matched controls) made center-out reaches to two 8-target sets differing only in their radial distances (6 cm vs 12 cm). To better isolate feedforward control, only endpoint visual feedback was provided. Using principal components analysis, we found that the PD group demonstrated larger errors in both movement extent and direction than controls, with more pronounced deficits for the far targets. The same participants were tested in Experiment 2, where we gradually perturbed visual feedback related to movement extent and angle using sinusoidal perturbation

schedules. Critically, the strong signal detection properties of this method allowed us to use perturbations that were a small fraction of the size that are typically used, thus minimizing cognitive contributions to performance changes. We examined adaptation in both movement extent and direction simultaneously by perturbing each at different temporal frequencies: 6 cycles/session sinusoid for movement gain (max: 8% change) and 4 cycles/session sinusoid for movement angle (max: 4 deg). Our frequency domain analyses showed that, while controls adapted robustly to these miniscule perturbations, the PD group were impaired in both gain and, surprisingly, angular adaptation. In addition, for the PD group, there was a gross association across experiments between planning and adaptation deficits. Taken together, PD broadly impacted motor planning and adaptation, results that are consistent with an expansive role for the basal ganglia in feedforward control.

Disclosures: H. Kim: None. **J. Tang:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.08

Topic: E.04. Voluntary Movements

Support: JSPS-JP16H06566

Title: Static and dynamic gaze sates can form conflicting visuomotor maps for arm-reaching

Authors: *N. ABEKAWA, H. GOMI;
NTT Communication Sci. Labs., NTT Communication Sci. Labs., Kanagawa, Japan

Abstract: In our daily lives, we perform skilled arm-reaching movements while looking at various locations. In addition, reaching movements are usually accompanied with eye movements. Although such static and dynamic gaze states are involved in reaching action, their potential contribution to arm motor learning has been largely overlooked. We have recently focused on two static gaze states, reaching to a target while looking at the target (Foveal-reach: FOV) and while looking elsewhere (Peripheral-reach: PER), and shown that two opposing motor skills (e.g., visuomotor rotations with different directions) can be learned simultaneously when each skill was associated with FOV and PER (Abekawa et al. 2022, Current Biology). While this result suggests that static gaze states can facilitate simultaneous learning, it still remains unknown whether and how dynamic chains of eye-hand movements are engaged in the learning of reaching. In Experiment 1, participants (n=12) performed reaching movements in three different gaze conditions. In the first two conditions, as in our previous study, participants reached to the target while maintaining gaze fixation (static conditions; FOV and PER). In the last condition, the trial started in the same way as in PER, but the fixation jumped to the target location before reaching was initiated. Participants had to make a saccade to a target followed by reaching toward that foveal target (dynamic condition; Sac-Reach). We applied visuomotor

rotation with its direction varied across trials, but was uniquely associated with the static or dynamic condition. In other words, clockwise rotation was applied for both FOV and PER, and counterclockwise rotation for Sac-Reach. The results showed that in all three conditions, the reaching error decreased to a similar degree throughout the training session (1200 trials), with clear aftereffects in the post-learning session where the rotation removed. This indicates that motor memoires for reaching can be separated by the gaze state prior to the reaching execution. To further examine if naturally chained eye-hand movements are necessary for such learning, we designed Experiment 2 (n=10), where a time gap of more than 1 second was inserted between eye and hand movements in the Sac-Reach condition. The results showed no progress in simultaneous learning, especially for the Sac-Reach condition. These results suggest that the dynamic gaze state, continuous chain of eye-hand movements, would activate neural states inherently involved in the visuomotor pathway, which can be tightly linked with the formation and retrieval of internal models of reaching.

Disclosures: N. Abekawa: None. **H. Gomi:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.09

Topic: E.04. Voluntary Movements

Support: ARC coAction UCLouvain

Title: Task reliability determines the arbitration between model-based and model-robust systems in human motor control

Authors: *H. KALIDINDI[1,2], F. CREVECOEUR[1,2];
[1]Inst. of Communication Technology, Electronics and Applied Mathematics, Univ. Catholique de Louvain, Louvain-la-Neuve, Belgium; [2]Inst. of Neurosci., Univ. Catholique de Louvain, Brussels, Belgium

Abstract: It is well known that humans use multiple systems of decision making, broadly categorized as model based (MB) and model free (MF) systems. An arbitration system is known to select between MB and MF systems depending on the task context to make the decision process flexible. However, in motor control much emphasis was placed on how the MB system maintains nominal movements under task perturbations. Recently, Crevecoeur et al., 2019 suggested a theory of model robust (MR) control that reduces the effect of task perturbations on movements, albeit incurring more effort. Day-to-day motor tasks are dynamic with varying levels of reliability. It is still unknown if a dynamic arbitration exists that selects between MB and MR motor controllers to optimize movements under different task conditions. So, we asked human participants (n=30) to perform reaching movements on the KINARM device in the presence of forcefields (FFs) in CW and CCW directions. The task reliability was controlled by

the probability of persistence in FF direction across trials. From optimal feedback control simulations, we found that the MR controller produces significantly different kinematics in the target direction compared to the MB controller which affected movements mainly in the perturbed directions (in terms of after effects). Moreover, the simulations predicted increased MR contribution with decreased task reliability. In the human experiments, the low reliability task had a higher MR and a lower MB contribution and vice versa. We further tested what happens when the task transitions from switching FFs (low reliability) to consistent FFs (max. reliability). We estimated the changes in the MB controller using force channel trials which let us measure the amount of model adaptation. In the low reliable condition, the adaptation was zero but surprisingly the hand deviation reduced exponentially by 33% across trials. Furthermore, the EMG activity around the peak in the relevant muscles linearly increased by 25%. These motor variable changes are predicted by an increasing contribution from the MR controller across trials. As the task transitioned to maximum reliability the MR contribution decayed exponentially and the MB contribution increased (measured from force channels). The hand deviation was further reduced by 17%, and the EMG activity reverted to baseline while the EMG peak shifted to the early movement period indicating anticipation. The arbitration process followed first order linear dynamics with a decay factor of 10 trials, with task reliability as its forcing term. This study highlights how humans flexibly arbitrate between different control systems to deal with changing task conditions

Disclosures: H. Kalidindi: None. **F. Crevecoeur:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.10

Topic: E.04. Voluntary Movements

Title: Effects of error size on sensorimotor learning under uncertainty

Authors: *C. L. HEWITSON[1], M. J. CROSSLEY[2], D. M. KAPLAN[2];
[1]Psychology, Yale Univ., New Haven, CT; [2]Sch. of Psychological Sci., Macquarie Univ., Sydney, Australia

Abstract: Adaptation of feedforward planning and feedback control of ongoing movements are essential error-reduction mechanisms for generating accurate motor behavior. Both the amount of adaptation induced by a movement error and the amount of feedback control correction issued to compensate for the real-time detection of that error are driven by its magnitude and direction. When measured in isolation from one another, the magnitudes of both of these error correction processes are inversely scaled by the operative level of sensory uncertainty. However, little is known about the role of sensory uncertainty for movement contexts that engage both feedforward and feedback correction mechanisms. In recent experiments, we found that in such contexts - when movement errors are constrained to be small - the effect of sensory uncertainty

on feedforward planning does not appear to modulate the error-driven response, but rather, exerts a powerful effect on other error-independent processes. However, it remains unknown whether this pattern holds for large error sizes. Here, we report on experiments showing that (1) sensory uncertainty influences feedforward adaptation in the presence of feedback integration even when errors are large, and (2) the influence of sensory uncertainty on feedback integration is eroded by large error sizes. This work helps to shed further light on the nature of sensorimotor learning under uncertainty.

Disclosures: C.L. Hewitson: A. Employment/Salary (full or part-time):; Postdoctoral Associate, full-time, Department of Psychology, Yale University. **M.J. Crossley:** A. Employment/Salary (full or part-time):; Senior Lecturer, full-time, School of Psychological Sciences, Macquarie University. **D.M. Kaplan:** A. Employment/Salary (full or part-time):; Deputy Head, full-time, School of Psychological Sciences, Macquarie University.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.11

Topic: E.04. Voluntary Movements

Title: Dissociating prediction and control during reaching under mirror reversal

Authors: X. DENG[1], C. YIN[1], A. M. HADJIOSIF[2], *A. M. HAITH[1];
[2]Neurology, Neurosci., [1]Johns Hopkins Univ., Baltimore, MD

Abstract: Numerous experimental findings suggest that the motor system contains both a controller, which generates a necessary motor command from desired outcome, and a predictor (or “forward model”), which generates expected outcome from an issued motor command. Theory suggests that prediction is inherently easier to learn than control, and it has thus long been thought that, when adapting one’s behavior to a novel environment, prediction is learned first, and is then used to guide control - potentially through an inversion of the predictor. We recently showed, however, that implicit adaptation to a mirror reversal is incompatible with this forward-model based account: errors induced by a mirror reversal get amplified, rather reduced, from one trial to the next, in a pattern that is incompatible with inversion of an updated predictor, but is compatible with a direct update of the controller (Hadjiosif et al., 2021). A limitation of this study, however, is that it did not directly dissociate changes in prediction and control and it remains unclear whether the predictor itself can adapt to a mirror reversal.We designed an experiment to measure both in parallel while reaching under mirror-reversed visual feedback. Participants made 2D reaching movements while direct vision of the arm was occluded; instead they could receive visual feedback on a screen mounted above their hand. After a familiarization/baseline session, we introduced a mirror reversal about the y-axis; this was followed by a block in which visual feedback was withheld to investigate how any adaptive changes to either predictor or controller decayed in the absence of error.To probe control, we

used trials where participants moved their hand through specific targets arranged close to the mirroring axis. To probe prediction, we used trials in which participants moved to self-selected locations close to the mirroring axis without visual feedback and, using the opposite hand, reported where they thought their hand just reached. Previous work has shown that such reports of hand location are strongly influenced by internal predictions about the consequences of motor commands (Synofzik, 2008). Predictor trials were interspersed with control trials at a ratio of 1:2. If the control function and the prediction function are inverse of each other, then there should be a close correspondence between patterns of reach errors in control trials, and reporting errors in prediction trials. If control and prediction are distinct functions that are learned independently, however, then we should instead expect a dissociation between prediction and control. Our results suggest that prediction and control are dissociable.

Disclosures: X. Deng: None. **C. Yin:** None. **A.M. Hadjiosif:** None. **A.M. Haith:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.12

Topic: E.04. Voluntary Movements

Support: NIH Grant NS116883
NIH Grant NS105839

Title: Interference of motor memories

Authors: *G. AVRAHAM, R. B. IVRY;
Univ. of California, Berkeley, Univ. of California, Berkeley, Berkeley, CA

Abstract: Savings refers to the gain in performance upon relearning. In sensorimotor adaptation, savings is tested by presenting the same perturbation over two learning blocks, separated by a washout phase. Savings has been attributed to the recall of an explicit strategy (e.g., Morehead et al. 2015). We have recently shown that implicit adaptation does not contribute to savings but is instead attenuated upon relearning (Avraham et al. 2021). Here, we hypothesize that this attenuation is due to feedback associated with the washout phase. When the perturbation is removed, participants experience an error in the opposite direction, a signal that drives behavior back to baseline. This experience may produce interference during relearning. We used a visuomotor task that isolates implicit adaptation. While reaching to a target, the cursor followed an invariant path with an angular offset from the target. Despite instructions to ignore the cursor, participants show implicit adaptation, with the hand path shifting in a direction away from the target (and cursor). In Exp 1, we replicated the attenuation effect in one group. For a second group, we eliminated all feedback during a long washout phase. This group did not show any attenuation, suggesting that attenuated relearning is due to interference from the feedback experienced during washout. In Exp 2, we asked if attenuation requires experience with the

salient opposite errors observed at early stages of washout. We eliminated these large errors by applying a rotation that was contingent on hand position, gradually decreasing the size of the rotation until the feedback became veridical. We then kept it veridical for an extended washout phase. Thus, participants experienced a distribution of small errors that was slightly shifted in the direction opposite from the error experienced during learning. Surprisingly, this group showed robust attenuation, suggesting that the interference effect does not depend on large opposite errors. Lastly, we asked if attenuation requires experience with opposite errors. In Exp 3, we tested learning after a long block of trials with veridical feedback. Interestingly, adaptation to a subsequent perturbation was attenuated. In Exps 4-6, we replicated this effect within-participants: The magnitude of attenuation was larger for targets that were previously associated with extended experience with veridical feedback. Thus, veridical feedback can produce proactive interference when experienced in a context of a subsequent perturbation. Overall, the results suggest that the formation and maintenance of motor memories is impacted by interference arising from prior experience in a similar context.

Disclosures: G. Avraham: None. **R.B. Ivry:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.13

Topic: E.04. Voluntary Movements

Support: CIHR PJT 165987

Title: Persistence of Adaptation following Visuomotor Training

Authors: *S. EBRAHIMI, D. OSTRY;
Psychology, McGill Univ., Montreal, QC, Canada

Abstract: Retention tests conducted after sensorimotor adaptation frequently exhibit a rapid return to baseline performance once the altered sensory feedback is removed. This so-called washout of learning stands in contrast with other demonstrations of retention, such as, savings on relearning and anterograde interference effects of initial learning on new learning. In the present study, we tested the hypothesis that washout occurs when there is a detectable discrepancy in retention tests between visual information on target position and somatosensory information on the position of the limb. Participants were tested following adaptation to gradually rotated visual feedback (15 degree or 30 degrees). In preliminary tests, we established that the smaller rotations were not detectable during learning and hence that any adaptation which occurred under these conditions was a likely consequence of implicit learning. Two different target types were used for retention testing, a point target in which a perceptual mismatch is possible, and an arc target which eliminated the mismatch. It was found that, with one exception, retention test movements were stable throughout after-effect trials, indicating no loss of information. Washout was only

observed in tests with a single point target, following adaptation to a large amplitude 30-degree rotation. This suggests that washout in aftereffect trials following visuomotor adaptation is due to a detectable mismatch between vision and somatosensation. Otherwise, there appears to be little loss of information.

Disclosures: S. Ebrahimi: None. **D. Ostry:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.14

Topic: E.04. Voluntary Movements

Support: NIH Grant R01DC017091

Title: Sensory errors drive speech adaptation even in the absence of overt movement

Authors: B. PARRELL[1], C. NABER[1], O. A. KIM[2], C. A. NIZIOLEK[1], S. D. MCDOUGLE[3]; [1]Univ. of Wisconsin–Madison, Madison, WI; [2]Princeton Univ., Princeton, NJ; [3]Yale Univ., Yale Univ., New Haven, CT

Abstract: When we make a movement, the observed outcomes of that movement sometimes differ from our expectations. These sensory prediction errors recalibrate the brain's internal models for motor control, reflected in alterations to subsequent movements that counteract these errors (sensorimotor adaptation). Leading theories of motor learning suggest that all forms of motor adaptation are driven by learning from sensory prediction errors (e.g., Krakauer et al., 2019; Hadjiosif et al., 2021). Conversely, dominant models of speech adaptation argue that adaptation results from integrating time-advanced copies of corrective feedback commands into feedforward motor programs (Guenther, 2016), which has also been suggested for reaching (e.g., Kawato et al., 1987; Albert & Shadmehr, 2016). Here, we test these alternative theories of speech adaptation by inducing planned, but not executed, speech: while the prediction error theory suggests that adaptation should only require a motor plan and a sensory error (Kim et al., 2022), adaptation in the feedback model requires overt speech acts. Human speakers (n = 17, 13F/4M) were prompted to speak a word and, on a subset of trials, were rapidly cued to withhold the prompted speech. On these "no-go" trials, just after withholding speech, speakers were exposed to playback of their own speech from the previous trial with an auditory perturbation of the first formant (i.e., the first resonant frequency of the vocal tract that helps to distinguish different vowels). Similar perturbations were also applied in real time to a subset of "go" trials to induce typical single-trial speech adaptation (Hantzsch et al. 2021). Results indicate that speakers adapt to auditory prediction errors on both go and no-go trials, altering the spectral content of the spoken vowel on subsequent trials to counteract the formant perturbation. Because adaptation occurred even in the absence of any movement, these results suggest that sensory prediction errors, rather than corrective motor commands, drive adaptation in speech.

Given that adaptation in speech has been extensively shown to involve only implicit learning processes, the current results solidify recent observations in reaching experiments also demonstrating motor adaptation evoked by sensory errors in the absence of overt movement (Kim et al., 2022). Our results extend that observation to a more complex movement system, one that relies on multi-dimensional auditory feedback rather than visual feedback. Our finding that sensory errors drive adaptation in speech even in the absence of movement points to a shared computational structure across human motor systems.

Disclosures: B. Parrell: None. **C. Naber:** None. **O.A. Kim:** None. **C.A. Niziolek:** None. **S.D. McDougle:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.15

Topic: E.04. Voluntary Movements

Support: NIH F31HD098796
NIH R56NS100528
NIH R21NS120274
NIH R01HD059783
NIH R41HD104296

Title: Adaptive Control is Reversed Between Hands After Left Hemisphere Stroke, and Lost Following Right Hemisphere Stroke

Authors: *R. VARGHESE[1,2], J. E. GORDON[3], R. L. SAINBURG[6], C. J. WINSTEIN[4], N. SCHWEIGHOFER[5];
[1]Johns Hopkins Univ., Johns Hopkins Med. Institutions, Baltimore, MD; [2]Ctr. for Movement Studies, Kennedy Krieger Inst., Baltimore, MD; [3]Biokinesiology & Physical Therapy, [4]Div. Biokinesiol/Physical Ther, [5]USC, Los Angeles, CA; [6]Kinesiology & Neurol., Penn State Univ., University Park, PA

Abstract: Human motor adaptability is of utmost utility after neurologic injury such as unilateral stroke. For successful adaptive control of movements, the nervous system must learn to correctly identify the source of a movement error and predictively compensate for it. Current understanding is that, in bimanual tasks, this process is flexible such that errors are assigned to, and compensated for, by the limb that is most likely to produce those errors. In right-handed adults, this is often the less-skilled, non-dominant left hand. After a stroke, muscle weakness, spasticity, and motor control deficits in the limb that is contralateral to the side of the stroke persist in the chronic phase causing it to become less skilled and produce more movement errors. Often, the non-paretic limb engages in prolonged functional practice and new skill learning to compensate for the paretic limb. The stark difference between the limbs in the likelihood of

movement errors and skill renders the chronic stroke model uniquely suited for the study of flexibility in the responsibility assignment process. Here, we tested the flexibility of the error assignment process in right-handed neurotypical adults (n = 20) and chronic stroke survivors (15 left and 15 right hemisphere stroke) using a redundant bimanual reaching task in which the hands jointly controlled a single cursor. We assayed adaptive control by examining compensation for two distinct error signals, within-trial compensation for sensory prediction errors and planned trial-by-trial adjustments for target errors. We predicted that errors will be assigned to, and compensated for, by the paretic hand, which is now more prone to error, regardless of the hemisphere of damage. We found that adaptive control is indeed reversed between hands after left hemisphere stroke, such that the paretic right hand predictively compensated for errors in cursor direction, more so than the non-paretic left hand. Furthermore, trial-by-trial adaptive response of the non-paretic left hand improved with time post stroke supporting the notion that adaptive control is learned and refined over time with practice. In individuals with right hemisphere stroke, however, rather than an exaggeration of existing interlimb asymmetries, adaptive control was lost for both hands. The loss of adaptive control observed in the right hemisphere stroke group seems inconsistent with the idea that responsibility assignment is fully flexible. The lack of compensatory adjustments for both types of errors in the right hemisphere stroke group point to the importance of right hemisphere circuits for flexible adaptive control of visually guided bimanual reaching movements.

Disclosures: R. Varghese: None. **J.E. Gordon:** None. **R.L. Sainburg:** None. **C.J. Winstein:** None. **N. Schweighofer:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.16

Topic: E.04. Voluntary Movements

Title: Evidence that human motor adaptation requires conscious awareness of error feedback

Authors: *A. D. FORRENCE, S. D. MCDOUGLE;
Psychology, Yale Univ., New Haven, CT

Abstract: What is the role of conscious awareness in motor adaptation? Studies on simple forms of associative and perceptual learning posit that learning can occur over subliminal inputs (Rosenthal et al., 2016; Pessiglioni et al., 2008). Crucially, some of this work has been called into question on methodological grounds (Skora et al., 2021), suggesting that key human learning processes may actually require conscious inputs. Although there is general agreement that the effects of motor adaptation on movement kinematics are implicit (Taylor et al., 2014; Tsay et al., 2020), it is not clear if adaptation is implicit "end-to-end." Here we ask if inputs to the adaptation system (i.e., errors) function when they are not consciously perceived. Subjects performed an error-clamp task that isolates implicit motor adaptation (Morehead et al.,

2017), moving to a single target using a mouse. At the start of movement, a visual mask was displayed (forward masking) and then quickly removed for a short interval during movement to reveal a feedback cursor rotated by 15° either CW or CCW (with direction randomized across trials). To induce unconscious perception, we rapidly applied a second mask (backward masking) after feedback exposure (Skora et al., 2021). Following each movement, subjects performed a forced choice concerning whether they perceived the feedback cursor or not. To maintain near-threshold conscious perception, we set the duration of error exposure using an adaptive staircase procedure. As expected, subjects showed significant single-trial motor adaptation following consciously detected error feedback. Crucially, adaptation was not seen following subliminal errors, and Bayes factors demonstrated strong evidence for the absence of adaptation on these trials. The amount of error exposure (i.e., the number of frames of visual error feedback) did not drive these effects, suggesting that differences in error exposure time could not explain the result. These findings provide evidence that motor adaptation does not proceed without conscious awareness of errors.

Disclosures: A.D. Forrence: None. **S.D. McDougle:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.17

Topic: E.04. Voluntary Movements

Support: CIHR Operating Grant MOP 106662
Faculty of Kinesiology
Calgary Health Trust

Title: Impairments in Visuomotor Adaptation are Associated with Motor Impairment After Stroke

Authors: *R. T. MOORE, M. A. PIITZ, N. SINGH, S. P. DUKELOW, T. CLUFF;
Univ. of Calgary, Calgary, AB, Canada

Abstract: Therapists use motor learning as a framework to help individuals with stroke relearn motor skills that are important for daily living. Past studies have shown the capacity to learn and adapt arm movements after stroke may be linked with motor impairment as measured by some clinical scales (e.g. Fugl-Meyer Assessment and Chedoke McMaster Stroke Assessment). However, these observer-based ordinal scales may not fully capture movement quality. There is consensus that detailed kinematic measures are needed to understand the relationship between motor control and learning, and that the approach may help identify behavioural biomarkers of motor recovery after stroke. Here, we examined the relationship between how individuals adapt their reaching movements to a visuomotor rotation and performance on a visually guided reaching task. Adults with first-time unilateral stroke were recruited within the sub-acute and

chronic stages of recovery. Controls matched for overall age and sex were recruited from the community. Subjects performed 1) a visuomotor adaptation task and 2) a visually guided reaching task in a Kinarm exoskeleton robot. Virtual targets and hand feedback were displayed in the subject's workspace using a virtual reality system. The adaptation task required subjects to reach back-and-forth between two targets (10 cm reach). Subjects performed 25 baseline trials with the feedback cursor aligned with their fingertip to measure their typical movement patterns. Subjects then made 125 movements with a 30 degree counter-clockwise rotation imposed on the motion of the feedback cursor. The angular deviation of the hand was measured 150 ms after the onset of each movement. This measure was used to assess the amount each subject adapted their movements and the number of trials they required to adapt to the rotation. The visually guided reaching task required that subjects reach from a central target to one of eight peripheral targets (10 cm reach). Kinematic measures assessed the straightness, smoothness, and timing of each movement. Group contrasts were conducted using bootstrap hypothesis tests. Spearman's correlations were used to examine relationships between motor adaptation and control measures. Subjects with stroke (n = 36) adapted slower and less than controls (n = 43). Impaired kinematics in the visually guided reaching task were associated with slower and reduced adaptation. Our results provide a deeper understanding of the link between impairments in motor control and adaptation after stroke. More research is needed to understand the link between motor learning, adaptation, and long-term motor recovery after stroke.

Disclosures: R.T. Moore: None. **M.A. Piitz:** None. **N. Singh:** None. **S.P. Dukelow:** None. **T. Cluff:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.18

Topic: E.04. Voluntary Movements

Support: Gift funding

Title: Exploring the benefits of immersive virtual reality exercise on cognition and sensorimotor control

Authors: *C. N. IRVING[1], J. S. TSAY[1], G. BROOKS[2], A. J. PECK[4], C. FUNG[4], R. IVRY[3]; [1]Psychology, [2]Integrative Biol., [3]Psychology, Helen Wills Neurosci. Inst., Univ. of California, Berkeley, Berkeley, CA; [4]Blue Goji, Austin, TX

Abstract: Two classes of interventions to reduce age-related declines in cognition center on physical exercise and cognitive training. Recent studies suggest that virtual reality games offer one platform for cognitive training, an engaging approach that should lead to better participant retention. This has motivated a new area of research at the intersection of physical fitness and gaming, or what has been called, "exergaming." Here, we asked whether exercising in an

immersive virtual reality platform would benefit cognition more than exercising alone. To test this, we compared the benefits of two types of exercise (n = 10 per group) on a battery of cognitive and sensorimotor control tasks. The immersive VR group played VR games while exercising on a customized treadmill system; the control exercise-only group exercised without the VR games. We evaluated cognition using the Adaptive Cognitive Evaluation (ACE), a battery that assesses attention, goal management, and working memory. We evaluated sensorimotor abilities with the Clinical Kinematic Assessment Tool, a battery that assesses human kinematics such as reaction time, movement time, and path accuracy in response to interactive visual stimuli. Each participant completed the psychometric assessment at four time points throughout the study: At initial study enrollment, after a two-week delay prior to the exercise program, and halfway and at the end of the exercise program. The exercise program lasted four weeks (three 35-minute sessions/week). We included two assessments prior to onset of the exercise to gauge performance changes due to time and/or repetition. All participants completed the trial and based on subjective reports, found the VR treadmill engaging and enjoyable. Individual differences were relatively stable across the assessments, indicating reasonable reliability of our measures. Preliminary results fail to show differences in improvement across sessions between the exercise-only and VR groups on any of the tasks. However, the initial results also point to limitations with our pilot study. The sample was composed of college students and most exercised on a regular basis. As such, the exercise intervention may not have been intensive enough to confer additional benefits. Additionally, performance on many of the tasks showed little improvement over time, pointing to a possible ceiling effect on these tasks for this population. Testing with older adults, including those who are more sedentary, would likely provide a more sensitive assay of the benefits of VR-enhanced exercise.

Disclosures: **C.N. Irving:** None. **J.S. Tsay:** None. **G. Brooks:** None. **A.J. Peck:** A. Employment/Salary (full or part-time):; BlueGoji. **C. Fung:** A. Employment/Salary (full or part-time):; BlueGoji CEO. **R. Ivry:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Co-founder of Magnetic Tides, Inc..

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.19

Topic: E.04. Voluntary Movements

Title: Does early-stage Alzheimer’s disease affect the dynamics of motor adaptation?

Authors: *K. SUTTER[1], L. OOSTWOUD WIJDENES[1], R. J. VAN BEERS[1,2], J. A. H. R. CLAASSEN[1,3], R. P. C. KESSELS[1,3,4], W. P. MEDENDORP[1];
[1]Donders Inst. for Brain, Cognition and Behaviour, Radboud Univ. Nijmegen, Nijmegen, Netherlands; [2]Dept. of Human Movement Sci., Vrije Univ. Amsterdam, Amsterdam,

Netherlands; [3]Dept. of Geriatric Med. & Radboudumc Alzheimer Ctr., [4]Dept. of Med. Psychology, Radboud Univ. Med. Ctr., Nijmegen, Netherlands

Abstract: Alzheimer's disease (AD) is the most common cause of dementia. The initial stages of the disease (referred to as mild cognitive impairment) are characterized by deficits in declarative memory, the recollection of explicit facts and events. Motor learning is traditionally viewed as a form of procedural learning, defined as the process of (re)gaining or retaining a given level of motor performance. While some studies suggest that AD patients are still able to (re)learn motor tasks, it is not clear how this motor learning is achieved.
Research in cognitively unimpaired participants has described motor adaptation as the sum of a fast process that learns and forgets quickly and a slow process that learns and forgets slowly. Because the fast process is driven by large, explicit errors, it has been suggested that it shares similarities with the declarative memory system. We therefore hypothesized that in AD patients the fast learning process is disturbed while adaptation follows the slow process that is still intact. Using a force-field adaptation task we evaluated the contribution of fast and slow processes of motor adaptation in AD patients (n = 20, age range 55-87, mean MoCA score 20.1, clinical dementia rating 0.5 - 1). Control groups were age-matched cognitively unimpaired participants (n = 21, mean MoCA score 26.4) and young healthy participants (n = 20). Participants performed reaching movements to a target while holding the handle of a robotic manipulandum. The experiment started with 100 trials without a force-field perturbing the reach, followed by 240 trials of a clock-wise force field with interspersed error-clamp trials, 30 trials of a counter-clockwise force-field and ended with 50 error-clamp trials. Motor adaptation was measured using the error-clamp trials in which participants reach to the target, but the movement was constrained to a straight line using a virtual channel, while the force in the channel was recorded. We computed the adaptation index (AI), i.e. the relative force that participants produced against the channel walls.
We used a hierarchical Bayesian two-state model to capture the dynamics of the AI by a fast and slow process. Our preliminary analyses are in line with previous observations showing that AD patients have selective deficits in motor adaptation, with a lower overall adaptation, and a time course that mostly hints at a slow learning process. Further analyses are currently under way to examine how AD patients differ from their age-matched controls as well as from young participants.

Disclosures: K. Sutter: None. **L. Oostwoud Wijdenes:** None. **R.J. van Beers:** None. **J.A.H.R. Claassen:** None. **R.P.C. Kessels:** None. **W.P. Medendorp:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.20

Topic: E.04. Voluntary Movements

Support: DST, Government of India

Title: Distinguishing the contributions of implicit and explicit mechanisms to savings in sensorimotor adaptation

Authors: *A. SHARMA, A. KUMAR, P. MUTHA;
Indian Inst. of Technol., Gandhinagar, India

Abstract: In sensorimotor adaptation, savings is defined as the faster relearning of a task that has been learned before. However, which of the many mechanisms that contribute to adaptation gives rise to savings, remains a matter of debate. One view is that savings arises from the recall of verbally-sensitive cognitive strategies employed largely to cancel out task performance errors during initial learning. However, other studies have argued that savings results from the operation of implicit mechanisms that bring about a reduction in limb-related sensory prediction errors. A primary reason for this debate is the difficulty associated with isolating the contributions of implicit and explicit mechanisms during adaptation. In order to address this issue, here we employed adaptation paradigms in which error reduction occurs either through implicit processes or explicit strategies, and then probed for savings during re-exposure to the same errors. Our first group of participants underwent visuomotor adaptation under conditions in which motion of the feedback cursor was clamped in a fixed direction regardless of the underlying hand motion. Furthermore, they were asked to ignore the cursor and reach to the on-screen target. Learning under such conditions is thought to proceed entirely implicitly, but it is unclear if any explicit strategies are also employed since a task error is always present. Following adaptation under these conditions, when subjects were re-exposed to the same error in a canonical visuomotor adaptation task, we did not find that learning was any faster than that of a control group that had not undergone any prior implicit learning. In other words, there was no evidence of savings. In a second group, we proceeded to remove any potential explicit learning by also shifting the location of the reach target such that the clamped cursor would always hit it, thereby eliminating task errors. Again, we found no evidence for savings in this group. Finally, a third group of subjects performed a task where the target jumped to a new location thereby producing a task error, but cursor feedback always remained veridical with the hand thereby eliminating any sensory prediction error. This ensured that learning occurred purely strategically and implicit mechanisms were not engaged. When this group was exposed to a rotation thereafter, they learned remarkably faster than controls, and demonstrated clear savings. Collectively, our results demonstrate that explicit strategy-driven mechanisms are necessary and sufficient to produce savings, and implicit-learning mechanisms do not contribute to faster relearning.

Disclosures: **A. Sharma:** None. **A. Kumar:** None. **P. Mutha:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.21

Topic: E.04. Voluntary Movements

Support: Facebook Technologies

Title: Cognitively demanding dual tasks do not strongly interfere with performance in the early stages of learning a new motor skill

Authors: *C. S. YANG[1], J. LIU[2], M. V. COLAVITO[1], J. W. KRAKAUER[1], A. M. HAITH[1]; [1]Neurol., [2]Biomed. Engin., Johns Hopkins Univ., Baltimore, MD

Abstract: Early in the process of learning a new skill, performance is typically thought to be cognitively demanding. For example, when initially learning to drive a car, it can be difficult to focus on anything other than driving, such as holding a conversation. However, with more practice, driving will demand fewer cognitive resources, allowing one to simultaneously perform other activities. Spatial working memory, in particular, has been suggested to be a key resource for learning visuomotor skills; abilities such as performing mental rotations and holding multiple spatial locations in working memory have been implicated as critical cognitive processes for motor skill learning. While the role of cognition has been explored in sensorimotor adaptation tasks, such as visuomotor rotations, these tasks may not provide a general model of skill learning as they can typically be learned quickly (within minutes) and are known to depend on implicit recalibration. Instead, we performed an experiment to examine whether, early in learning, spatial working memory would be required for the performance of "*de novo*" learned skills, i.e., skills that are learned from scratch. Participants (n = 16) learned to control an on-screen cursor using a non-intuitive bimanual mapping by performing point-to-point reaches. Previously, we have demonstrated that this mapping is learned *de novo* and that participants require roughly five days of practice for their performance improvements to asymptote. After two days of practice (i.e., early in learning), we measured the extent to which participants' ability to control the cursor under the bimanual mapping became worse while they simultaneously performed two other tasks that required spatial working memory. The first task involved mentally rotating one's reaching goal. The second task involved reaching towards the remembered positions of a sequence of targets. If the kinematic quality of participants' bimanual control declined relative to baseline while simultaneously performing the two tasks, this would suggest that the performance of the bimanual mapping depended on the cognitive resources that these tasks engaged. Surprisingly, we did not find any difference in the effect of the additional tasks on bimanual versus baseline control of the cursor. Critically, we did observe dual task interference between the two additional tasks, demonstrating that our results could not be explained by the fact that the tasks we chose were simply not cognitively demanding. These data suggest that the spatial working memory demands of performing motor skills can be low early in learning, even before one has become highly skilled.

Disclosures: C.S. Yang: None. **J. Liu:** None. **M.V. Colavito:** None. **J.W. Krakauer:** None. **A.M. Haith:** None.

Poster

548. Motor Learning: Human Psychophysics

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 548.22

Topic: E.04. Voluntary Movements

Title: Acquired skills can undergo implicit recalibration, but only after they have been extensively practiced

Authors: *A. M. HADJIOSIF, K. KITA, M. V. COLAVITO, A. M. HAITH;
Neurol., Johns Hopkins Univ., Baltimore, MD

Abstract: Perturbing movement-related sensory feedback typically leads to implicit, temporary, and reversible recalibration of the motor controller involved - a characteristic of established controllers involved in, e.g., arm reaching. Here we ask whether this kind of adaptability can arise in newly acquired controllers associated with a novel skill. We hypothesized that a well-practiced skill would exhibit implicit adaptation to an imposed perturbation similar to an established behavior like reaching, but that this capacity would not be present when the skill is less practiced.

We challenged healthy participants to learn a new skill in which they were required to control an on-screen cursor to a series of targets via a novel mapping from their hands to the cursor position. Forward-backward movement of the left hand controlled left-right movement of the cursor, and left-right movement of the right hand controlled forward-backward movement of the cursor. We have previously shown that this task is not learned through adaptation but is instead learned by building a new controller "do novo" over multiple days of practice (Haith et al., 2021). We compared two groups who either had substantial experience with the task (Experienced group, 6 day experience) or minimal experience (Novice group, 1 day experience). To evaluate how the newly learned skill would adapt to a perturbation, we subsequently exposed participants to a 15° visual-clamp perturbation (Morehead et al., 2017) in a center-out, target-shooting paradigm. To ensure that we only elicited implicit learning mechanisms, participants were told to keep aiming their (bimanually-controlled) hand to the target - an instruction mirroring what's told to participants in the standard, unimanual version of the task. This 15° visual-clamp was followed by a decay block in which the visual-clamp was set to zero, to observe the decay of adaptive changes in the absence of visual error.

The Experienced group implicitly adapted to the visual-clamp, with performance gradually drifting away from the target to about 20° and resulting in aftereffects that decayed gradually in subsequent zero-error clamp trials - paralleling established findings for reaching movements. In the Novice group, implicit adaptation to the visual-clamp was markedly reduced and any adaptive changes that did occur did not exhibit decay in zero-error clamp trials, suggesting a more permanent reconfiguration of the previously learned controller. Our results suggest that implicit, reversible recalibration in response to sensory errors can occur for a de novo learned controller, but only emerges after sufficient practice.

Disclosures: A.M. Hadjiosif: None. **K. Kita:** None. **M.V. Colavito:** None. **A.M. Haith:** None.

Poster

549. Neuroprosthetics, Brain Computer Interfaces, and Body Machine Interfaces: Motor Learning and Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 549.01

Topic: E.05. Brain-Machine Interface

Support: Intramural Research Program of the National Institute of Mental Health (ZIA-MH002893)

Title: Decoding action and observation of biological and robotic hand gestures

Authors: H. SCHONE[1,2], G. EDWARDS[1], S. JAPEE[1], T. R. MAKIN[2,3], ***C. I. BAKER**[1]; [1]Lab. of Brain and Cognition, NIH, Bethesda, MD; [2]Inst. of Cognitive Neurosci., [3]Wellcome Ctr. for Human Neuroimaging, Univ. Col. London, London, United Kingdom

Abstract: Research surrounding human augmentation and restoration technologies has sought to understand the flexibility of neural body representations to support the control of external devices, a concept known as *technological embodiment*. To test the validity of this framework in the brain, we investigated the neural representations supporting both biological and robotic hands, in the context of a longitudinal robotic hand training study. We trained two groups of able-bodied participants (n = 40) to use a multi-articulating myoelectric robotic hand over the course of four days (2 hours/day). The robotic hand was attached to participant's left arm and was operated by an 8-channel EMG pattern recognition system. Additionally, a separate control group (n = 20) was tested with the robotic hand without training. All participants underwent functional MRI (fMRI) scans before and after training (1-week apart for the controls). To characterize both visual and motor features of hand representation, participants performed a visuomotor task that required participants to either execute a specific hand gesture or to observe a first-person video of either a biological or robotic hand performing a gesture. Here, we primarily focus on the pre-training fMRI data (n = 60) to characterize the representational differences that exist between biological and robotic hands prior to training. Using representational similarity analysis, we measured differences in the representational structure of biological and robotic hands. Within visual brain regions associated with hands and tools, we observe a similar representational structure for viewing biological and robotic hands. Further, there was a distinct representational structure for executing gestures. However, we observed limited correspondence between the representational structure when viewing biological hand gestures and executing the same gestures. Ongoing analyses will attempt to decode across modalities (action, observation) and hand-type (robotic, biological) in occipitotemporal, sensorimotor, premotor, and parietal regions of interest. Overall, this work provides a critical framework for exploring the neural capacity for technological embodiment.

Disclosures: H. Schone: None. **G. Edwards:** None. **S. Japee:** None. **T.R. Makin:** None. **C.I. Baker:** None.

Poster

549. Neuroprosthetics, Brain Computer Interfaces, and Body Machine Interfaces: Motor Learning and Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 549.02

Topic: E.05. Brain-Machine Interface

Support: Intramural Research Program of the National Institute of Mental Health (ZIA-MH002893)

Title: Is the human body the best model for controlling artificial limbs? Comparing biomimetic and arbitrary control strategies

Authors: H. SCHONE[1,2], ***M. UDEOZOR**[1], M. MONINGHOFF[1], B. RISPOLI[1], J. VANDERSEA[4], B. LOCK[5], L. J. HARGROVE[6,7], T. R. MAKIN[2,3], C. I. BAKER[1]; [1]Lab. of Brain and Cognition, NIH, Bethesda, MD; [2]Inst. of Cognitive Neurosci., [3]Wellcome Ctr. for Human Neuroimaging, Univ. Col. London, London, United Kingdom; [4]Med. Ctr. Orthotics & Prosthetics, Silver Spring, MD; [5]Coapt Engin., Chicago, IL; [6]Dept. of Physical Med. and Rehabil., Northwestern Univ., Chicago, IL; [7]The Regenstein Fndn. Ctr. for Bionic Med., Shirley Ryan AbilityLab, Chicago, IL

Abstract: For individuals missing a limb, the development of robotic prostheses and their control often centers on embodiment as the ultimate goal: making devices more like the biological system (biomimetic). The untested assumption is that by using control strategies that are biomimetic, the user is likely to recruit already existing motor skill to control the prosthesis, therefore providing more intuitive control which will reduce the burden of skill training and improve the generalizability of learning. However, given clear differences between biological and prosthetic limbs, biomimetic control strategies might actually hinder usage, due to known neurocognitive bottlenecks. We compared two motor control strategies in participants controlling a robotic hand: biomimetic (mimicking the desired movement with their own hand) versus arbitrary (pairing an unrelated hand gesture with the desired movement). We focused on skill learning and generalizability. To accomplish this, we trained two groups of able-bodied participants (n = 40) to use a multi-articulating myoelectric robotic hand over the course of five days (2 hours/day). The robotic hand was attached to participant's left arm and was operated by an 8-channel EMG pattern recognition system, positioned around the forearm. Additionally, a separate control group (n = 20) was tested with the robotic hand without training. We used pre- and post- comparison measures to assess behavioural outcomes of robotic hand usage. Questionnaires for explicit sense of embodiment revealed increased embodiment over the robotic hand specifically in the trained users, with no differences across motor control strategies. The biomimetic group performed better than the arbitrary group on speed tasks, but both groups performed similarly on other aspects of robotic hand control (i.e. grasping precision and gesture switching). Crucially, when assessing how well the training generalized to a novel hybrid control strategy, the biomimetic group showed no benefits of training, and performed similarly to the no-training control group. In contrast, the arbitrary users showed significant behavioural gains which were further reflected in subjective ratings for control difficulty, relative to both other groups. Overall, our findings suggest that biomimetic and arbitrary control strategies may provide different benefits. The best strategy likely depends on the strengths of the technology, training opportunities and user requirements.

Disclosures: **H. Schone:** None. **M. Udeozor:** None. **M. Moninghoff:** None. **B. Rispoli:** None. **J. Vandersea:** None. **B. Lock:** None. **L.J. Hargrove:** None. **T.R. Makin:** None. **C.I. Baker:** None.

Poster

549. Neuroprosthetics, Brain Computer Interfaces, and Body Machine Interfaces: Motor Learning and Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 549.03

Topic: E.05. Brain-Machine Interface

Support: NIH 1 DP2 HD087955

Title: Leveraging rapid representational mesoscale neural plasticity for stable high degree of freedom brain computer interfaces

Authors: *N. NATRAJ[1], **S. SEKO**[1], R. ABIRI[3], A. TU-CHAN[1], E. F. CHANG[2], K. GANGULY[1];
[1]Neurol., [2]Neurolog. Surgery, Univ. of California, San Francisco, San Francisco, CA; [3]Dept. of Biomed. Engin., Univ. of Rhode Island, Kingston, RI

Abstract: Brain-computer interface (BCI) control of high degree of freedom (DoF) assistive devices (such as a robotic arm) have enormous potential in enabling reach-to-grasp function in human patients with severe motor disabilities. While impressive proof-of-principle demonstrations have been shown using spike-based BCIs, clinical translation to daily use has been challenging due to the difficulty in achieving daily reliable control of high DoF assistive devices, with long decoder calibration times and learning periods. Our recent work (Silversmith et. al., 2020) has shown that the stability of mesoscale electrocorticographic signals (ECoG) can be used to track neural plasticity and decoder learning, resulting in stable 'plug-and-play' (PnP) BCI control without the need for recalibration; however, this was done in the context of 2D cursor control. Here, we present a framework of leveraging the stability of ECoG, its representational content (Natraj et. al., 2022) and its ability to track neural plasticity across different time scales to achieve reliable PnP control of high DoF BCIs with minimal training and across a wide variety of use-contexts. Our BCI system involved a 128 channel ECoG grid placed over left sensorimotor cortex in a right-handed male tetraplegic. We extracted delta (0.5-4Hz), beta (13-30Hz) and high gamma (70-150Hz) power at each channel, subsequently binned at 5Hz. We first show that stable neural representations for imagined actions of the entire body are encoded by the single unihemispheric grid, with diverse spatial activations specific to the imagined action. The mesoscale representations were remarkably stable at the single trial level, allowing us to seed and rapidly consolidate a discrete high DoF decoder. We found evidence of rapid plasticity from imagined seeding to online BCI control, assessed using deep autoencoders, both within a single session and across a few days, resulting in a PnP high DoF decoder that was stable for up to 4 weeks (~2 bits per second on average). This decoder was then used to provide

user-inputs to control the 3D pose and gripper dynamics of a Kinova Jaco robotic arm. Our subject was able to proficiently control the robotic arm for reaching and grasping across a wide-variety of use-contexts (horizontal vs. vertical), perspectives (left-handed vs. right-handed, allocentric vs. egocentric) and environments (virtual vs. real-world physical). Median task completion times for reaching, grasping and object transport was 45s, with an accuracy of 100% within a week of practice. Our results thus highlight the potential of leveraging the rapid representational neural plasticity with ECoG for stable high DoF BCIs.

Disclosures: **N. Natraj:** None. **S. Seko:** None. **R. Abiri:** None. **A. Tu-Chan:** None. **E.F. Chang:** None. **K. Ganguly:** None.

Poster

549. Neuroprosthetics, Brain Computer Interfaces, and Body Machine Interfaces: Motor Learning and Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 549.04

Topic: E.05. Brain-Machine Interface

Support: ERC Grant Agreement No. 864729
DGA AID

Title: Comparison of strategies to elicit motor imagery-related brain patterns in multimodal BCI settings

Authors: *T. VENOT[1], A. DESBOIS[1], M.-C. CORSI[1], L. HUGUEVILLE[2], L. SAINT-BAUZEL[3], F. DE VICO FALLANI[1];
[1]Sorbonne Université, Inst. du Cerveau - Paris Brain Inst. - ICM, CNRS, Inria, Inserm, AP-HP, Hôpital de la Pitié Salpêtrière, Paris, France; [2]Sorbonne Université, Inst. du Cerveau - Paris Brain Inst. - ICM, CNRS, Inserm, AP-HP, Hôpital de la Pitié Salpêtrière, Paris, France; [3]ISIR, Sorbonne Univ., Paris, France

Abstract: Cognitive tasks such as motor imagery (MI) used in Brain Machine Interface (BMI) present many issues: they are demanding, often counter-intuitive, and complex to describe to the subject during the instruction. Engaging feedback related to brain activity are key to maintain the subject involved in the task. We build a framework where the subject controls a robotic arm both by gaze and brain activity in an enriched environment using eye tracking glasses and electro-encephalography (EEG).
In our study, we tackle the important question of the preferable moment to perform the MI task in the context of the robotic arm control. To answer this question, we design a protocol where subjects are placed in front of the robotic arm and choose with gaze which object to seize. Then based on stimuli blended in an augmented table, the subjects perform MI or resting state tasks. The stimuli consist of a red (MI) or blue (Resting) dot circling the object to seize. At the end of a MI task, the hand should close. There are three strategies corresponding to three different

moments when to perform the mental task, 1) after the robot's movement towards the object, 2) before the robot's movement, 3) during the robot's movement. The experimentation is split into a calibration and two control phases, in the calibration phase, the hand always close during MI task, and in the control phases, it relies on the subject's brain activity.
We used power spectral density estimates using Burg Auto regressive method to differentiate between MI and resting state in the alpha and beta bands (8-35 Hz).
Our method to compare the strategies relies on classification performance (LDA 2 classes) using sensitivity scores and statistical differences between conditions (R-squared map).
The early results on the first 10 subjects only showed significant differences between strategy 1 and 2 in terms of offline classification using SVM with RBF kernel ($p < 0.01$ on sensitivity, 2-way permuted ANOVA test) and online performance scores. In all subjects this difference was accompanied by a significant brain activity localized in the sensorimotor cortex ($p < 0.01$ between MI and resting state using Wilcoxon test). These findings indicated that providing subjects with eye-gaze control increase their sense of agency and improve the subsequent BCI accuracy.
Taken together, our results indicate that the moment when to perform a MI in a BCI task is crucial to the overall system performance.

Disclosures: **T. Venot:** None. **A. Desbois:** None. **M. Corsi:** None. **L. Hugueville:** None. **L. Saint-Bauzel:** None. **F. De Vico Fallani:** None.

Poster

549. Neuroprosthetics, Brain Computer Interfaces, and Body Machine Interfaces: Motor Learning and Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 549.05

Topic: E.05. Brain-Machine Interface

Support: Coleman Fung's Gift

Title: Transcutaneous Electricl Spinal Stimulation Fosters BCI Skill Acquisition

Authors: H. ALAWIEH, D. LIU, J. MADERA, S. KUMAR, F. RACZ, A. MAJEWICZ FEY, *J. MILLAN;
Univ. of Texas, Austin, Austin, TX

Abstract: Non-invasive brain computer interfaces (BCIs) present promising solutions for patients with neuromuscular impairments by providing alternative interaction channels for the control of a wide range of assistive devices. BCIs are commonly controlled through the imagination of limb movements - motor imagery (MI), which relies on voluntarily modulated sensorimotor rhythms (SMRs) that can be detected noninvasively from brain signals and then translated into external commands. However, a major challenge in controlling MI-based BCIs is the instability of SMRs, which require feedback training for MI skill acquisition. Recent research

has shown that coupling BCI training with cortical or peripheral electrical stimulation leads to neurophysiological changes that correlate with enhanced MI skills. While such approaches aimed at increasing the excitability of the sensorimotor networks within the corticospinal tract, less focus has been given to the inhibitory circuits involved in MI. As an emerging stimulation technique, Transcutaneous Electrical Spinal Stimulation (TESS) has shown excitatory effects at the spinal level and inhibitory effects at the cortical level. It has also demonstrated great potential in motor rehabilitation and functional recovery. We show that incorporating TESS in BCI training fosters MI skill acquisition and leads to better and faster BCI control. Within a controlled BCI study, 10 out of 20 participants received 20 minutes of TESS before each MI session in a five-days training protocol. We found significant improvement in BCI performance, measured by the percentage of time participants correctly control the interface, after two training sessions in the TESS group ($p < 0.001$, n=40, Linear Mixed-effect (LME) modeling) opposed to four sessions in the control ($p < 0.005$, n=40, LME). Notably, the TESS group had significantly better performance compared to the control starting on the second training session ($p < 0.001$, n=40) until the end of training (TESS: 74.4% ± 13.5%, Control: 62.9% ± 19.5%, $p < 0.01$, n=40, LME). The performance in the TESS group was also correlated with evidence of increased focal activation and functional reorganization in cortical networks, which is consistent with the expected focal activation/surround inhibition phenomenon during MI. In contrast to previous works, our findings highlight the role of inhibition in MI skill acquisition. We propose that using TESS to induce cortical inhibition before focally activating the motor cortex with MI promotes the acquisition of the MI skill for better BCI control. This not only has implications for replacing lost functions, but also for rehabilitation.

Disclosures: H. Alawieh: None. **D. Liu:** None. **J. Madera:** None. **S. Kumar:** None. **F. Racz:** None. **A. Majewicz Fey:** None. **J. Millan:** F. Consulting Fees (e.g., advisory boards); MindMaze.

Poster

549. Neuroprosthetics, Brain Computer Interfaces, and Body Machine Interfaces: Motor Learning and Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 549.06

Topic: E.05. Brain-Machine Interface

Title: The development of a corticomuscular coherence brain-computer interface to treat impairments in sensorimotor control

Authors: *S. FOGLIA[1], H. SHANTHANNA[2], Z. GAO[3], A. J. NELSON[4];
[1]Sch. of Biomed. Engin., [2]Fac. of Hlth. Sci., [3]W Booth Sch. of Engin. Practice and Technol., [4]Kinesiology, McMaster Univ., Hamilton, ON, Canada

Abstract: Human motor control is predicated on the ability for the motor system to generate efferent commands that travel to a specific muscle as well as receive afferent feedback

throughout a movement. Following a neurological injury or trauma however, there may be disruption to this feedback cycle which manifests as impairment in sensorimotor control during motor actions. The goal of this project was to develop a novel brain-computer interface (BCI) rehabilitation technology that records cortical and muscle activity and provides this as neurofeedback to individuals using a visual display to improve sensorimotor control following neurological injury. Simultaneous recording of electroencephalography (EEG) and electromyography (EMG) allows for the calculation of corticomuscular coherence (CMC) and can be used as neurofeedback reflecting both cortical and muscular involvement in the real-time control of movement. CMC is suggested to reflect the flow of information from the sensorimotor cortex to the muscle, as well as feedback from the limb back to the brain. CMC has been used to assess improvement during post-stroke motor recovery, however, its use as a method of rehabilitation has not yet been explored. In this BCI application, EMG and EEG signals are simultaneously recorded during a movement and processed to remove noise and increase feature separation between the two signals. Specifically, the mu band is extracted from the EEG signal from channels positioned over the contralateral sensorimotor cortex and surface EMG from the enervated muscle. CMC is calculated using the correlation between band-limited power time-courses method. This method computes the correlation between event related desynchronization observed in EEG and the increase in power in EMG during a movement. This method allows for CMC to be provided as neurofeedback after each trial as traditional methods for estimating CMC from single trial data can be inaccurate. The calculated CMC value is the neurofeedback that is presented to the participant as a correlation (0 to 100%) after each action. This feedback allows participants to engage in and modify cortical processes implicated in sensorimotor control that are not normally perceptible. CMC can be intentionally modulated through different mental strategies such as mental imagery and attention, which can improve synchronization between the brain and active muscle. This novel technology may have beneficial impact as a rehabilitation tool in clinical populations such as stroke, incomplete spinal cord injury, and chronic pain to improve sensorimotor control and improve activities of daily living and quality of life.

Disclosures: **S. Foglia:** None. **H. Shanthanna:** None. **Z. Gao:** None. **A.J. Nelson:** None.

Poster

549. Neuroprosthetics, Brain Computer Interfaces, and Body Machine Interfaces: Motor Learning and Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 549.07

Topic: E.05. Brain-Machine Interface

Support: This work was supported by the National Research Foundation of Korea (NRF) through the Korea Government [Ministry of Science and ICT (MSIT)] under Grant NRF-2022R1A2B5B01001443

Title: Brain computer interface based on action observation and peripheral electrical stimulation to enhance sensorimotor cortical activation in stroke patients

Authors: H. LIM[1], Y. KANG[2], *J. KU[1];
[1]Keimyung Univ., Daegu, Korea, Republic of; [2]Nowon Eulji Med. Ctr., Seoul, Korea, Republic of

Abstract: Action observation (AO) and peripheral electrical stimulation (PES) are conventional methods used to enhance rehabilitation outcomes by promoting neural plasticity. In providing AO and PES simultaneously, one's attentional state could affect the brain plasticity. Brain-computer interface (BCI) is one of promising techniques which can enable the identification of subjects' intentions and attentional state so that it could provide them with brain state-dependent feedback. In this study, we assessed the effects of attentional state-dependent feedback in the combined application of BCI-AO with PES on sensorimotor cortical activation in patients after stroke. Our approach involved showing all participants a video with repetitive grasping actions under two different tasks. One condition has feedback stimulation controlled by BCI and other conditions have feedback stimulation provided regardless of subjects' attentional state. There were significant differences in mu suppression in the affected motor and frontal cortices in stroke patients between two conditions. The involvement of both frontal and motor cortices became prominent in BCI-AO+triggered PES tasks in which feedback was given to patients according to their attentive watching, while conditions in which the condition in which the stimulation was not controlled showed activations only in the occipital lobe which processes visual information. Our findings suggest that synchronous stimulation according to patient attention is important in combining BCI-AO feedback with PES for the neurorehabilitation of stroke patients. BCI-AO feedback combined with PES is shown to be more effective for facilitating sensorimotor cortical activation in the affected hemispheres of stroke patients.

Disclosures: H. Lim: None. **Y. Kang:** None. **J. Ku:** None.

Poster

549. Neuroprosthetics, Brain Computer Interfaces, and Body Machine Interfaces: Motor Learning and Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 549.08

Topic: E.05. Brain-Machine Interface

Support: R01NS105646
R01NS123378
R01NS117568
P50HD105353
1R01NS105 646-01A1
RC1MH090912-01
T32GM008692

Title: Stroke survivor reported improvements in impaired hand function following multimodal BCI-FES intervention for upper extremity motor rehabilitation

Authors: *T. HOSSEINI[1], A. B. REMSIK[1], P. VAN KAN[1], N. ADLURU[1], V. A. NAIR[2], J. C. WILLIAMS[3], V. PRABHAKARAN[4];
[1]Univ. of Wisconsin-Madison, Madison, WI; [2]UW-MADISON, Madison, WI; [3]Arizona State Univ., Tempe, AZ; [4]Dept Neurosci, Univ. of Wisconsin, Madison, WI

Abstract: Objective Stroke often results in upper extremity motor impairment which can be characterized by a loss of function and capacity in one or both hands. Stroke-impaired hand strength and quality of function are deficits that negatively impact activities of daily living for survivors, limiting their independence. Multimodal BCI-FES interventions show efficacy in rehabilitating stroke-impaired hand function. This analysis seeks to determine if multimodal BCI-FES intervention is able to deliver perceivable improvements in hand function relative to a time-matched control group of stroke survivors. **Methods** Stroke survivors were randomized to either an immediate BCI-FES intervention group (n = 35, 16 females, mean age 62 yrs±12.8 yrs) where they completed <30 hours of multimodal BCI-FES intervention, or to a waitlist control (n= 21, 9 females, mean age 59.4 yrs±14.2 yrs) that was time matched to the intervention phase, after which the stroke control participants crossed over into the intervention phase. All were assessed with behavioral measures at baseline (M1), midpoint of intervention (M2), and at completion (M3) that included the Stroke Impact Scale-strength (SIS-S) domain and the Motor Activity Log's How Well scale (MAL-HW). Means at each timepoint were calculated for each phase and compared using independent samples t-tests. **Results** There were greater mean scores at each timepoint in intervention compared to controls in SIS-S and MAL-HW and greater mean change for intervention compared to the waitlist control in both measures (SIS-S intervention mean difference = 0.257, SD=±0.78, SIS-S waitlist control mean difference = 0.1904, SD=±1.078) (MAL-HW intervention mean difference = 2.91, SD=±0.801, MAL-HW waitlist control mean difference = 2.29, SD=±2.702) though mean change over time in either measure was not significantly different between the groups. The mean differences over time (M1 to M3) between the groups were not significant in either the SIS-S or the MAL-HW measures. Mean differences at completion between the intervention and waitlist control groups were also recorded in SIS-S (mean difference = 0.11, SD=±0.13) and MAL-HW (mean difference = 2.27, SD=±1.95), however, neither difference reached significance. **Discussion** While participants in the intervention phase may report greater improvements in perceived stroke-impaired hand function as compared to controls, the difference between means and outcome measure over time are not significant. **Conclusion** Multimodal BCI-FES can induce clinically meaningful motor recovery to the BCI-FES users but, in this sample, perceived change as well as change reported by participants are not significant.

Disclosures: T. Hosseini: None. **A.B. Remsik:** None. **P. van Kan:** None. **N. Adluru:** None. **V.A. Nair:** None. **J.C. Williams:** None. **V. Prabhakaran:** None.

Poster

549. Neuroprosthetics, Brain Computer Interfaces, and Body Machine Interfaces: Motor Learning and Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 549.09

Topic: E.05. Brain-Machine Interface

Support: American Heart Association 16IRG26960017
U.S. Army Research Office W911NNF-14-D-0005
National Institutes of Health K01HD091283

Title: Home-based Electromyography Neurofeedback System for Sensorimotor Recovery Post Stroke: A Pilot Study

Authors: *M. R. DONNELLY[1], O. MARIN-PARDO[2], C. S. PHANORD[1], K. WONG[1], S.-L. LIEW[1];
[1]Chan Div. of Occup. Sci. and Occup. Therapy, [2]Viterbi Sch. of Engin., USC, Los Angeles, CA

Abstract: Introduction: There are limited therapy options for stroke survivors with severe arm impairment. Repetitive task practice facilitates recovery, but standard therapy for severe stroke tends to yield low doses of practice. Telerehabilitation can increase the dose of therapy, but many paradigms rely on active movement, rendering them inappropriate for severe cases. Electromyography (EMG) biofeedback provides high repetition arm training in the absence of active movement and has been shown to reduce synergies and improve motor control. The purpose of this exploratory study is to present preliminary data from an at-home EMG biofeedback system (Tele-REINVENT) for severe post-stroke upper extremity hemiparesis.**Methods:** Tele-REINVENT is a neurofeedback system that acquires, processes, and transforms surface EMG signals into game activity. We piloted Tele-REINVENT with 4 chronic stroke survivors (age=56-74 years) with severe hemiparesis. Participants were asked to use Tele-REINVENT for 30, 1-hour remote sessions over 6 weeks. Signals were recorded from antagonistic muscles in the affected arm as participants attempted wrist extension in the games. The system reinforced individuated muscle activity by calculating a ratio of extensor to flexor activity and rewarding extensor recruitment in the game. Additionally, we collected clinical assessment data, including the Fugl-Meyer Assessment, Action Research Arm Test, grip strength, and range of motion (ROM) during in-person evaluations before and after the remote sessions. Finally, we also included a test of isometric muscle control during these evaluations.**Results:** Results from the pilot data suggest greater individuation of muscle activity across the 6-week trial and modest improvement on clinical assessments. Participants demonstrated improved isolated extension during the muscle control test, which was statistically significant for two of them. Furthermore, average muscle activity during the sessions improved for 3 of 4 participants; however, only 1 showed a statistically significant improvement. Finally, all participants showed improvements in different clinical assessments. However, the only improvement consistent across all participants was passive extension ROM. **Conclusion:** Tele-REINVENT is a promising telerehabilitation option to improve sensorimotor function for stroke survivors with severe hemiparesis. Despite a small sample, preliminary results suggest Tele-REINVENT is feasible and may reduce dysfunctional coactivation. Future work will examine the efficacy of Tele-REINVENT and characterize recovery patterns to determine for whom Tele-REINVENT is most appropriate.

Disclosures: M.R. Donnelly: None. **O. Marin-Pardo:** None. **C.S. Phanord:** None. **K. Wong:** None. **S. Liew:** None.

Poster

549. Neuroprosthetics, Brain Computer Interfaces, and Body Machine Interfaces: Motor Learning and Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 549.10

Topic: E.05. Brain-Machine Interface

Support: NSF Grant 1849213
Halcomb Fellowship for Dr. Haddix

Title: Prediction and confirmation of effort associated with graded finger extension in individuals with hemiparetic stroke

Authors: *M. BATES[1], C. HADDIX[1], S. GARCIA PAVA[1], E. POWELL[2], L. SAWAKI-ADAMS[2], S. SUNDERAM[1];
[1]F. Joseph Halcomb III, MD, Dept. of Biomed. Engin., [2]Dept. of Physical Med. and Rehabil., Univ. of Kentucky, Lexington, KY

Abstract: Strokes are a leading cause of lifelong disability (e.g., impaired hand function). Therefore, stroke survivors must undergo extensive therapies to attempt to regain motor control. Certain technologies are being developed to help improve these rehabilitation strategies. Brain-computer interfaces (BCIs) offer disabled individuals the means to interact with external devices by decoding electroencephalogram (EEG) signals. In the case of hand rehabilitation, integrating a BCI system with a sensor glove can provide vital information on fine motor control in individuals with large cortical lesions. Here we investigate the feasibility of predicting motor effort from the EEG associated with graded finger extension as measured by a sensor glove in stroke patients with left-hand paresis and healthy controls of similar age. Following an IRB-approved protocol, participants extended fingers of one hand in response to visual cues to one of four levels: low, medium, high, or “no-go” (i.e., none). Hand, extensor muscle, and brain activity were monitored using motion capture, electromyography (EMG), and EEG (32 channels), respectively. Event-related desynchronization (ERD) of the sensorimotor EEG was measured as the 8-30 Hz signal power at each scalp location relative to a pre-trial reference period. A quadratic classifier was trained on this ERD feature vector to predict the level of finger extension and its accuracy assessed using k-fold cross-validation in each participant. In both stroke and control groups, the ERD classifier gave 46-67% accuracy per target on either hand—much greater than chance (25%)—despite severe spasticity in the stroke-impaired hand. In contrast, an EMG power classifier gave only 34-44% accuracy per target on the stroke-impaired hand but 44-97% in the unimpaired hand and in controls. Hence, the EEG captures the effort or intent associated with finger extension regardless of success or failure in the task. The motion capture glove provided confirmation of attempted or accomplished movement. Work is ongoing to extend this protocol to include hand contraction and grip force using a custom-designed sensor glove. This system could be useful in rehabilitative BCI protocols that emphasize fine control.
Disclaimer: This work was performed while Lumy Sawaki was employed at University of

Kentucky. The opinions expressed in this article are the author's own and do not reflect the view of the National Institutes of Health, the Department of Health and Human Services, or the United States government.

Disclosures: M. Bates: None. **C. Haddix:** None. **S. Garcia Pava:** None. **E. Powell:** None. **L. Sawaki-Adams:** None. **S. Sunderam:** None.

Poster

549. Neuroprosthetics, Brain Computer Interfaces, and Body Machine Interfaces: Motor Learning and Plasticity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 549.11

Topic: E.04. Voluntary Movements

Support: European Union's Horizon 2020 research and innovation programme under the Marie Sklodowska-Curie grant agreement No 896892
NSF 2054406
NIDILRR 90REGE0005

Title: Effects of synergic practice on motor learning

Authors: *A. BELLITTO[1], M. CASADIO[1], F. A. MUSSA-IVALDI[3,4], C. PIERELLA[2,1]; [1]Dept. of Computer Science, Bioengineering, Robotics and Systems Engin., [2]Dept. of Neurology, Rehabilitation, Ophthalmology, Genetics, Maternal and Child Hlth., Univ. of Genoa, Genova, Italy; [3]Dept. of Physiol., Northwestern U./RIC, Chicago, IL; [4]Shirley Ryan Ability Lab., Chicago, IL

Abstract: Body-Machine Interfaces (BoMIs) exploit the many degrees of freedom of the body by mapping them into a smaller number of commands that are used to control a device. Due to this feature, BoMIs can be used to evaluate how a user's internal model of the body and device changes over time when learning a new skill. Studies have shown how, despite presenting subjects with an ill-posed inverse problem, through training, BoMI users converged to a stable model. So far, motor learning has been considered as a process taking place within single individuals. Here, we focus on how shared practice of a new skill by a group affects the concurrent individual and collective learning dynamics. Our aim was to characterize and compare learning as it occurs when using a BoMI solo or in a dyadic manner and also to investigate the evolution of the related internal models through time. We recruited 20 pairs of naïve participants. Each participant controlled a cursor presented on two computer monitors. Body motions captured by 4 Inertial Measurement Units, positioned on the upper arms of both users, were mapped onto the positions of the two cursors. Participants performed a reaching task and underwent 3 phases of training:1-individual, 2-social and 3-individual practice. The body-to-cursor map changed according to the type of practice. In phases 1 and 3 each participant used an individual map and had complete control of their computer cursor. In phase 2 participants shared

cursor control via a common map, and the same cursor positions were displayed on both screens. With practice, all participants learned to control the cursor. When investigating their behaviour during phase 2, we observed how, in each couple, one participant contributed more than the other to the task performance. We compared these 2 responses and, despite not noticing a difference in the individual performance either before or after social practice, we observed a difference in the evolution of their individual inverse model. In phase 2, participants who contributed less to the cursor's control adopted a new model when sharing the cursor, while those who contributed more kept a consistent model, similar to that used in the individual practice. Results indicate that synergic learning is likely to influence the evolution of the inverse model even after an initial individual practice. It remains to be established how different types of interactions guide the formation and evolution of the internal model when learning a new skill.

Disclosures: A. Bellitto: None. **M. Casadio:** None. **F.A. Mussa-Ivaldi:** None. **C. Pierella:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.01

Topic: E.06. Posture and Gait

Title: Bodyweight Unloading Influences Lower Extremity Kinematics and Muscle Activity During Walking

Authors: *C. S. LAYNE[1], C. A. MALAYA[2], A. RIAZ[2], S. CHANDRASEKARAN[2], Z. MOHIUDDIN[2];
[1]Univ. Houston, Univ. Houston, Houston, TX; [2]Ctr. for Neuromotor and Biomechanics Res., Univ. of Houston, Houston, TX

Abstract: Background: Changes in gravitational load can drive adaptations in leg speed, foot placement and pressure distribution, as well as affect joint angles and muscle activity during walking. There are still unanswered questions about how different levels of simulated gravitational loading can influence kinematics and muscle activity; in particular, it is unclear if the effects seen are specific to a certain level of unloading, or if the previous level of unloading influences the current one. **Objective:** To explore the effects of simulated gravitational unloading on kinematics and muscle activity during walking. **Methods:** 15 healthy adult participants walked for trials of 1 minute in a positive pressure unloading treadmill at a comfortable speed. Each participant experienced a series of unloading steps from 100% to 20%-unit earth gravity, in 20% increments. There was no delay between conditions. Mean and difference kinematic and electromyographic waveforms were compared using statistical parametric mapping (SPM). Phase diagrams, phase diagram areas, root mean square and integrated areas were calculated for each condition. **Results:** Testing revealed differences between conditions and for difference waveforms for both kinematics and electromyographic

variables across the hip and knee, as well as the rectus femoris and medial gastrocnemius. The ankle, anterior tibialis, and biceps femoris did not appear to be affected by the unloading. RMS, integrated areas, phase portraits and phase areas were also modified by unloading. **Conclusions**: Simulated gravitational unloading appears to modify kinematic and neuromuscular activity in the joints and muscles of the lower extremities, particularly in those resisting gravitational forces. Differences in joint angles and muscles also appear non-linear, and each joint and muscle displays a distinct pattern of adaptation to unloading.

Disclosures: C.S. Layne: None. **C.A. Malaya:** None. **A. Riaz:** None. **S. Chandrasekaran:** None. **Z. Mohiuddin:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.02

Topic: E.06. Posture and Gait

Support: NSF CRCNS 2113028
NSF NeuroNex 2015317

Title: Exploring the effects of tarsal segment morphology on insect strain sensing

Authors: *C. A. GOLDSMITH[1], W. P. ZYHOWSKI[1], S. N. ZILL[2], N. S. SZCZECINSKI[1]; [1]West Virginia Univ., Morgantown, WV; [2]Biomed. Sciences/Anatomy, J.C. Edwards Sch. Med., Huntington, WV

Abstract: Sensory feedback is a crucial part of how the nervous system controls robust and agile walking. Dynamic feedback from sense organs helps continually influence activity in the low-level motor networks and high-level control centers, which in turn dictate limb movements. Furthermore, the sensory feedback available to the nervous system is highly dependent on the mechanical structure of the organism. The foot (or tarsus) is one key part of this structure, as its morphology influences the organism's interaction with the ground, and subsequently the sensory feedback collected from load sensors. In many insects, the tarsus is comprised of several nested segments actuated by a singular elastic tendon running through the center. This allows the tarsus to both passively deform to the ground and actively grip onto it when the tendon is pulled. Additionally, tarsal segments contain groupings of campaniform sensilla (CS), which measure the strain of the cuticle. These strain measurements, in addition to those collected by CS in various locations on the limbs, provide load feedback to the nervous system to influence stepping motions by activating muscle synergies that resist such loads. Due to the nature of strain, these sensors are influenced both by external forces as well as the internal limb structure (e.g., its geometry and compliance).
In order to further elucidate the interplay between tarsal structure and CS readings in the insect nervous system, we developed tarsi with different degrees of biomimetic structure, compliance,

substrate gripping capability, and sensing, and tested them on our physical robotic limb performing stepping. We have previously developed a physical robotic model of a stick insect leg with biomimetic strain sensing in reported CS locations on the limbs in order to replicate the sensory feedback available to the nervous system. However, our previous data collection with this model utilized a simplified, semi-spherical, rigid tarsal segment. Our new experiments enable us to extract strain readings analogous to CS discharge for differing tarsal functionality, which can be used to infer what sensory information is available to the nervous system during walking in different conditions. These readings can then be used to explore how active substrate grip could be controlled by the nervous system by developing a synthetic nervous system controller that relies on strain feedback to modify stepping. Such a controller utilizes dynamical neurons arranged in morphological configurations, so its structure aids in understanding how the nervous system may integrate sensory information to control stepping.

Disclosures: C.A. Goldsmith: None. **W.P. Zyhowski:** None. **S.N. Zill:** None. **N.S. Szczecinski:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.03

Topic: E.06. Posture and Gait

Title: Effects of motion display rate and coherence on balancing a visual inverted pendulum

Authors: *P. DIZIO[1], H. N. DAVIES[1,2,3];
[1]Volen Ctr. for Complex Systems, Brandeis Univ., Waltham, MA; [2]Marcus Autism Ctr., Children's Healthcare of Atlanta, Atlanta, GA; [3]Dept. of Pediatrics, Emory Univ. Sch. of Med., Atlanta, GA

Abstract: Manually keeping a visual inverted pendulum (VIP) upright requires dynamic visual information coupled to rapid selection and execution of motor commands. Current models of human VIP balancing focus on continuous or intermittent visual feedback about pendulum state (position and velocity), and this study is the first to differentiate the roles of visual streams of information about motion, orientation, and their combination in the perception-action cycle for balancing. In this VIP paradigm, a circular random dot kinematogram (RDK) composed of 100 dots, subtending 10.3° visual angle, underwent simple inverted pendulum roll motion ($\ddot{\Theta}=K_P \sin\Theta$) about its fixed center. Left-right joystick deflections accelerated the RDK ipsilaterally. Subjects (N=32) were instructed to keep the RDK as upright and stable as possible without exceeding programed fall limits at ±180°. The pendulum constant, K_P, was individually calibrated to produce about 10 falls in the first trial, and then subjects completed 36 30-sec trials in 3 randomized conditions: 1) 50 Hz frame rate with 50% coherent dot motion, activating first-order, low-level retinal motion detectors, 2) 8 Hz strobe rate with 100% coherence, activating long-range orientation and motion perception, and 3) 50 Hz with 100% coherence, activating

both the low- and high-level systems. VIP performance was best (fewest falls and lowest sway velocity) in the naturalistic condition which provided visual orientation and motion information via low- and high-level pathways. The 50% coherent condition (only low-level motion information) showed a significant performance decrement, and the 100% coherent 8 Hz strobe condition (no low-level motion information) showed an additional significant decrement. Performance improvement over repetitions was inversely proportional to the average performance per condition. The incidence of destabilizing joystick commands (which accelerate the pendulum toward the nearest fall boundary) varied in parallel with performance under the different visual conditions, and with learning. Reactive and anticipatory joystick commands were significantly less frequent in the condition which produced the worst performance - 8 Hz, 100% coherent - than the other two conditions. A second experiment varied the strobe rate of 100% coherent RDKs, and showed a significant exponential, asymptotic improvement in performance from 5 to 8 Hz. Conclusion: Loss of high-level orientation and motion information degrades VIP balancing more than loss of low-level visual motion information. Future studies will assess the differential roles of low- and high-level pathways in single and dual task bipedal balance.

Disclosures: P. DiZio: None. **H.N. Davies:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.04

Topic: E.06. Posture and Gait

Support: NSF CRCNS 1822568

Title: Effect of Cadence on the Relationship between Center of Mass and Foot Placement

Authors: *J. GRAY[1], A. SANSARE[2], M. VAN LEEUWEN[3], S. M. BRUIJN[3], H. REIMANN[1]; [1]Kinesiology and Applied Physiol., [2]Physical Therapy, Univ. of Delaware, Newark, DE; [3]Res. Inst. MOVE, Fac. of Human Movement, Amsterdam, Netherlands

Abstract: Controlling balance while walking requires intentional foot placement based on the body's Center of Mass (CoM). Our study explored how the neural control processes of walking change with steady state walking cadence. Specifically we ask how foot placement, as a mechanism of balance regulation, changes with cadence. Prior work has shown no effect of walking speed on foot placement within a narrow range of speeds, but here we considered a wide range of walking cadences. Previous studies established a linear relationship between CoM state and next foot placement during steady state walking. We measured the slope and explanatory power of this linear relationship between CoM state and medial-lateral foot placement and analyzed changes with walking cadence. 24 young healthy participants walked at one of five designated metronome paces (50, 75, 100, 120, and 140 bpm) on a self-paced treadmill in trials of 100 strides. They completed three trials at each cadence, with order of metronome pace

randomized within blocks of five trials. We measured kinematic data from 44 markers in Qualisys Motion Capture, kinematic data from force plates captured kinetic data. and calculated the whole-body CoM position relative to stance foot ankle. We found high R^2 values across all cadences in our data, which confirms that these linear models have high predictive value within the range of cadences studied. We found a statistically significant decrease in slopes of the linear model relating CoM state to foot placement with increasing cadence. The decrease in slopes with increasing cadence suggests that neural processes change with walking speed or stepping frequency. In particular, this suggests that feedback plays a lesser role at higher walking speeds. Our results confirm that foot placement can be predicted by the COM state across a range of walking cadences as neural control parameters change with pace. However, clinical populations with slower paces around 50bpm may not have a clear relationship between CoM state and foot placement, which suggests that they may use other balance mechanisms to regulate balance during walking.

Disclosures: J. Gray: None. **A. Sansare:** None. **M. van Leeuwen:** None. **S.M. Bruijn:** None. **H. Reimann:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.05

Title: WITHDRAWN

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.06

Topic: E.06. Posture and Gait

Support: NIH Grant NS110550
NIH Grant NS100928
NIH Grant HD032571
NSF Grant 2024414

Title: The role of forelimb muscle morphology in muscle motor and sensory function during locomotion in the cat

Authors: S. M. A. RAHMATI[1], A. KLISHKO[1], R. S. MARTIN[2], N. E. BUNDERSON[3], T. R. NICHOLS[1], I. A. RYBAK[4], A. FRIGON[5], T. BURKHOLDER[1], *B. I. PRILUTSKY[1];

[1]Georgia Inst. Technol., Atlanta, GA; [2]AVEVA, San Leandro, CA; [3]Autonomous Solutions, Petersboro, UT; [4]Dept. of Neurobio. and Anat., Drexel Univ., Philadelphia, PA; [5]Dept. of Pharmacology-Physiology, Univ. de Sherbrooke, Sherbrooke, QC, Canada

Abstract: Previous studies established a strong link between morphological characteristics of cat hindlimb muscles and their sensorimotor functions during locomotion. Less is known about the role of cat forelimb muscle morphology in producing motor outputs and generating length- and force-dependent sensory signals. This information is needed to understand contributions of the forelimbs to the spinal control of quadrupedal locomotion. To evaluate the role of forelimb muscle morphology in muscle sensorimotor function during locomotion, we used morphological characteristics of 40 forelimb muscles measured in 4 adult cats. These characteristics included 3D coordinates of muscle attachments; muscle physiological cross-sectional area (PCSA); sarcomere length; etc. We also recorded full-body mechanics and EMG activity of 12 forelimb muscles during level walking. For each walking cycle, we computed moment arms, muscle-tendon unit length, and resultant joint moments. Using the above information, we computed forces of 40 forelimb muscles during walking along with muscle force- and length-dependent sensory signals mapped onto the corresponding cervical spinal segments. The firing rates of muscle spindle group Ia and II and Golgi tendon group Ib afferents were computed from the muscle-tendon unit length, velocity, activation and force during walking using regression equations developed by Prochazka and Gorassini (1998). Muscle morphology contributed strongly to the estimated motor and sensory functions of forelimb muscles. For example, patterns of computed forces of forelimb muscles and the activities of group Ib afferents were affected by the muscle moment arm and PCSA of individual muscles. Muscle moment arms also strongly effected muscle-tendon unit length and velocity changes and thus activity patterns of spindle group Ia and II afferents. We identified forelimb muscles that likely trigger the swing-to-stance (spinodeltoideus, teres major) and stance-to-swing (supraspinatus, biceps brachii, extensor carpi radialis) transitions based on their length-dependent afferent activities. Similar to the hindlimb, these muscles cross mainly proximal limb joints. We conclude that forelimb muscle morphology contributes substantially to sensorimotor locomotor muscle function in the cat.

Disclosures: S.M.A. Rahmati: None. **A. Klishko:** None. **R.S. Martin:** None. **N.E. Bunderson:** None. **T.R. Nichols:** None. **I.A. Rybak:** None. **A. Frigon:** None. **T. Burkholder:** None. **B.I. Prilutsky:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.07

Topic: E.06. Posture and Gait

Support: NIH Grant R21 EB032059

Title: An ultrasound-based model for tremor suppression with peripheral electrical stimulation

Authors: A. IYER[1], X. XUE[2], D. ROQUE[3], *N. SHARMA[1];
[1]North Carolina State Univ., Raleigh, NC; [2]North Carolina State Univ., RALEIGH, NC; [3]Univ. of North Carolina Chapel Hill, Chapel Hill, NC

Abstract: Tremor is a neuromuscular disorder that leads to involuntary oscillations of the upper limb in persons with Parkinson's disease (PD) and Essential Tremor (ET). These oscillations significantly hinder well-being when performing activities of daily living. Current medications for tremor suppression have limited efficacy. Surgical treatments such as deep brain stimulation and high-intensity focused ultrasound, although effective, risk side effects due to permanent damage to the healthy brain tissue. Recently non-invasive peripheral nerve stimulation has shown promising results. It is devoid of side effects, and one such method has even garnered FDA approval for transient reduction of moderate to severe tremors. However, there is a limited understanding of the mechanisms involved in tremor suppression due to the electrical stimulation. Improved modeling of the involved mechanisms and closed-loop algorithms based on these models can determine the correct stimulation dosage and timing. Sensing modalities to measure tremors include inertial measurement units (IMU), motion capture, and electromyography (EMG). The data collected from these sensors is crucial for the creation of a mathematical model of tremor. However, these modalities have inherent limitations. IMUs measure limb kinematics during the tremor but cannot visualize tremor-causing muscles thus providing limited insight into tremors at the neuromuscular level. EMG is affected by stimulation artifacts during high frequency electrical stimulation making it difficult to determine the tremor signals during electrical stimulation. We believe ultrasound (US) imaging is a novel tremor sensing method to accurately model and assess tremor severity as it can provide direct visualization of the tremor-causing muscles. We have previously shown that US can detect the tremor frequency of participants with PD and ET. To expand the study, we collected US images and EMG and IMU signals from three groups of human participants (ET, PD, and healthy control). Participants performed a grasping motion while we used a range of sensory stimulation parameters to model the tremor behavior. Our preliminary results show that a data-driven model based on US-derived tissue position and velocity measurements can predict the wrist joint position during a tremor, within 3.5% of the wrist position measured by the IMU. The preliminary analysis and electrical stimulation testing show that US imaging provides vital information to create models of tremors on the muscular level and has the potential to be used as a feedback signal in closed-loop control for sensory stimulation.

Disclosures: A. Iyer: None. **X. Xue:** None. **D. Roque:** None. **N. Sharma:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.08

Topic: E.06. Posture and Gait

Support: PF-JFA-2036

Title: The Condition for Dynamic Stability in Walking with Foot Placement Control

Authors: *H. REIMANN;
Univ. of Delaware, Newark, DE

Abstract: The upright human body is mechanically unstable. The nervous system must continuously regulate motor activity based on sensory information to prevent loss of balance. This sensorimotor control process has been studied intensively in standing, where balance is largely regulated by continuous modulation of ankle musculature, changing the center of pressure under the feet. Stability in standing can be quantified in purely biomechanical terms by the Margin of Stability, which is proportional to the impulse the standing body is able to withstand without having to take a step. In walking, this principle is less meaningful, because stepping is a regular and necessary component of locomotion. Our research goal is to characterize the stability of the walking human body in a meaningful way. One way to regulate balance in walking is to modulate the location of the foot placement at each step based on the kinematic state of the center of mass. Human experiments show that the foot placement location can be reliably predicted from the kinematic state of the the center of mass at midstance, indicating that humans do use feedback control of foot placement for balance during walking. The behavior of this feedback-controlled system, including stability in the sense of robustness against perturbations, depends both on the biomechanics of the body and the specific details of the feedback control law for foot placement. In this work, we model human walking as a linearized single-link inverted pendulum controlled by taking a step after a fixed time to a location determined by a proportional-derivative control law based on the position and velocity of the center of mass at midstance. We show that for each combination of proportional and derivative control gains, this system has a periodic limit cycle solution corresponding to a gait at a stepping frequency determined by the step time parameter. To characterize the stability of the system, we analyze the transition matrix that represents how small perturbations at midstance are carried over into the next step. This allows us to determine which combinations of control parameters result in a stable gait in the sense that the system will eventually return to the limit cycle after a perturbation. We find that the control gains for which the system is stable depend on the stepping frequency, with slow-paced gaits requiring higher control gains for foot placement. The model result implies that humans have to adjust feedback gains for balance control during walking depending on the walking cadence.

Disclosures: H. Reimann: None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.09

Topic: E.06. Posture and Gait

Support: NSERC RGPIN-2017-04175

Title: Light touch reduces, but does not eliminate, sway evoked by a rapid visual disturbance during standing.

Authors: *J. E. MISIASZEK[1,2], S. HEMAKUMARA[3], J. FORERO[3], K. K. FENRICH[3,2];
[1]Occup. Therapy, [2]Neurosci. and Mental Hlth. Inst., [3]Fac. of Rehabil. Med., Univ. of Alberta, Edmonton, AB, Canada

Abstract: Postural sway increases when standing with eyes closed, highlighting the importance of visual inputs to balance control. Moreover, movement of the visual surround entrains sway to an oscillating stimulus or induces lean with a persistent stimulus. These effects are reduced when touching a stable reference. However, the effect of a short, rapid visual field displacement (VFD) on standing balance is not yet known. In this study, we tested the effect of a short, but rapid VFD on standing balance with or without touching of a stable reference. We hypothesized that participants would sway in the direction of the VFD and that touch would reduce the effect. To test this, participants stood on a foam pad atop a force plate, surrounded by an enclosure mounted on a set of rails. The surround was driven by a motor to produced a 2.5 cm linear displacement with a peak velocity of 124 mm/s. A total of 40 naïve participants (4 groups of 10), received either forward or backward VFD, with or without touch of a stable reference. Responses to the VFD were measured as 1) the anterior-posterior sway in the center of pressure (COPap) data, 2) ankle joint motion, and 3) electromyographic (EMG) activity of leg muscles. VFD in either direction produced a biphasic displacement of the COPap, with the initial, smaller deflection moving the opposite direction of the VFD. The larger second deflection moved in the same direction as the VFD. A similar biphasic pattern was observed in the ankle goniometer trace. Despite the consistent behaviour observed across participants in the mechanical data, the EMG data did not reveal a consistent pattern of activation across, or within participants. The amplitudes of the first and second peaks in the COPap deflections were reduced with light touch, for both forward and backward VFD. In contrast, whereas the amplitude of the initial peak habituated with repeated stimuli in both directions, the amplitude of the second peak habituated with repeated backward stimuli, but not forward stimuli. Overall, short, rapid VFD produced a long-latency postural reaction in the direction of the VFD, consistent with previous reports. The shorter-latency deflection in the COPap and ankle goniometer data are likely due to postural changes needed to allow the force necessary to propel the body in the direction of the stimulus. Touching a stable reference reduced, but did not eliminate, the visually evoked sway. This indicates that the potentially threatening stimulus introduced by the VFD is difficult to ignore, despite the lack of corroborating sensory inputs from other sources.

Disclosures: J.E. Misiaszek: None. **S. Hemakumara:** None. **J. Forero:** None. **K.K. Fenrich:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.10

Topic: E.06. Posture and Gait

Support: NIH Grant R01 NS115900
NIH Grant R01 NS112304
NIH Grant R01 NS100928
NIH Grant R01 NS110550
NSF Grant 2113069

Title: Comparative investigation of control mechanisms of locomotion and turning using a simulated quadrupedal robot

Authors: *A. B. LOCKHART[1], M. A. PARRILLA[2], S. N. MARKIN[1], S. TATA RAMALINGASETTY[1], N. A. SHEVTSOVA[1], Y. I. MOLKOV[3], J. AUSBORN[1], I. A. RYBAK[1], S. M. DANNER[1];
[1]Dept. of Neurobio. and Anat., Drexel Univ. Col. of Med., Philadelphia, PA; [2]Dept. of Physics, Univ. of Salamanca, Salamanca, Spain; [3]Dept. of Mathematics and Statistics, Georgia State Univ., Atlanta, GA

Abstract: Locomotion allows animals to move in the world. The underlying neuronal control involves rhythm-generating and limb-coordinating circuitry in the spinal cord that integrates afferent feedback to execute descending commands. Most of what we know about the neuronal control of locomotion pertains to straightforward locomotion; how turning or the change in direction is controlled, remains poorly understood. Various left-right asymmetries could lead to changes in movement direction, but it is not clear which turning strategies optimally maintain stability and how these strategies depend on speed, gait, and environmental conditions. Here, we present a simulation of a quadrupedal robot controlled by a model of the spinal locomotor circuits to study various mechanisms of turning. Using a robot allows us to study the impact of these turning strategies on stability and balance. The quadrupedal robot model has 13 degrees of freedom: three joints per limb and one joint within the torso that allows for lateral bending of the body. The neuronal model controlling the robot includes four coupled rhythm generators, each controlling one limb. These rhythm generators receive sensory feedback that characterizes loading and extension of the corresponding limb. The robot model was able to locomote at different speeds and gaits while exhibiting speed-dependent changes of stance and swing phase durations. Furthermore, the model was able to adapt to changes in the environment and external perturbations. We studied the effectiveness of left-right asymmetric changes in locomotor frequency, duty factor, stride length, medio-lateral paw placement, and body bending to induce turning at different speeds and gaits. Our model predicts that each turning strategy leads to a change in direction but differently affects the stability of locomotion. Lastly, our model suggests that a combination of strategies is needed to effectively turn while maintaining stability, and that the optimal strategy depends on the locomotor gait and speed. Thus, control of turning likely involves task- and speed-dependent modulation of the spinal neuronal circuits at multiple levels. We are currently working on a physical robot that will allow us to validate our simulation results in the future.

Disclosures: A.B. Lockhart: None. **M.A. Parrilla:** None. **S.N. Markin:** None. **S. Tata Ramalingasetty:** None. **N.A. Shevtsova:** None. **Y.I. Molkov:** None. **J. Ausborn:** None. **I.A. Rybak:** None. **S.M. Danner:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.11

Topic: E.06. Posture and Gait

Support: NIH Grant R01 NS100928
NIH Grant R01 NS110550
NIH Grant R01 NS112304
NIH Grant R01 NS115900
NSF Grant 2113069

Title: On the role of sensory feedback and central neuronal interactions in locomotor gait and balance control in quadrupeds

Authors: G. YU[1], S. N. MARKIN[3], N. A. SHEVTSOVA[3], S. M. DANNER[3], *I. A. RYBAK[3], Y. I. MOLKOV[1,2];
[1]Dept. of Mathematics and Statistics, [2]Neurosci. Inst., Georgia State Univ., Atlanta, GA; [3]Dept. of Neurobio. and Anat., Drexel Univ., Philadelphia, PA

Abstract: Locomotion in quadrupeds is a complex process that involves coordination of movements of four limbs actuated by numerous muscles. This coordination defines locomotor gait and ensures postural stability during locomotion at the desired speed and direction, and their changes. Due to the complexity of the system, researchers often resort to studying locomotion in simplified quadrupedal robots and mathematical models which allows them to limit the system's complexity and focus on general principles.
The basic pattern of limb movements during locomotion is produced by rhythm-generating neural circuits within the spinal cord controlling each limb. Interlimb coordination is provided in part by central interactions between these circuits. The rhythm-generating and limb-coordinating spinal circuits represent the central neural controller, whose operation is modulated by sensory feedback from the limbs that informs the central controller about the state and position of the limbs and the body (posture). The interactions between the central controller and sensory feedback and their role in locomotor gait and posture adjustment during locomotion are poorly understood. In this study, we address these issues by combining some fundamental findings from the biomechanical studies on quadrupedal locomotion and recent discoveries on neural interactions within the spinal locomotor circuitry in the form of a tractable mathematical model. The model is minimalistically designed to account for feedback signals arising from loading and extending each limb, as well as postural stability. The central controller in the model includes a minimal model of the locomotor rhythm generators whose interconnections and inputs have clear interpretation in terms of specific neuronal interactions and sensory signals. We systematically explore the model behavior in a range of locomotor speeds and test it against existing experimental data from mouse locomotion. Our model predicts that in freely moving mice, some of the locomotor phase transitions are triggered by specific feedback signals while other

transitions are controlled by the central interactions. Moreover, the mechanisms of these transitions may change with speed and gait. Our model can be scaled for other quadrupedal animals and be used as a convenient theoretical framework that couples neural dynamics with mathematically tractable biomechanics.

Disclosures: G. Yu: None. **S.N. Markin:** None. **N.A. Shevtsova:** None. **S.M. Danner:** None. **I.A. Rybak:** None. **Y.I. Molkov:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.12

Topic: E.06. Posture and Gait

Support: NIH Grant R01 NS115900
NIH Grant R01 NS112304
NIH Grant R01 NS100928
NIH Grant R01 NS110550
NSF Grant 2113069

Title: Studying interactions between somatosensory feedback and spinal circuitry during locomotion using a mouse neuromuscular model

Authors: *S. TATA RAMALINGASETTY[1], S. N. MARKIN[1], A. B. LOCKHART[1], N. A. SHEVTSOVA[1], J. ARREGUIT[2], A. J. IJSPEERT[2], I. A. RYBAK[1], S. M. DANNER[1]; [1]Dept. of Neurobio. and Anat., Drexel Univ. Col. of Med., Philadelphia, PA; [2]Inst. of Bioengineering, École Polytechnique Fédérale de Lausanne, Lausanne, Switzerland

Abstract: Locomotion in animals results from a complex interplay between neuronal circuitry in the spinal cord, descending supra-spinal signals, the musculoskeletal system, and sensory feedback. With the advent of modern molecular-genetic tools, significant strides have been made in identifying specific neuron types in the spinal cord and brainstem involved in different aspects of locomotion. However, how somatosensory feedback interacts with the spinal circuitry to provide stable, adaptive locomotion remains poorly understood. Here, we present a neuromuscular model of mouse locomotion and use it to explore interactions between the spinal circuitry and afferent feedback and their role in the control of locomotion. We adapted the previously developed three-dimensional (3D) musculoskeletal model of the mouse and integrated it with a model of spinal locomotor circuitry. Motor outputs of the spinal circuit model was used to drive muscles of the biomechanical model; proprioceptive (spindle Ia and II, and Golgi tendon Ib) and exteroceptive feedback signals provided sensory feedback to the spinal circuit model. The spinal circuit model included rhythm generators, each controlling one limb, that defined the locomotor frequency and flexor-extensor alternation. The rhythm generators controlled the pattern formation circuits which generated muscle synergies and created muscle-specific

activation patterns. Afferent feedback was connected to the rhythm generators (affecting timing of phase-transitions), to the pattern formation circuits (affecting muscle synergies), and directly to the motoneurons, forming basic reflex circuits. The closed-loop neuromuscular interactions were simulated in a 3D physics environment (MuJoCo). The model reproduced speed-dependent gait expression of mice while being able to adapt to different changes in the environment. We systematically studied the influence of somatosensory afferent pathways by manipulating them during different locomotor behaviors. Our results suggest that task-dependent modulation of afferent feedback is essential for robust changes of speed and gait. In the future, we will extend our model to simulate neuromuscular control of 3D-quadrupedal locomotion with the aim to provide an open-source model that can be used as a testbed to explore different hypotheses on the interactions between spinal circuits and sensory feedback and the role of feedback in the control of locomotion under different conditions.

Disclosures: S. Tata Ramalingasetty: None. **S.N. Markin:** None. **A.B. Lockhart:** None. **N.A. Shevtsova:** None. **J. Arreguit:** None. **A.J. Ijspeert:** None. **I.A. Rybak:** None. **S.M. Danner:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.13

Topic: E.06. Posture and Gait

Support: KAKENHI: 18H03141
KAKENHI: 18H04038
KAKENHI: 18H04958
KAKENHI: 18H05287
KAKENHI: 20H05714

Title: Stimulus intensity- and location-specific activation of human locomotor circuitry by non-invasive transvertebral magnetic stimulation

Authors: K. KAWAI[1], **T. TAZOE**[2], *Y. NISHIMURA[2];
[1]Col. of Sports Sci., Nihon Univ., Tokyo, Japan; [2]Tokyo Metropolitan Inst. of Med. Sci., Tokyo, Japan

Abstract: Transvertebral magnetic stimulation (TVMS) of the human lumbar spinal cord can evoke bilateral rhythmic leg movement as walking, supposedly trough the activation of spinal locomotor circuitry. However, an appropriate stimulus condition to effectively evoked walking movement remains to be elucidated. As spinal motoneurons innervating leg muscles that engage in walking movement are present in the entire lumbosacral spinal cord, the change of stimulus intensity or stimulus location may induce ununiform patterns of activations in spinal motor network and then modulate the pattern of evoked leg movement. Here, we systematically

investigated the effects of stimulus intensity and location on the bilateral leg movements induced by non-invasive TVMS. Neurologically healthy adult participants were tested in semi-prone posture on a bed with both legs suspended from the ceiling and received the bursts of TVMS via a closed-loop paradigm while legs were fully relaxed. In the first experiment, TVMS was delivered over an intervertebral space of the lumbar cord (L1-L2) at the stimulus intensity from 10% to 70% of maximum stimulator output. In the second experiment, a fixed intensity of TVMS was given to a 6 by 3 stimulation target grid over the back that covered 6 intervertebral spaces from T11 to L5 and ~3 cm left to right from the midline. The rhythmically controlled bursts of TVMS induced two major patterns of cyclic leg movements in which left-right movement cycles were coordinated with different phase relationship; hopping-like movements, in which both legs moved in the same direction in phase, and walking-like movements, in which both legs moved alternatively in anti-phase. In experiment 1, the stimulus-evoked rhythmic movements changed from the hopping-like movements to walking-like movements as stimulus intensity was increased. In experiment 2, the walking-like movement were evoked when TVMS was given at around T12-L2, whereas the hopping-like movement were mostly evoked when TVMS was given to either T11-T12 or L2-L5. We demonstrated that TVMS induced different patterns of cyclic leg movement depending on the stimulus intensity and location. These results suggest that TVMS activates distinct neural modules in the human spinal cord to generate hopping- and walking-like movements.

Disclosures: **K. Kawai:** None. **T. Tazoe:** None. **Y. Nishimura:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.14

Topic: E.06. Posture and Gait

Support: KAKENHI: 18H03141
KAKENHI: 18H04038
KAKENHI: 18H04958
KAKENHI: 18H05287
KAKENHI: 20H05714

Title: Reorganization of human spinal locomotor circuitry after spinal cord injury

Authors: *T. TAZOE[1], T. MURAYAMA[2], T. TOSAKA[2], M. KANESHIGE[1], M. SUZUKI[1], N. KIKUCHI[3], Y. UGAWA[4], Y. NISHIMURA[1];
[1]Neural Prosthetics Project, Tokyo Metropolitan Inst. of Med. Sci., Tokyo, Japan; [2]Dept. of Rehabil. Treatment, [3]Dept. of Rehabil. Med., Chiba Rehabil. Ctr., Chiba, Japan; [4]Dept. of Human Neurophysiology, Sch. of Med., Fukushima Med. Univ., Fukushima, Japan

Abstract: Bipedal human walking is mediated by spinal locomotor circuitry in the lumbar cord. As transvertebral magnetic stimulation (TVMS) to human lumbar cord is capable of inducing a cyclic alternative leg movement as in walking, it is presumed that TVMS activates a neural module involved in the spinal locomotor circuitry to induce walking movement. Using TVMS technique, here, we aimed to discover potential adaptive changes in the spinal locomotor circuitry caused by spinal cord injury (SCI). Thirteen patients with chronic SCI and nine intact controls were participated in the experiment. Six out of thirteen patients and the others were clinically diagnosed as incomplete and complete loss of lower extremities sensorimotor functions, respectively. The participants were in semi-prone posture on a bed with both legs suspended from the ceiling and received the bursts of TVMS via a closed-loop paradigm while legs were fully relaxed. A 6 by 3 stimulation target grid was arranged over the back that covered 6 intervertebral spaces from T11 to L5 and ~3 cm left to right from the midline. The rhythmically controlled bursts of TVMS induced the cyclic bilateral leg movements in all participants. Notably, the phase relationship between the left and right leg cycles was changed with shifting the stimulus site in all participants. However, the number of sites inducing the walking-like, left-right alternative leg movement was different among the groups. In controls and incomplete SCI, the stimulus sites inducing walking-like movement were confined to the intervertebral spaces between T11-L2. TVMS to the other sites mostly induced the hopping-like, bilateral cyclic movement in-phase. In contrast, the walking-like movements were induced at border sites in complete SCI. The complete SCI patients had more stimulus sites inducing the walking-like movements than incomplete SCI patients or controls (complete SCI=5.0±3.2; incomplete SCI=1.2±1.2; controls=2.4±1.2), whereas the stimulus sites inducing the hopping-like movements were less in complete SCI patients (complete SCI=2.9±2.5; incomplete SCI=8.2±2.7; controls=11.3±3.5). We demonstrated that the disconnection of neural pathways between the brain and the lumbar cord lead a new spinal locomotor circuitry in the humans. This new circuitry may be the primitive spinal locomotor circuitry unmasked by the SCI which disconnected the spinal locomotor circuitry with the descending commands from the brain.

Disclosures: **T. Tazoe:** None. **T. Murayama:** None. **T. Tosaka:** None. **M. Kaneshige:** None. **M. Suzuki:** None. **N. Kikuchi:** None. **Y. Ugawa:** None. **Y. Nishimura:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.15

Topic: E.06. Posture and Gait

Title: Visual reliance is related to sensory deficits in individuals with and without cerebral Palsy

Authors: *A. SANSARE, M. ARCODIA, S. C. K. LEE, J. JEKA, H. REIMANN;
Univ. of Delaware, Univ. of Delaware, Newark, DE

Abstract: Individuals with cerebral palsy (CP) have lower limb sensory deficits, such as difficulty in perceiving touch, vibrations, position of joints and limbs, and in discriminating between two points in space. They compensate for these deficits typically by relying on vision over other senses for balance control. They have magnified responses to visual perturbations, implying that they are more affected by changes in visual input. The aim of this study was to investigate the relationship between sensory deficits and responses to visual perturbations during walking. Our hypothesis, based on previous studies on standing balance, is that individuals with degraded sensory processing will have a larger response to visual perturbations. Twenty-eight individuals (14 with CP, 14 age-and sex-matched controls) walked on a self-paced treadmill inside a virtual reality cave that induced a visual fall stimulus in the frontal plane every 10-12 steps. Each trial lasted two minutes and was repeated ten times, with intermittent rest breaks. We correlated the magnitude of the center of mass response to the visual perturbations with their performance on sensory tests, including ability to perceive touch, vibration, joint position sense and 2-point discrimination. We found significant ($p<0.05$) and strong correlations between the response to visual perturbations and performance on tests of vibration ($r=-0.74$), joint position sense ($r=0.71$) and 2-point discrimination ($r=0.62$). These results support our hypothesis that those individuals with degraded sensory processing are more affected by visual input during walking. We speculate that they rely more on visual information for balance control to compensate for deficits in somatosensory and proprioceptive processing. Also, while our results showed a strong correlation between sensory deficits and a response to a visual perturbation for both the CP as well as the healthy control group, the relationship was stronger for CP ($r=0.80$, 0.88, and 0.77 for vibration, joint position and 2-point discrimination respectively) and the non-dominant side in the control group ($r=0.74$, 0.84, and 0.70 for vibration, joint position and 2-point discrimination respectively). Overall, our findings suggest a direct link between sensory deficits and visual reliance during walking. While most assessments as well as treatments for balance rehabilitation in CP address motor deficits, our findings highlight the importance of considering sensory regulation of walking balance in rehabilitation protocols.

Disclosures: **A. Sansare:** None. **M. Arcodia:** None. **S.C.K. Lee:** None. **J. Jeka:** None. **H. Reimann:** None.

Poster

550. Posture and Gait: Afferent Control

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 550.16

Topic: E.06. Posture and Gait

Support: This work was supported by an Aston funded PhD studentship (A/C Code: 10656)

Title: Sensorimotor recalibration of postural control strategies occurs after whole body vibration

Authors: I. RIGONI[1,2], G. DEGANO[2], M. HASSAN[3], *A. FRATINI[1];
[1]Aston Univ., Birmingham, United Kingdom; [2]Univ. of Geneva, Geneva, Switzerland;
[3]Reykjavik Univ., Reykjavik, Iceland

Abstract: Efficient postural control results from an effective interplay between sensory feedback integration and muscle recruitment and can be affected by ageing, neuromuscular injuries as well as by external stimuli. Whole body vibration (WBV) is a renowned way to stimulate muscle spindles firing rate with mechanical vibrations delivered to the body via mean of a platform and has recently been incorporated in many rehabilitative programs. However, the effect of this stimulation on muscle modulation for postural control is poorly understood. Here, we investigate the latter by measuring centre of pressure (COP) displacement via a force platform one and four minutes before and after WBVs delivered at 30 Hz. To characterize muscle modulation elicited by the central nervous system (CNS), we measured electroencephalography (EEG) and electromyography from 10 calf and thigh muscles of 17 healthy subjects. We investigated the postural control strategies via mean of corticomuscular and intramuscular coherence (CMC and IMC), cortical beta-band activity (BBA, 13-30 Hz) and COP complexity index (CI). Most differences were noticeable in the short term (one minute after the WBV), rather than in the long one. The CMC between dominant calf muscles (gastrocnemius lateralis, GL, and soleus, SOL) and a set of contralateral central EEG sensors increased after the WBVs, suggesting that the communication between the CNS and the periphery was enhanced, as expected after such stimulation of the spindles. We also found a bigger BBA over the central electrodes, supporting the hypothesis that a greater cortical effort was necessary to maintain the current physical output and, on the same line, we found that balance worsened in the acute term, as the COP CI increased in the medial-lateral direction. However, it recovered over the four minutes, as a lower anterior-posterior CI indicates a more spontaneous and efficient postural control. Finally, the network analyses performed on the IMC highlighted a short-term reconfiguration of muscle recruitment patterns pointing at a more individualized modulation –likely from supraspinal origins rather than subcortical ones- of the dominant GL and SOL. Altogether, our results suggest that the WBV stimulation is able to affect the interplay between the CNS and the peripheral muscles not only *during* the stimulation but also once it ends, leading to a greater contribution of cortical activity and recalibration of the sensorimotor set. On the posturography side, although balance seems disrupted at first, the ability to maintain an upright stable posture seems to be even bettered by the stimulation in the longer term.

Disclosures: I. Rigoni: None. **G. Degano:** None. **M. Hassan:** A. Employment/Salary (full or part-time):; MINDig, Rennes, France. **A. Fratini:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.01

Topic: F.01. Neuroethology

Support: Canadian Institute of Health Research (CIHR)

Title: Neural heterogeneities improve coding of prey location in weakly electric fish

Authors: *M. HAGGARD[1], M. J. CHACRON[2];
[1]Quantitative Life Sci., [2]Physiol., McGill Univ., Montreal, QC, Canada

Abstract: Despite recent technological advances, how the activities of sensory neural populations are combined to give rise to perception and behavior remains poorly understood. This is in part because neurons typically display strongly heterogeneous responses to stimulation. Behavioral studies have shown that weakly electric fish are highly skilled at detecting and locating prey that give rise to extremely weak sensory input (<0.1% contrast). To understand how these remarkable abilities arise, we investigated how heterogeneous neural populations within the electrosensory system of the weakly electric fish *Apteronotus leptorhynchus* encode the location of prey-like stimuli. Large scale neural recordings within the electrosensory lateral line lobe (ELL) were made using high-density electrode arrays (i.e., Neuropixel probes) in response to stimuli positioned at different locations along the animal's rostro-caudal axis. We found that ELL pyramidal cell receptive fields (i.e., the firing rate modulation as a function of spatial location) displayed an antagonistic center-surround organization that varied greatly across neurons (i.e., was highly heterogeneous) in terms of attributes such as depth of modulation, spatial dimensions, and location. Using Fisher information, our data demonstrate that, to accurately localize a small prey item, ~3 mm in diameter, as few as ~30 neurons are needed. To better understand how neural heterogeneities contribute to localization performance, we built a mathematical model that captures the essential aspects of neural responses. Varying heterogeneities in the model surprisingly revealed that the level of receptive field spatial location seen experimentally was optimal and thus gives rise to the maximum possible amount of information. Moreover, analysis of correlations between the trial-to-trial variabilities of neural responses (i.e., noise correlations) revealed a strong dependence on spatial position. Specifically, correlation magnitude increased at the receptive field edges and was negligible within receptive field centers. Interestingly, we found that spatially dependent correlations increase information transmission relative to spatial independence, suggesting that the spatial dependency helps mitigate the negative impact of increased redundancy. Taken together, our results strongly suggest that neural heterogeneities within the ELL pyramidal cell population benefit the animal's ability to successfully locate prey. Because of strong similarities between the electrosensory system and other senses (e.g., vision, audition, vestibular), it is likely that our results will be generally applicable.

Disclosures: M. Haggard: None. **M.J. Chacron:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.02

Topic: F.01. Neuroethology

Support: UNAM-DGAPA-PAPIIT IN231620

Title: The electrophysiological brain response to urine detection and its relation with the hierarchical order in P. clarkii

Authors: *I. HERNANDEZ PRIOR, M. OSORIO PALACIOS, K. MENDOZA ÁNGELES, J. HERNÁNDEZ FALCÓN;
Univ. Nacional Autónoma De México, Univ. Nacional Autónoma De México, Ciudad de México, Mexico

Abstract: In crayfish, agonistic encounters are behavioral displays that regulate the spatial distribution of animals and access to food supplies and mates. When a triad of unfamiliar crayfish is placed in the same circular aquarium, animals display a patterned behavior, they begin aggressive confrontation. The first agonistic encounter is usually intense, then, encounters de-escalate as time goes on, and a dominant crayfish emerges with two submissive ones. When agonistic encounters are repeated daily, their intensity decreases until a steady level is reached. This decrease seems to depend on individual recognition of the dominant animal through some substance present in its urine. It is supposed that this putative substance should reach the olfactory receptors located in the antennule, and then activate the olfactory lobe in the brain. However, there are not studies on the sequence of events until reaching the brain. The main goal of this paper was to analyze the antennule and brain electrical responses of crayfish from a triad to urine of their members. We recorded the electrical response from the antennule and, simultaneously, the olfactory lobe from dominant, submissive 1 and submissive 2 animals in the presence of urine from their conspecifics. We analyzed the electrical responses through time-frequency analysis, Wavelet Transform. Our results showed that, the latency, magnitude and duration of the brain's response depended on the source of urine. The dominant animal responds energetically to submissive-1 urine, and not to that from submissive-2. Submissive-2 responds mainly to submissive 1 urine. Submissive-1 is able to recognize the urine from the dominant or the submissive-2 animals. The response is higher when the applied urine comes from a dominant animal, with an increase in the power of frequencies from 30 to 60 Hz and a decrease in the frequencies from 1-20 Hz. For all animals, the average power analysis through frequency bands, shows that the main changes are those that occur between 30-60 Hz. Urine from unknown naïve animals or from the animal itself did not elicit a significant response in any crayfish from a hierarchical stable triad.

Disclosures: I. Hernandez Prior: None. **M. Osorio Palacios:** None. **K. Mendoza Ángeles:** None. **J. Hernández Falcón:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.03

Topic: F.01. Neuroethology

Support: CONACYT-GRANT 415154

Title: Mother and sibling interactions during the pre-weaning period influence myelination and impulse propagation of the sensory sural nerve in the adult rat

Authors: *V. MARTINEZ-ALVAREZ[1,2], B. SEGURA-ALEGRIA[2], E. RODRÍGUEZ-TORRES[3], M. PORRAS-GRACIELA[4], E. AGUIRRE-BENITEZ[4], M. GONZÁLEZ DEL PLIEGO[4], R. HUDSON[5], S. QUIROZ-GONZÁLEZ[6], A. I. MELO[7], I. JIMENEZ-ESTRADA[1]; [1]Physiology, Biophysics and Neurosci., CINVESTAV-IPN, Mexico City, Mexico; [2]UNAM-FES Iztacala, Mexico City, Mexico; [3]Univ. Autonoma del Estado de Hidalgo, Hidalgo, Mexico, Mexico; [4]Facultad de Medicina-UNAM, Mexico City, Mexico; [5]Inst. de Investigaciones Biomedicas, Univesidad Nacional Autonoma de Mexico, Mexico City, Mexico; [6]Univ. Estatal Del Valle De Ecatepec, State of Mexico, Mexico; [7]Cinvestav-lab.tlax.Universidad Autónoma De Tlaxcala, Cinvestav-lab.tlax.Universidad Autónoma De Tlaxcala, Tlaxcala, Mexico

Abstract: Myelin is directly associated with the functionality of axons such as the propagation of action potentials and its formation is associated to the nutritional condition, social context and sensory experience of mammals during early development. In this study we investigate whether mother and sibling interactions during the pre-weaning period influence the histological and electrophysiological characteristics of the sensory sural (SU) nerve at postnatal day 60 (PND60) in the rat. For this purpose, the following experimental groups were formed: A) Sibling deprivation group (1 male pup per litter; n = 9 litters) and B) Sibling groups (3, 6, 9 and 12 male pups per litter n=5 litters); Litters with 6, 9 or 12 male pups were formed at PND2 with natural (pups born and raised with their own mother) and foster pups (incorporated to the litter) from synchronous births of two or three pregnant females. The body weight of each pup was measured routinely until PND60 and within-litter analysis was performed in the lighter and heaviest pup of litters with 6,9 and 12 pups. During the pre-weaning period, milk intake and maternal care at PND9-10 were determined. At PND60, the left leg SU nerve of selected littermates was dissected and prepared for the electrophysiological recording of the Compound Action Potential (CAP) and the right nerve was prepared for histological measurements (axon diameter and myelin thickness). Our results indicate that pups from litters with 3 or 6 pups showed greater milk consumption and body weight than pups without siblings (1P) or from 9P or 12P litters. Analysis of maternal licking showed that pups from large litters received considerably fewer licks per pup than pups from small size litters. At PND60, SU nerves of rats from 6P and 9P litters had greater CAP amplitude and area and higher proportion of axons with large myelin thickness than nerves from rats of 1P, 3P, or 12P litters. Finally, no significant differences in body weight, PAC propagation and axon myelin thickness of SU nerve were found between natural and foster littermates of large size litters, indicating that fostering seems not affect the body weight and electrophysiological and histological characteristics of axons in nerves of littermates in large size litters (6P, 9P and 12P). Accordingly, we propose that a complex interplay of sensory, social and nutritional factors arising from mother and littermate interactions during the pre-weaning period influence the myelination and propagation of action potentials in the SU of rats into adulthood.

Disclosures: V. Martinez-Alvarez: None. **B. Segura-Alegria:** None. **E. Rodríguez-Torres:** None. **M. Porras-Graciela:** None. **E. Aguirre-Benitez:** None. **M. González del Pliego:**

None. **R. Hudson:** None. **S. Quiroz-González:** None. **A.I. Melo:** None. **I. Jimenez-Estrada:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.04

Topic: F.01. Neuroethology

Support: Wellcome 219627/Z/19/Z
Gatsby Charitable Foundation GAT3755

Title: Activity of dopaminergic neurons in the tail of the striatum increases with threat and is reduced following escape suppression in mice

Authors: *S. C. LENZI, T. BRANCO, M. STEPHENSON-JONES, T. W. MARGRIE; Sainsbury Wellcome Ctr. for Neural Circuits and Behaviour, UCL, London, United Kingdom

Abstract: Sensory stimuli that might signal threatening events are parsed by the brain to select behavioral responses that are fundamental to survival. Over the lifespan of an individual, however, constant revision of the threat value of stimuli in the environment is also believed to be essential for optimizing behavioral selection (Evans et al. 2019). Recently we have described a new behavioral paradigm in laboratory mice that results in a rapidly learned and stimulus-specific long-term suppression of escape to looming stimuli - Learned Suppression of Escape (LSE) (Lenzi et al. 2022). To understand how the escape circuitry supports such flexible behaviour we have investigated whether dopaminergic neurons in the Substantia Nigra pars Lateralis (SNL), which project to the Tail of the Striatum (TS), could signal changes in threat intensity (Menegas et al. 2018) that may be used to update escape decisions. First, we found that lesions of the SNL and/or TS abolish normal robust escape behavior. We next injected AAV5-TRE-GCaMP6f into the SNL of dopaminergic-neuron-specific tTA expressing (DAT-tTA) mice and recorded photometry signals from SNL dopaminergic axons in the TS. We observed reliable, large calcium responses to looming stimuli that correlated with threat level and escape probability. These calcium responses were reduced during and following LSE. Similarly, both D1R and D2R-expressing neurons in the TS showed reduced responses following LSE, indicating that both pathways undergo experience-dependent modulation. Further, we found that previous threat escape within 24 hours prior to the LSE protocol, which occludes LSE, results in normal escape behavior and does not lead to attenuation of these SNL calcium signals recorded in the TS. We suggest that dopaminergic TS signals form part of the neuronal machinery required for flexible escape behavior that is dependent on prior experience and threat prediction.

Disclosures: S.C. Lenzi: None. **T. Branco:** None. **M. Stephenson-Jones:** None. **T.W. Margrie:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.05

Topic: F.01. Neuroethology

Support: NIH Grant R01 GM130842-01

Title: Feel the heat: role of TRPA1 in thermotactic response to innocuous temperatures in Drosophila melanogaster larva

Authors: *S. LAZOPULO[1,2], A. SAMUEL[1,2];
[1]Dept. of Physics, [2]Ctr. for Brain Sci., Harvard Univ., Cambridge, MA

Abstract: Complex behavioral responses require integration of inputs from multiple types of receptors both within and across modalities. To study integration, we investigate thermosensing in *Drosophila melanogaster* larva, a genetically and neuronally accessible model organism. In endotherms, body temperature is maintained through internal homeostatic control. For small ectotherms like larvae, which receive heat from outside, homeostatic control is achieved by navigating the environment. Previous studies identified receptors that specifically respond to temperature changes. However, homeostatic control requires additional sensors to compare ambient temperature to the set point. Here, we use genetic manipulations combined with quantitative behavioral analysis and fluorescent imaging of neuronal activity in living animals to elucidate the molecular and cellular receptors that control thermotactic response towards the homeostatic set point. Larval exploratory behavior primarily consists of forward peristaltic movement (runs) interrupted by reorientations (turns). We found that larvae respond to changing temperature through increased turning. Below the preferred temperature - at 18°C - cooling elicits strong turning response (Δ=65% increase of turning) while warming doesn't (Δ=0%). In contrast, above preferred temperature - at 28°C - cooling response is suppressed (Δ=20%) and warming - induced (Δ=20%). This temperature dependency was lost in animals lacking transient receptor potential ankyrin 1 (TRPA1, either through null mutation or RNA interference mediated depletion). Using genetic knockouts and rescues we further found that it is specifically the TRPA1 isoform A that is necessary and sufficient for the proper response. Next, we analyzed temperature dependent activity of TRPA1-expressing neurons using calcium imaging. We did not detect temperature dependent activity in TRPA1-expressing multidendritic sensory neurons across the studied temperature range. Instead, we found activity in novel pair of neurons in the anterior ventral part of the brain. Their activity level correlated with temperature, implicating these neurons as potential sensors of internal absolute temperature. TRPA1 has been previously identified as a temperature sensor required for temperature preference behavior in adult flies. But in larvae, TRPA1 has primarily been studied in its responses to noxious stimuli. We suggest a novel role of TRPA1 in modulating response to temperature change in the innocuous range. Together with the previously identified thermosensors, our results are unveiling a complete and integrative process of larval thermotactic behavior.

Disclosures: S. Lazopulo: None. **A. Samuel:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.06

Topic: F.01. Neuroethology

Support: Grant PGC2018-097769-B-C22 from the Spanish Ministry of Science and Innovation
Spanish Ministry of Economy and Competitiveness and the European Union (via ERDF funds) through the research project (TIN2017-84968-R)

Title: Reversal training facilitates acquisition of new learning in a virtual reality maze

Authors: *P. OGALLAR[1], J. MORENO[1], J. M. ROSAS[1], J. M. JURADO[1], J. A. ALCALA[2], J. R. JIMENEZ-PEREZ[1], J. E. CALLEJAS-AGUILERA[1];
[1]Univ. de Jaén, Jaén, Spain; [2]Univ. Complutense de Madrid, Madrid, Spain

Abstract: The Morris water maze (MWM) has become one of the most widely used laboratory tools in behavioral neuroscience. It has been used in some of the most sophisticated experiments in the study of spatial learning and memory with animals. Recent advances in virtual reality (VR) make it possible to generate 3D environments with a high level of realism and user immersion. Current VR systems integrate eye-tracking devices to measure the user's attention on virtual entities. The main goal of this study was to replicate in humans the results found by Alcalá et al. (2020) with rats in which reversal training facilitated the acquisition of new learning in a Morris water maze, and thus validate VR technology as a novel way for the study on learning and spatial memory with human participants. The participants wore a 3D headset and could navigate the environment by using the handsets of the equipment. The task was to find a treasure buried under the ground in a localization that was signaled by two landmarks. During the acquisition phase (16 training trials) participants in all groups were required to find the treasure localized between landmarks A and B. During a second phase (8 training trials), for the Long Acquisition group training continued as before; for the Reversal group the treasure was placed in the opposite quadrant from the first phase; and for the Short Acquisition group there was no training in this phase. Finally, all groups received 8 training trials in which the treasure was placed between a new landmark (C) and landmark A. The results found in this study replicated those reported by Alcala et al. (2020) suggesting that the reversal training facilitated learning of a new treasure position signaled by landmarks C and A. This result is consistent with the idea that the increase on the prediction error attained to reversal training leads to an activation of the exploratory attention mechanism that facilitates subsequent learning.
Alcalá, J. A., Callejas-Aguilera, J. E., Nelson, J. B., & Rosas, J. M. (2020). Reversal training facilitates acquisition of new learning in a Morris water maze. *Learning & behavior, 48*(2), 208-220. doi: 10.3758/s13420-019-00392-7

Disclosures: **P. Ogallar:** None. **J. Moreno:** None. **J.M. Rosas:** None. **J.M. Jurado:** None. **J.A. Alcala:** None. **J.R. Jimenez-Perez:** None. **J.E. Callejas-Aguilera:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.07

Topic: F.01. Neuroethology

Support: IBS Grant IBS-R001-D2

Title: Characterization of TMEM43 as a novel ion channel

Authors: M. W. JANG, J. WON, Y.-E. HAN, C. J. LEE;
Ctr. for Cognition and Sociality, Inst. for Basic Sci., Daejeon, Korea, Republic of

Abstract: TMEM43 is a transmembrane protein with 4 transmembrane (TM) domains. A nonsense *TMEM43* variant p.(Arg372Ter) has been shown to cause auditory neuropathy spectrum disorder (ANSD) in two Asian families from South Korea and China. A knock-in mouse with the p.(Arg372Ter) variant that recapitulates a progressive hearing loss in humans, revealed that TMEM43 interacts with the Connexin26 and Connexin30 gap junction channels in the cochlea and mediates passive conductance current, that is critical for normal hearing. In this study, we examined if TMEM43 can function as an ion channel. Heterologous expression of TMEM43 demonstrated that TMEM43 is permeable to Na^+, K^+, and Cs^+ ions, indicating that TMEM43 is a non-selective cation channel. The TMEM43-mediated current decreased gradually with lowering external solution pH, further characterizing TMEM43 as an external-pH sensing channel. Utilizing the endogenous cysteine residue at TM3, we could predict that the pore-forming residue lies near TM3 and Loop2 domain. Importantly, stochastic channel-opening of the lipid-bilayer-reconstituted purified TMEM43 protein was observed, strengthening the proposal of TMEM43 as an ion channel. Lastly, heterologous expression of *TMEM43*-p.(Arg372Ter) resulted in a loss of channel activity in dominant-negative fashion, as in the hearing loss phenotype. These results together provide molecular and functional properties of TMEM43 and identify TMEM43 as a novel ion channel.

Disclosures: **M.W. Jang:** None. **J. Won:** None. **Y. Han:** None. **C.J. Lee:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.08

Topic: F.01. Neuroethology

Support: NSF Grant DBI2021795

Title: Role of Inactivating Potassium Current in Contrast-Dependent Collision Detection

Authors: *G. SHAULSKY[1], R. B. DEWELL[2], F. GABBIANI[3];
[1]Neurosci., [2]Dept of Neurosci., Baylor Col. of Med., Houston, TX; [3]Baylor Col. Med., Houston, TX

Abstract: In grasshoppers, visual detection of approaching predators is accomplished by the Lobula Giant Movement Detector (LGMD). The LGMD integrates OFF retinotopic inputs originating from every facet of the eye within a large dendritic field A. An inactivating K+ conductance (K_D) in field A is critical to discriminate the spatial coherence of black looming stimuli. ON excitation for white looming stimuli impinges non-retinotopically on a separate dendritic field C, and the LGMD does not discriminate their spatial coherence. Block of K_D increased responses to white looms, independent of coherence. To further determine how K_D impacts ON/OFF spatial coherence computations we applied blockers to dendritic fields A and C while presenting ON and OFF looming stimuli and used a biophysical LGMD model to test the influence of K_D channel localization. These experiments and simulations provide insights into dendritic computations required for contrast-polarity specific approaching object segmentation.

Disclosures: G. Shaulsky: None. **R.B. Dewell:** None. **F. Gabbiani:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.09

Topic: F.01. Neuroethology

Support: NINDS Grant R21 NS116319

Title: Magnetsearch: a collective science project to identify animal magnetoreceptors

Authors: D. POLLAK, M. H. ROSENBERG, D. A. WAGENAAR, *M. MEISTER;
Biol. and Biol. Engin., Caltech, Pasadena, CA

Abstract: If biological cells could be controlled using magnetic fields, this would enable unprecedented approaches to basic biology and medicine. Ideally one wants a genetic tool that can render arbitrary cells magnetically sensitive, a goal now known as “magnetogenetics”. Several attempts to accomplish this by de novo bioengineering have failed, mainly because magnetic fields interact only weakly with biological molecules. At the same time we know that certain animal species have the remarkable ability to sense the Earth’s magnetic field and to use this information for orientation and navigation. Thus there must exist nerve cells with the mechanism to transduce even weak magnetic fields: the magnetoreceptors. If one could find

those receptor neurons, they would reveal a cellular pathway that could be used for magnetic control. We have initiated a collective science project to find magnetoreceptors. The approach is to first search for neural signals anywhere in the brain that respond to magnetic stimuli. Based on such signals one can then pursue a magnetic scanning method to localize where the magnetic responses originate. Many research groups today are engaged in high-throughput neuroscience pursuing a broad range of questions in diverse species, using revolutionary methods that record signals from hundreds to thousands of neurons at a time. The project transiently engages many of these groups in a broad unbiased search for magnetoreceptors. The Caltech team constructs electromagnetic stimulators that produce a defined magnetic field and ships these to each partner lab. The device can be added easily to an ongoing experiment, and a mere 20 minutes of recording will reveal whether any of the neurons under study carry magnetic signals. Here we will report on the design of this project, and present data analysis methods designed to reveal a small sensory modulation with high sensitivity. We will also report preliminary results on recordings under magnetic stimuli from many thousands of neurons in diverse brain areas of mammals, fish, and birds.

Disclosures: D. Pollak: None. **M.H. Rosenberg:** None. **D.A. Wagenaar:** None. **M. Meister:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.10

Topic: F.01. Neuroethology

Support: R01MH127820
R01MH114882
R01MH104559
R01MH120514
R01MH120637

Title: The cortical amygdala contributes to distinct social behaviors in a sexually dimorphic manner

Authors: *A. V. AUBRY, L. LI, R. DURAND-DE CUTTOLI, S. J. RUSSO;
Icahn Sch. of Med., New York, NY

Abstract: Aggression is an evolutionarily conserved behavior that controls social hierarchies and protects valuable resources like mates, food, and territory. In humans, however, some forms of aggression are considered pathological when they threaten lives and or increase the risk of psychiatric impairment in victims. Physically aggressive individuals have a tendency to identify ambiguous faces as threatening or angry, suggesting that these individuals have a perceptual bias. There is ample evidence in human and non-human primates that the amygdala plays a key

role in social perception. Studies with human and non-human primates examining the amygdala's role in social perception have focused on the lateral and basolateral amygdala given their extensive connections with the visual system because they are a highly dependent on vision. Conversely, rodents are more reliant on olfaction, and thus, we need a deeper understanding of amygdala interactions with olfactory structures during social encounters. The posterior cortical amygdala (COAp) is one such region which receives input from olfactory structures. Our data demonstrate that ESR-1 cells in the COAp show an increase in activity during attack behavior in males, and during bouts of investigation which precede an attack. Furthermore, chemogenetic activation of ESR-1 cells in the COAp promote aggression and inhibition of these cells promote pro-social interactions in aggressive males. We also found that the projections of this cell population to the ventrolateral portion of the ventromedial hypothalamus and central amygdala and are necessary for aggressive behavior to occur in males. Collectively, these data suggest that in aggressive males, the COAp is highly responsive to odors emitted by the intruder, thereby enhancing their salience and promoting attack behavior.

Disclosures: A.V. Aubry: None. **L. Li:** None. **R. Durand-De Cuttoli:** None. **S.J. Russo:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.11

Topic: F.01. Neuroethology

Support: NSF NeuroNex Innovation Award

Title: Interphotoreceptor connections in Heliconius lamina

Authors: *D. PAUKNER[1,3,2], S. E. PALMER[2], N. KASTHURI[1,3];
[1]Neurobio., [2]Organismal Biol. and Anat., Univ. of Chicago, Chicago, IL; [3]Argonne Natl. Lab., Lemont, IL

Abstract: The way nervous systems evolve to produce different behaviors is an important question for understanding how organisms adapt to their environments. Heliconius cydno alithea butterflies provide a novel model for addressing this question. Male Heliconius cydno alithea prefer mating with females of their own color, which is causing this species to diverge. While this behavior is visually mediated, the neural mechanism is still unknown. A proposed mechanism is at the level of synapses, ultraviolet (UV) photoreceptors in the lamina of yellow male Heliconius receive more synaptic inhibition from long-wavelength photoreceptors than in white male Heliconius. We will use large volume serial electron microscopy - connectomics- to test this hypothesis, reconstructing the size and number of individual synapses between UV and long wavelength photoreceptors in white and yellow male butterflies. I have preliminary SEM

data that encompasses the length of the male Heliconius lamina with a resolution of 5nm/pixel, which I have used to identify synapses between photoreceptors.

Disclosures: D. Paukner: None. **S.E. Palmer:** None. **N. Kasthuri:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.12

Topic: F.01. Neuroethology

Support: CONACYT- CVU 1084476

Title: Milk fat concentration deppends on litter sex proportion in the rat

Authors: *V. CUBRIA[1], B. MOLINA[2], E. CUEVAS-ROMERO[1], L. M. STEVENS[3], M. A. LARA GARCIA[4], P. PACHECO[5];
[1]Univ. Autónoma de Tlaxcala, Tlaxcala, Mexico; [2]Univ. Nacional Autónoma de Mexico, Xalapa, Mexico; [3]Psychology, Dalhousie Univ., Halifax, NS, Canada; [4]Inst. de Neuroetologia, Univ. Veracruzana, Xalapa, Mexico; [5]Inst. de Investigaciones Biomédicas, Univ. Nacional Autonoma de México, Xalapa, Mexico

Abstract: During lactation, the most important stimuli that the mother receives are those which activate specific receptors located in/and around the nipple during suckling and exteroceptive stimuli which emanate from the offspring and which are perceived by her special organ senses. These stimuli are responsible for the majority of the endocrine, metabolic and behavioral adjustments that develop in the lactating animal and which enable her to care for her offspring. Recently it was proposed that in *Macacca mullata,* mothers of male pups have a higher concentration of fat milk. In the present study, it was explored in the rat, if there was also a correlation of the offspring sex with the milk fat concentration. We used lactating mothers with a compound of litters of 8 female pups (n=6) or 8 males pups (n=6) or 4-4 females/males pups (n=6). The fat concentration was measured using a simplified method of Rose-Gottlieb, on milk obtained by mummifying the suckling by manual stimulation in the mother, previously injected with i.p 1.5 UI of oxytocin. The measurements were done on days 5, 12, and 19 postpartum. Additionally, the anogenital licking behavior of the mother throughout her litter was also measured using a 60 minutes videotape recording on individual litters. Results.- The milk fat concentration was higher (68 mg) in the group of 8 females pups as compared with the group with 8 males (52 mg). In the mixed group, there was obtained a 60 mg. On the Anogenital Licking behavior, there was observed that mothers provide more lickings in males (17 lickings in 60 minutes) than on females (5 lickings in 60 minutes). Discussion.- The milk on the rat mammary gland is produced by the epithelial cell of the alveoli. This cell is modulated by prolactin and adrenaline liberated by suckling. Possibly, pheromones; electrolytes obtained from the pup urine; ultrasonic vocalizations produced by pups (more in males than in females).

Disclosures: **V. Cubria:** None. **B. Molina:** None. **E. Cuevas-Romero:** None. **L.M. Stevens:** None. **M.A. Lara Garcia:** None. **P. Pacheco:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.13

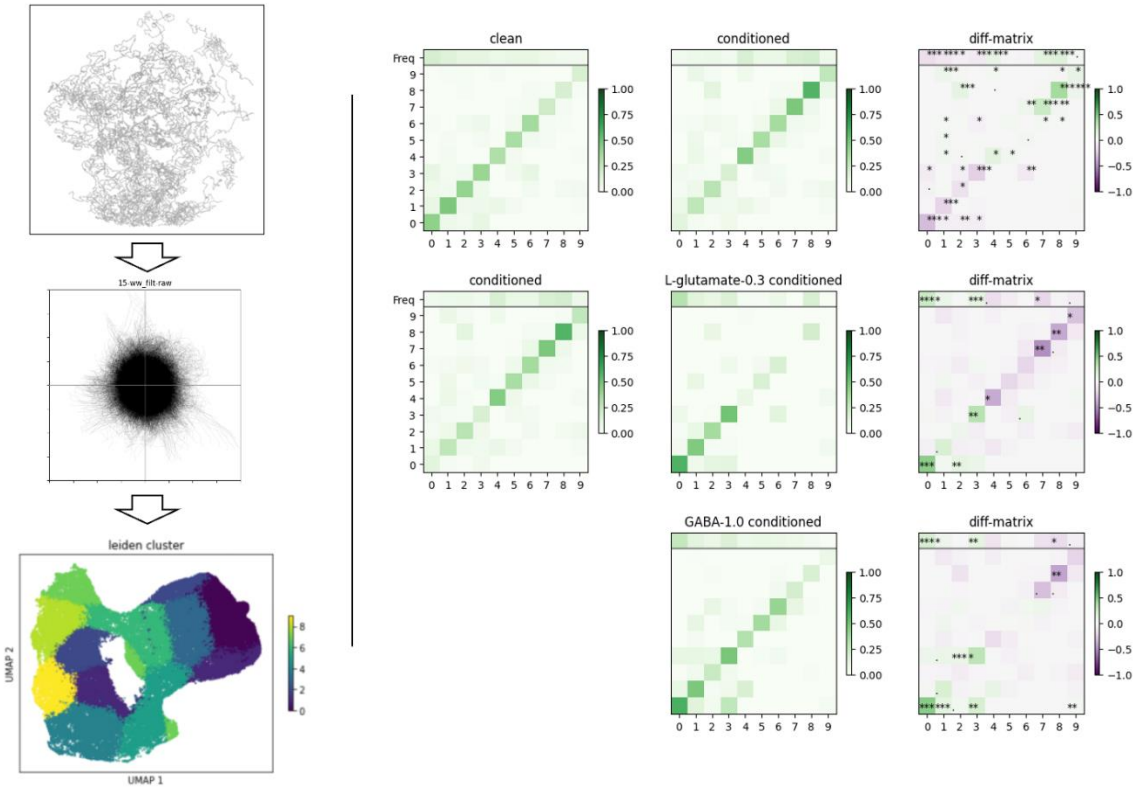
Topic: F.01. Neuroethology

Support: NSF
NIH

Title: Deciphering structure of behavior in the simplest free living animal with machine learning

Authors: ***W. WEERTMAN**[1], C. SUN[2], D. ROMANOVA[3], M. NIKITIN[4], L. L. MOROZ[5]; [1]Univ. of Washington, Seattle, WA; [2]Computer Engin., Univ. of Florida, Gainesville, FL; [3]Lab. of Cell. Neurobio. of Learning, Inst. of Higher Nervous Activity and Neurophysiol., Moscow, Russian Federation; [4]Moscow State Univ., Moscow, Russian Federation; [5]The Whitney laboratory for Marine Biosci., Univ. of Florida Dept. of Neurosci., Saint Augustine, FL

Abstract: Placozoans are simplest free living animals with just 6-10 described cell types. Despite such apparent morphological simplicity, placozoans exhibit fast and complex behaviors, including social behaviors. How is this possible in an organism lacking both neuronal and muscle cells, and without electrical and chemical synapses? Behaviors exhibited by placozoans are not well described and difficult to robustly sort by eye. Here, we developed and implemented a broad spectrum of machine learning algorithms and approaches to unbiasedly decipher the scope and functional significance of various behavior patterns in these enigmatic animals. Tractlets of several hundreds of individual placozoans were initially tracked using ImageJ. These positions were then windowed and Uniform Manifold Approximation and Projection was used to build low-dimensional manifolds of behaviors for visualization and construct a k-nearest neighbor graph for Leiden/Louvain community based clustering. Speed, entropy, and curvature for the different communities were assessed and similar communities were combined. Markov chain transition matrices and cluster frequencies were then assessed for each animal before and after application of candidate signaling molecules, identified in microchemical and scRNA-seq experiments. Before, after, and control Markov chain transition matrices were compared using pair wise statistics. To our surprise, we detected unprecedented behavioral variance among genetically similar clonal animals. Nevertheless, machine learning approaches reproducibly identified distinct behavioral structures (motifs), which were selectively and deferentially affected by individual signal molecules (and their different concentrations), implying integrative functions of volume transmission in these nerveless organisms. Unsupervised machine learning can be applicable to the broadest spectrum of organisms. This, it serves as a powerful tool for discovering behavioral modifications across phyla both in natural habitats and pharmacologically. Supported by NSF and NIH to LLM



Disclosures: **W. Weertman:** None. **C. Sun:** None. **D. Romanova:** None. **M. Nikitin:** None. **L.L. Moroz:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.14

Topic: F.01. Neuroethology

Support: Deutsche Forschungsgemeinschaft (DFG)
SFB Grant 1089
ISF Grant 1539/17
Grant from Pasteur-Weizmann Council
Marianne Manoville Beck Laboratory for Research

Title: Object-specific neuronal response in the auditory system during active whisking

Authors: *B. EFRON, Y. KATZ, I. LAMPL;
Weizmann Inst. of Sci., Weizmann Inst. of Sci., Rehovot, Israel

Abstract: Integrating information from different sensory systems is essential for comprehensively representing the external world. The different senses represent various aspects of the surrounding world at multiple time scales and distances from the body. For example, tactile sensation enables sensing proximal objects, whereas the auditory system also allows sensing distant objects. In many instances, animals perceive things simultaneously using two or more modalities, which can improve performance in different tasks. Although the mechanisms of multimodal integration have been studied in rodents, usually, the experimentalists provided unrelated artificial stimuli from two or more modalities and asked where and how they are integrated. Here we illustrate that the vibrissa system of rodents can create multimodal interactions and perception. The vibrissa sensory system of the mouse is its primary tactile system, allowing it to sense its environment by active whisking.In contrast to the standard view of this system, we show for the first time that the vibrissa system is not solely tactile but can be an audio-tactile system. We found that when mice whisk against certain objects, for example, dry leaves, whisking-touch events produce audible object-specific sounds that alter the firing rate of neurons in the inferior colliculus (N = 2,n = 22) and the auditory cortex. To show that the change in the firing rate of neurons in the auditory cortex (N = 3, n = 140) is caused by the whisking-touch induced sound and not due to tactile signals from the somatosensory system, we cut the Infraorbital nerve (ION) that conveys information from the whiskers to the brain. Under this condition, most neurons in the auditory cortex still responded to whisking-touch audible events. Importantly, different neuronal responses were found, including elevated and reduced firing in response to audible objects and whisking-only units. Furthermore, a simple neural network classifier showed that neuronal response captured the mouse's behavior (non-whisking, free whisking) and differentiated contacts with a noisy vs. an attenuated object. (i.e., an object identical in coarseness to the noisy object but much less noisy).These novel findings are potentially relevant to the natural behavior of mice and other rodents that actively explore their environment by whisking. These auditory-tactile signals can improve the perception of objects by providing complementary auditory information to tactile information. Primary and multimodal sensory areas in the cortex might integrate the complementary sensory information and allow a better perception of the environment.

Disclosures: **B. Efron:** None. **Y. Katz:** None. **I. Lampl:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.15

Topic: F.01. Neuroethology

Support: R01NS118442
NSF EAGER Rules of Life #1838346

Title: The effect of temperature on predator-prey interactions: learning and metabolic asymmetry shape hunting outcomes

Authors: *J. L. AMME, J. M. GRADY, S. J. BRUNWASSER, A. I. DELL, K. B. HENGEN; Biol., Washington Univ. In St. Louis, St. Louis, MO

Abstract: The role of environmental temperature in shaping species interactions is under increasing scrutiny, yet the extent to which learning contributes to how animals respond to changing environmental conditions is largely unexplored. Currently, the effect of temperature on animal behavior is primarily studied through a metabolic lens: temperature imposes constraints on movement and behavior as a function of metabolic profile (e.g., warm or cold-blooded), which in turn shapes species interactions. In the case of predation, hunting outcomes predicted by this model are based solely on differences in the metabolism of interacting species and their resultant asymmetric performance across a temperature gradient. However, learning may also shape behavioral responses and hunting success across changing temperature. In this study we aimed to quantify the effect of temperature on prey capture. We hypothesized that predatory learning, in addition to metabolic asymmetry between predator and prey, modulates changes in hunting success across a range of temperatures. To test this, we developed an experimental housing arena in which we can vary ambient temperature while also tracking multispecies interactions using machine vision. We first trained adult C57BL/6 mice (n=8, sex-balanced) to hunt cockroaches (*Shelfordella lateralis*) at room temperature. We found that mice achieve consistent capture performance after 1 week. We then altered ambient temperature across a ~25C range to measure its effect on capture performance. We found that hunting success, as measured by time to capture, varied robustly as a log-linear function of temperature, with time to capture increasing four-fold with ambient temperature ($p < 0.001$). However, our results deviated from predictions based on metabolic theory; namely, mice tended to maintain consistent levels of performance at hotter temperatures despite the cockroaches, as cold-blooded organisms, moving fastest in these conditions. Using machine vision, we conducted pose estimation on mouse and cockroach in each hunting trial (n=2560). We found that mice exhibited a repertoire of temperature-dependent behavioral patterns during hunting and may use temperature as a predictive cue to select hunting strategy. Together, these results suggest that learning and behavioral plasticity play important roles in further shaping predator-prey outcomes beyond that which is predicted by metabolic constraints alone. Consideration of prey capture - a ubiquitous and ethologically relevant behavior - offers unique insights into how animals adapt to changing environmental conditions.

Disclosures: J.L. Amme: None. **J.M. Grady:** None. **S.J. Brunwasser:** None. **A.I. Dell:** None. **K.B. Hengen:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.16

Topic: F.01. Neuroethology

Support: NIH Grant EY026031

Title: Columnar T3 neurons support saccadic object vision in flies

Authors: *G. FRIGHETTO, M. A. FRYE;
Integrative Biol. and Physiol., UCLA, Los Angeles, CA

Abstract: Detecting salient visual objects against a moving background is anything but simple and the mechanisms remain enigmatic in visual neuroscience. The fruit fly has been a highly successful model organism for understanding a complimentary challenge: how the visual system detects the direction of motion. Motion direction is computed by the columnar T4 and T5 cells (i.e., the first-order direction selective neurons) that segregate ON and OFF signals, respectively. These cells supply wide-field integrating neurons of the lobula plate that encode patterns of panoramic optic flow and coordinate directional gaze stabilization behavior, but these neurons are not suited for the robust object tracking behaviors that flies produce. Here, we hypothesized that a visual pathway parallel to the T4/T5 direction selective cells encode small moving targets and drive body saccades toward them. We combined physiological and behavioral experiments to investigate the role of another class of T- cells, columnar T3 neurons, innervating the lobula. By using two-photon calcium imaging, we demonstrated that T3 neurons are well tuned to motion-defined bars. Then, we showed that T3 contribute to the normal counter-directional orientation response typically deployed for tracking motion-defined bars by rigidly tethered flies. In real closed-loop conditions, by using magnetically tethered flies, we showed that silencing of T3 reduce the number of saccades while the optogenetic activation increase them. Finally, we accounted for the saccade trigger mechanism through an integrate-and-fire model physiologically inspired by the calcium dynamic of T3 neurons. Our results represent the first evidence for a parallel motion detection pathway involved in small object tracking behavior through saccades whose computation is based on direction-agnostic contrast changes.

Disclosures: G. Frighetto: None. **M.A. Frye:** None.

Poster

551. Sensing and Detecting Movement

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 551.17

Topic: F.01. Neuroethology

Support: NSF BRAIN EAGER 1451221
NIMH R01 5R01MH110514-02
Kavli Institute for Brain and Mind

Title: Brain and Body Coupling in Neural Circuitry Underlying Social Assessment

Authors: *E. LEONARDIS[1], L. BRESTON[3], R. LUCERO-MOORE[2], R. KOHLI[2], L. BARTON-GLUZMAN[2], M. AGUILAR-RIVERA[4], L. QUINN[2], J. WILES[6], A. A. CHIBA[5];
[1]UC San Diego, San Diego, CA; [2]UC San Diego, La Jolla, CA; [3]Univ. of California San Diego,

LA JOLLA, CA; [4]Bioengineering, [5]Cognitive Sci. and Program in Neurosci., UCSD, La Jolla, CA; [6]Univ. of Queensland, Brisbane, Australia

Abstract: Social interactions rely on internal and external contexts which scaffold the complexity of human and animal behavior. Social influences exert a pronounced effect on the relationship between neural circuits and the body. Brain and body coupling during interoception and exteroception allows for agents to assess social situations in relation to salience, safety, and uncertainty. This study will examine dynamics in neural circuitry underlying social assessments by recording local field potentials in the olfactory bulb, amygdala, hippocampus and insular cortex simultaneously. A study was performed to characterize behavioral, autonomic, and multi-region brain dynamics during interaction between rodents and other conspecifics, robots, and stationary objects. The use of interactive robots served as a control condition while also revealing novel findings about social dynamics. The results from these experiments highlight the importance of naturalistic regulatory behaviors, which serve an important functional role in interoception and stabilizing the nervous system. This functional role was examined with respect to multi-region brain coupling by applying a novel method for estimating dynamic coupling to identify bidirectional coupling between regions as well as coherence analysis. Rather than advocating for the "communication-through-coherence" hypothesis, these results suggest that "communication-through-coupling" may be a more compelling approach to understanding the dynamics of neural circuits.

Disclosures: E. Leonardis: None. **L. Breston:** None. **R. Lucero-Moore:** None. **R. Kohli:** None. **L. Barton-Gluzman:** None. **M. Aguilar-Rivera:** None. **L. Quinn:** None. **J. Wiles:** None. **A.A. Chiba:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.01

Topic: F.03. Stress and the Brain

Title: Amygdala control of the emotional motor system

Authors: *G. HOLSTEGE[1], H. H. SUBRAMANIAN[2];
[1]Groningen Academic Hosp., Den Haag, Netherlands; [2]Boston Scientific Neuromodulation, Los Angeles, CA

Abstract: The amygdala regulates fear and anxiety. Expression of fear and anxiety is associated with changes in the emotional motor system control of breathing, cardiovascular and pelvic organ function. The amygdala via its strong projections to the periaqueductal gray (PAG) could function as one of the emotional motor pathways for the modulation of fear and anxiety. In that case, stimulation of the amygdala should have a direct causal influence on breathing as well as on cardiovascular and pelvic organ functions. This was investigated by stereotaxic glutamate

stimulation of different portions of the amygdala in both cats and rats. Glutamate only stimulates the cell bodies and not the fibers of passage. Using this technique it was possible to find out whether within the amygdala a topographical organization of emotional functions exist.
Results from the cat studies **Respiration:** In the cat, stimulation of the lateral amygdala generated hyperpnea and tachypnea, while stimulation of the central amygdala induced dyspnea, bradypnea, inspiratory apneusis and double diaphragm breathing patterns. Stimulation in the amygdala never generated apneas. **Cardiovascular effects:** Stimulation in both the lateral and central amygdala induced hyper- as well as hypotension. **Micturition:** In the cat stimulation of central amygdala generated micturition, but stimulation in the lateral amygdala did not.
Results from the rat studies Focal stimulation of the central amygdala in the rat induced both tachypnea/hypertension as well as apnea/hypotension. Topical stimulation in the lateral portion of the central amygdala produced breath-hold. Focal glutamate stimulation in the lateral amygdala did not result in any cardiorespiratory effect. In the rat micturition was never generated in either the lateral or the central amygdala.
ConclusionThe lateral and the central amygdalar areas produce distinct modulations of the emotional motor system depending on its association with stress, fear and anxiety. Differences between the cat and rat exist in terms of modulation of the emotional motor system.
Acknowledgement and Declaration The animal studies were undertaken by GH and HHS wholly at The University of Queensland between 2013-2017 under UQ institutional ethics approval. HHS and GH designed the project, performed the experiments, analysed primary data and made figure illustrations. GH curated and approved the final data/figure representation in this presentation/poster. None of work described here were undertaken at either of the current work designations of Hari Subramanian or Gert Holstege.

Disclosures: G. Holstege: None. **H.H. Subramanian:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.02

Topic: F.03. Stress and the Brain

Support: NIH Grant R01MH118237

Title: Social defeat alters the firing rate of medial amygdala neurons

Authors: *A. C. RITGER, M. K. LOH, N. C. FERRARA, C. P. STICKLING, J. A. ROSENKRANZ;
Ctr. For Neurobio. of Stress Resilience and Psychiatric Disorders, Rosalind Franklin Univ. of Med. and Sci., North Chicago, IL

Abstract: Depression is characterized by social withdrawal and avoidance and can emerge after exposure to a social stressor. The medial amygdala (MeA) regulates social behaviors and

responds to social cues, and there is evidence of reduced MeA volume and connectivity in people with depression. The MeA projects to the bed nucleus of the stria terminalis (BNST) and ventromedial hypothalamus (VMH), which are also involved in social avoidance and approach behaviors, suggesting MeA-related circuits may regulate social symptoms of depression. To better understand the MeA's response to social stressors, we used 5 days of repeated social defeat stress using the resident intruder paradigm or a transport cage control in adult (PND 71-83) male Sprague Dawley rats (n=84) and recorded the firing rate of spontaneously active neurons in the MeA using anesthetized *in vivo* extracellular single-unit electrophysiology. We found that MeA neurons fired faster in socially defeated rats relative to controls, and the number of attacks a stressed rat experienced per day trended towards a positive correlation with their average MeA firing rate, suggesting a direct relationship between stressor intensity and MeA firing. This increase in MeA activity after stress was driven by neurons in the posterodorsal subnucleus (MeApd), whereas the posteroventral (MeApv) firing rate was similar between groups. Cumulative frequency distributions of MeA, MeApd, and MeApv firing rates all revealed significant differences in firing rate distribution between treatment groups, suggesting distinct populations of neurons in these regions may respond differently to social stress. To examine differences in MeA projections, we used antidromic stimulation of axonal terminals to identify MeA-BNST or MeA-VMH neurons. While preliminary results indicated that firing rates were similar in MeA-BNST and -VMH neurons between groups, MeA-VMH neurons trended towards firing faster than MeA-BNST neurons in the control group. Future work will test whether differences in MeA-BNST or -VMH firing are associated with changes in social avoidance or approach behaviors following social stress. These results indicate that the MeA is sensitive to prolonged social stressors and that social defeat differentially affects the activity of MeA subnuclei. These findings point to dysregulated MeA activity as one possible mechanism for social stressors to drive maladaptive social behaviors.

Disclosures: A.C. Ritger: None. **M.K. Loh:** None. **N.C. Ferrara:** None. **C.P. Stickling:** None. **J.A. Rosenkranz:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.03

Topic: F.03. Stress and the Brain

Support: NIH Grant 5P50MH115874
NIH Grant R01MH108665
NIH Grant F32MH125634

Title: An amygdala Crh+ cell activity signature for attack initiation

Authors: *E. L. NEWMAN[1,2], K. J. THREADGILL[1,2], E. HISEY[1,2], N. RESSLER[1,2], H. R. BERMUDEZ[3], K. J. RESSLER[1,2];

[1]Psychiatry, McLean Hosp., Belmont, MA; [2]Harvard Med. Sch., Boston, MA; [3]Tufts Univ., Medford, MA

Abstract: Background: Patients with stress-induced psychopathologies like posttraumatic stress disorder experience debilitating changes in emotional reactivity that can lead to aggression. The central amygdala (CeA) is a point of intersection for neural circuits that regulate both threat responding and aggression. Here, we leveraged mouse models of naturalistic agonistic behavior to examine the neural bases of territorial and self-defensive attacks. We used single-cell calcium imaging and chemogenetics to evaluate CeA neurons that express the stress-signaling neuropeptide, corticotropin releasing hormone (Crh). **Methods**: Male CRH-ires-Cre mice received intra-CeA AAV5-EF1a-DIO-GCaMP6s and were implanted with a gradient index lens to detect calcium-dependent activity in Crh+ CeA cells. Cell activity (*n*=16-45 cells/mouse) was video-recorded using a miniature microscope during 5-minute home-cage territorial aggressive encounters with a submissive intruder mouse or during 1-minute self-defensive interactions with a highly aggressive intruder. Crh+ CeA cell ensembles were identified based on their activity (3 z-score peak event threshold) relative to territorial attack initiation. Additional CRH-ires-Cre males were infused with intra-CeA AAV5-hSyn-DIO-hM4D(Gi)-mCherry to drive inhibitory Designer Receptors Exclusively Activated by Designer Drugs (DREADDs) or control virus in Crh+ CeA neurons. Mice received systemic vehicle or deschloroclozapine (0.02 mg/kg) 5 minutes before territorial or self-defensive aggression tests. Behavioral videos were analyzed using supervised machine learning; pose estimations generated by multi-animal deeplabcut (Lauer et al. 2022) served as the input for simple behavioral analysis (Nilsson et al. bioRxiv) to classify social approach, social contact, and attacks **Results:** Three Crh+ CeA cell ensembles were identified - 41% were active in the 10 seconds before territorial attack initiation, 32% were active during attacks, and 27% were inactive during aggression. Chemogenetic inhibition of Crh+ CeA cells selectively prevented territorial attacks without affecting social approach, contact, or self-defensive bites toward an attacking intruder. **Conclusions**: CeA Crh+ cell activity is essential for the initiation of territorial attacks. Cell activity was not necessary for self-defensive bites, suggesting distinct neural circuitry underlying territorial vs. self-defensive aggression. Ongoing studies use all-optical closed-loop approaches to block attack initiation using Crh+ CeA cell activity to trigger real-time inhibitory optogenetics.

Disclosures: E.L. Newman: None. **K.J. Threadgill:** None. **E. Hisey:** None. **N. Ressler:** None. **H.R. Bermudez:** None. **K.J. Ressler:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.04

Topic: F.03. Stress and the Brain

Support: NIH Grant R01MH115016

Title: Corticotropin releasing factor (CRF) in glutamatergic, GABAergic, and GABA/glutamate neurons in the primate central extended amygdala and ventral pallidum

Authors: *J. L. FUDGE[1], E. A. KELLY[1], T. A. HACKETT[2];
[1]Neurosci., Univ. of Rochester Med. Ctr., Rochester, NY; [2]Neurosci., Vanderbilt Univ., Nashville, TN

Abstract: The central extended amygdala (CEA) and ventral pallidum are involved in diverse motivated behaviors, and encompass a large region of the basal forebrain in primates. Corticotropin releasing factor (CRF), a canonical 'stress molecule' is enriched in the CEA, and is dynamically regulated. CRF's role in behavior--particularly in higher species--has been hard to parse, in part because it modulates primary 'fast' transmitters in neural circuits. To clarify CRF's role in the context of its primary transmitters, we used a combination of immunocytochemical and spatial transcriptomics approaches to 1) survey the distribution of CRF-positive neurons in subregions of the CEA and ventral pallidum, 2) examine the overall proportions of GABAergic and glutamatergic neurons by subregion, and 3) determine the proportion of CRF-mRNA neurons that co-express glutamate (Vglut2 mRNA) and GABA (Vgat mRNA) by subregion. Young male macaques were used. CRF-protein and CRF mRNA labeled neurons were broadly dispersed in all CEA subregions, and also in the ventral pallidum and globus pallidus, pars interna (GPi). Overall, single-labeled Vgat-mRNA positive cells were the most prevalent cell type in the CEA and ventral pallidum (80%); however, Vglut2 mRNA was expressed in 20% of all neurons, 10% of which were Vgat/Vglut2 positive. Regional differences in the distribution of Vgat- and Vglut2- mRNA positive cells across CEA and ventral pallidal subregions were striking. With respect to the CRF neuronal population, CRF/Vgat-only neurons were found in highest proportions in lateral central bed nucleus, lateral central amygdala nucleus, and medial central amygdala nucleus (74%, 73%, and 85%, respectively). Lower percentages of CRF/Vgat labeled cells (53%, 54%, 17%, respectively) characterized the sublenticular extended amygdala, ventrolateral bed nucleus, and ventral pallidum. These regions had higher complements of CRF/Vgat/Vglut2 labeled neurons (33%, 29%, 67%, respectively). Across all subregions, relatively stable, low proportions of CRF/Vglut2 and CRF-mRNA single-labeled cells comprised the balance of the CRF labeled cells. In sum, the dorsal bed nucleus and central nucleus have relatively homogeneous populations of CRF- GABAergic neurons, mirroring their relatively restricted efferent systems. In contrast, the ventral lateral bed nucleus and sublenticular extended amygdala, and the ventral pallidum have heterogeneous CRF cell types, including mixed GABA/glutamatergic subpopulations, and broader efferent paths. Important next steps will be to map cell-type specific CRF circuitry associated with the CEA subregions and ventral pallidum.

Disclosures: J.L. Fudge: None. **E.A. Kelly:** None. **T.A. Hackett:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.05

Topic: F.03. Stress and the Brain

Support: VA merit Grant – I01BX002188

Title: Exposure to Behavioral Therapies Can Reverse Persistent Stress Induced Changes in Amygdalar Glucocorticoid Receptor Expression to Ameliorate Chronic Visceral Hypersensitivity.

Authors: *A. OROCK, B. GREENWOOD-VAN MEEVELD;
Physiol., Univ. of Oklahoma Hlth. Sci. Ctr., Oklahoma City, OK

Abstract: Introduction: Chronic visceral pain, often exacerbated by stress, is one of the main features of irritable bowel syndrome (IBS). Evidence suggests that behavioral therapies can be effective in managing IBS symptoms, however the mechanisms of action remain to be explored. Our previous research has revealed that stress induces persistent changes in glucocorticoid receptor (GR) function at the central nucleus of the amygdala (CeA) to cause chronic visceral hypersensitivity. Here we test the hypothesis that exposure to an enriched environment (EE), a rodent analog of behavioral therapies, is capable of ameliorating persistent visceral hypersensitivity in rats previously subjected to chronic stress by reversing stress induced changes in GR expression at the CeA. **Methods**: Male and female rats (n=4/group) were exposed to water avoidance stress (WAS) for 7 days (1hr/day). Colonic sensitivity was assessed on day 8 by measuring the number of abdominal contractions to colorectal distension pressures (0-60mmHg). The animals were then returned to standard cages (SH) for one additional week after which a cohort of WAS animals were transferred to EE cages for 14 days. On day 28 post WAS, colonic sensitivity was once again measured. CeA was also collected from the rats on day 28 to assess the effects of EE on WAS induced changes in GR expression via western blot. Results were expressed as mean ± SD. **Results**: On day 8 post-WAS, we observed colonic hypersensitivity in male (Male 60mmHg: SHAM=16±2 vs. WAS=25±3 contractions; $p<0.001$) and female rats (60mmHg SHAM=19±3 vs. WAS=27±4 contractions; $p<0.01$), which persisted until day 28 post-WAS (60mmHg: Male =24±3; Female=33±6 contractions; $p<0.001$). However, animals which were exposed to WAS and then housed in EE from days 14-28 showed visceral sensitivity (EE+WAS: Male=17±1; Female=22±2 contractions) that was comparable to SHAM controls and significantly lower than SH+WAS animals either on day 8 or day 28 ($p<0.01$, 2-way ANOVA). WAS caused a persistent decease in GR expression in the CeA in both males and females (SHAM=100±10; WAS=50±3; $p<0.001$) and EE able to partially reverse the effects of WAS (EE+WAS=68±11. $p=0.03$; 1-way ANOVA). **Summary and Conclusions**: Our data reveals that a brief exposure to EE is capable of reversing chronic stress-induced visceral hypersensitivity in male and female rats along with a partial restoration of GR expression in the CeA. These findings show that GR plays a critical role in chronic stress induced visceral hypersensitivity. Our data also suggests behavioral therapies may work through GR signaling and additional pathways to normalize stress-induced visceral hypersensitivity.

Disclosures: A. Orock: None. **B. Greenwood-Van Meeveld:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.06

Topic: F.03. Stress and the Brain

Title: Basolateral Amygdala Circuitry and Stress-induced Repetitive Behavior

Authors: *A. GLORIUS[1], J. P. HERMAN[2];
[1]Pharmacol. & Systems Physiol., Univ. of Cincinnati, Reading, OH; [2]Dept Pharmacol. and Systems Physiol., Univ. of Cincinnati, Cincinnati, OH

Abstract: Obsessive Compulsive Disorder (OCD) is a highly disruptive chronic condition that affects approximately 1.2% of the adult U.S. population. OCD is commonly characterized by the presence of thoughts (obsessions) that trigger the execution of repetitive behaviors (compulsions). Rodent models of OCD typically examine highly stereotyped behaviors such as grooming. Self-grooming increases in the presence of stress in numerous rodent models. Mechanisms underlying thus process remain to be delineated. we propose that a stress triggers activation of BLA, synapses in the NAc, which in turn drives a striatonigral pathway that is known to induce grooming. We used tracing approaches in Fischer 344 rats to assess this pathway. Rats underwent stereotaxis procedures and received a unilateral PHA-L (anterograde tracer) iontophoretic injection in the BLA and a complimentary ipsilateral/unilateral red Retrobead (retrograde tracer) injection in the SNr. We identified PHA-L labeled terminals in both the NAc and in the ventromedial aspect of the SNr, indicting the existence of BLA projections. Positive retrograde signal was not found in the NAc but was observed dorsomedial to the BLA in the striatum. Subsequent experiments tested grooming as a parameter for stress-based behavior, as a means of identifying a reproducible model of stress grooming in our lab. Fischer 344 rats (a strain know for high levels of grooming) were exposed to a modified stress enhanced fear learning (SEFL) paradigm following prior stress exposure (restraint stress/elevated plus maze). No significant changes in grooming were seen during initial stress exposures. However, significant increases in grooming duration were observed during SEFL training in groups exposed to prior stress. Additionally, groups not exposed to prior stress showed decreases in grooming duration. The data suggest a lasting impact of stress exposure on fear learning, a process thought to be mediated by the BLA. The data indicate that grooming may be a potential stress-scoping mechanism, possibly triggered by BLA circuitry.

Disclosures: A. Glorius: None. **J.P. Herman:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.07

Topic: F.03. Stress and the Brain

Support: NIH Grant R15 MH125306-01A1
NIH Grant R15 MH104485-01A1

Title: Female social defeat behavior is modified through orexin 2 receptor activity, balancing pro- and anti-stress circuitry in basolateral amygdala

Authors: J. D. W. YAEGER[1], M. M. JOHN[2], L. J. LEDESMA[2], T. R. SUMMERS[2], W. J. KORZAN[3], R. P. WATERS[4], ***C. H. SUMMERS**[2];
[1]Pediatrics and Rare Dis. Group, Sanford Res., Sioux Falls, SD; [2]Dept. of Biol., Univ. of South Dakota, Vermillion, SD; [3]Norwegian Univ. of Life Sci., Oslo, Norway; [4]Univ. of Mary Washington, Univ. of Mary Washington, Fredericksburg, VA

Abstract: Balanced Stress responsiveness is derived from signaling in brain regions associated with the promotion of pro- and anti-stress behavioral output. Biases in these circuits shift behavioral patterns and define phenotypes that exhibit stress resilience or vulnerability. Transcriptomic analyses reveal distinct expression profiles in pro-stress regions, prelimbic cortex and anterior basolateral amygdala of mice exhibiting active avoidance (Escape) or accepting confrontation (Stay) behaviors in a social stress paradigm (Stress Alternatives Model; SAM). Further, intra-BLA manipulation of orexin 2 receptors (Orx_2R) shifts phenotype-specific behaviors. As the orexin system mediates stress responses in a sex-dependent fashion, we developed a model for investigating social stress in female mice using shock-induced aggression (SIA). Unlike males exposed to the SAM paradigm, most females display Escape behavior, which can be altered by including additional social stress bouts to the paradigm to enhance the stress state. Further, female mice possess more BLA cells that express Orx_2R mRNA (*Hcrtr2*) compared to males. Antagonizing Orx_2R subcutaneously at a low dose (30 nmol) in female animals resulted in phenotype divergence with a proportion of mice displaying slower escape ($Escape^S$), a result replicated in all females administered the anxiogenic drug yohimbine (α_2 receptor antagonist). Like yohimbine-treated mice, females of the $Escape^S$ phenotype also showed reduced social preference and enhanced cued fear freezing. Additionally, $Escape^S$ animals had more *Hcrtr2*-positive cells in the BLA. These results suggest that Orx_2R mediate stress responsivity, likely by balancing pro- and anti-stress circuitry.

Disclosures: J.D.W. Yaeger: None. **M.M. John:** None. **L.J. Ledesma:** None. **T.R. Summers:** None. **W.J. Korzan:** None. **R.P. Waters:** None. **C.H. Summers:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.08

Topic: F.03. Stress and the Brain

Support: NIH Grant MH122946

Title: Sex Differences in the Formation and Maintenance of Dominance Relationships in Syrian hamsters

Authors: J. N. KEARNEY, C. J. WHITTEN, M. K. HOOKER, *M. A. COOPER; Psychology, Univ. of Tennessee Knoxville, Knoxville, TN

Abstract: A poor understanding of sex differences in animal models of stress-related psychiatric disorders has contributed to greater misdiagnosis of depression and anxiety disorders in women compared to men. Dominance hierarchies produce experience-dependent neural plasticity and generate individual differences in stress sensitivity such that subordinate animals are often more vulnerable to stress than dominants. However, status-dependent differences in stress sensitivity are primarily limited to findings in males and it is unknown whether differences in dominance relationships between males and females produce sex differences in stress sensitivity. The aim of this study was to characterize sex differences in the formation and maintenance of dominance relationships in same-sex pairs of male and female Syrian hamsters. We age-matched and weight-matched all subjects, and estrus-cycle-matched females to minimize pre-existing differences between subjects. We then exposed hamsters to daily social encounters for two weeks in a resident-intruder format. Because female hamsters show low and inconsistent aggression during estrous, we skipped social encounters every fourth day. We conducted a meta-analysis on agonistic behavior from multiple cohorts of hamsters across several studies. In total, we obtained behavioral data from 68 male dyads and 88 female dyads. We found that female hamsters attacked more quickly and at a higher rate compared to males regardless of dominance status. In addition, resident female hamsters attacked more quickly and at a higher rate than intruder females, but aggression in males did not depend on residency status. Female subordinates submitted more quickly to their dominant counterparts and fled at higher rates from their dominant counterparts compared to male subordinates. Intruder subordinate females were found to submit more quickly and flee at a higher rate than resident subordinate females. Females were also more resistant than males to becoming subordinate in that they fought back more consistently and were more likely to flip their dominance status during the 2 weeks of social encounters. These findings suggest that dominance relationships are less stable in females compared to males and that residency status may have a larger impact on status-dependent neural plasticity in females than in males. In addition, sex differences in the formation and maintenance of dominance relationships may contribute to variation in stress sensitivity between males and females.

Disclosures: J.N. Kearney: None. **C.J. Whitten:** None. **M.K. Hooker:** None. **M.A. Cooper:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.09

Title: WITHDRAWN

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.10

Topic: F.03. Stress and the Brain

Support: NIH Grant MH122946

Title: Experience-dependent Changes in Activation of Infralimbic and Medial Amygdala Neural Ensembles Associated with Stress Coping

Authors: *J. LAYMON, M. HOOKER, A. BREWER, M. COOPER;
Univ. of Tennessee, Knoxville, TN

Abstract: Experience-dependent activity of specific neural ensembles is known to generate individual differences in stress reactivity and can alter risk for stress-related psychiatric disorders. Dominance status involves a critical social experience that modulates neurochemical, neuroendocrine, and behavioral responses to stress. Dominant animals often show greater stress resistance than subordinates. We have shown that dominant hamsters exhibit greater stress-induced neural activity in the infralimbic (IL) region of the ventromedial prefrontal cortex and medial amygdala (MeA). We have also shown that dominants exhibit less stress-induced social anxiety than subordinates. In this project we are using a viral vector approach to label immediate early gene expression during stress to quantify subsequent stress-related behavior in the same animals. We are tagging activated neural ensembles with a robust activity marker (RAM) during an acute social defeat stressor and measuring social exploration 48 hours after stress. Our preliminary data indicate that RAM expression in the IL and MeA can be temporally controlled with doxycycline chow and is activated by a social defeat experience. Specifically, we have shown that defeated animals exhibit greater RAM expression in the IL and MeA compared to non-defeat animals. Also, defeat-induced RAM expression in both the IL and MeA is positively correlated with defeat-induced social exploration 48 hours later. We are currently creating dominance relationships to test whether status-dependent differences in IL and MeA RAM expression correlate with status-dependent differences in defeat-induced social exploration. These findings suggest that elevated neural activity in the IL and MeA during stress predicts reduced social anxiety-like behavior in the future. Also, this study supports the use of a RAM approach for identifying neural ensembles that regulate long-term changes in behavior.

Disclosures: J. Laymon: None. **M. Hooker:** None. **A. Brewer:** None. **M. Cooper:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.11

Topic: F.03. Stress and the Brain

Support: NIH Grant R15MH122946
Ford Fellowship

Title: Androgen receptor-expressing neurons in select neuronal ensembles within the medial amygdala play a role in status-dependent differences in stress responsivity

Authors: *C. WHITTEN, J. E. KING, R. M. RODRIGUEZ, L. M. HENNON, M. K. HOOKER, M. C. SCARBROUGH, M. S. JENKINS, M. A. COOPER;
Dept. of Psychology, Univ. of Tennessee, Knoxville, TN

Abstract: Social stress is a negative emotional experience that leads to individual differences in fear and anxiety. Dominance status is an environmental factor that can alter the way individuals react to and cope with stressful events. The underlying neurobiology of how social dominance produces stress resistance remains elusive, although experience-dependent changes in androgen receptor (AR) expression is thought to play an essential role. Using a Syrian hamster (Mesocricetus auratus) model, we investigated whether dominant individuals activate more AR-expressing neurons in the posterodorsal and posteroventral regions of the medial amygdala (MePD, MePV), activate more neurons in MePD and MePV that project to the bed nucleus of the stria terminalis (BNST), and display less social anxiety following social defeat stress compared to subordinate counterparts. We allowed male hamsters to form and maintain a dyadic dominance relationship for 12 days, exposed them to social defeat stress, and then tested their approach-avoidance behaviors using a social interaction test. During social defeat stress, dominant subjects showed greater c-Fos expression in AR-expressing cells in the MePD/MePV as well as increased c-Fos expression in a MePD/MePV-BNST pathway compared to subordinates. We found that acute social defeat reduced the amount of time animals spent interacting with a novel conspecific, although there was no effect of dominance status, which is in contrast to status-dependent changes in a conditioned defeat response. Also, there were no status-dependent differences in c-Fos expression in MePD/MePV AR-positive neurons during social interaction testing. These findings suggest that activation of select neural ensembles in the MePD/MePV during social defeat stress contributes to status-dependent changes in stress responsivity and gives insight into the neuroendocrine mechanisms by which social experience alters stress coping.

Disclosures: C. Whitten: None. **J.E. King:** None. **R.M. Rodriguez:** None. **L.M. Hennon:** None. **M.K. Hooker:** None. **M.C. Scarbrough:** None. **M.S. Jenkins:** None. **M.A. Cooper:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.12

Topic: F.03. Stress and the Brain

Support: NIH Grant R15 MH122946

Title: The role of the medial amygdala in development of status-dependent differences in stress vulnerability

Authors: J. R. KELLY, C. J. WHITTEN, ***M. K. HOOKER**, J. N. KEARNEY, L. M. HENNON, M. S. JENKINS, M. C. SCARBROUGH, M. A. COOPER;
Dept. of Psychology, Univ. of Tennessee, Knoxville, TN

Abstract: Dominance relationships modulate stress sensitivity in males within a variety of species, however less is known about the consequences of dominance status in females. The posterodorsal and posteroventral medial amygdala (MePD, MePV) are part of a social behavior neural network and plasticity within these regions contribute to experience-dependent changes in behavior. Using a Syrian hamster (Mesocricetus auratus) model, we established dominance relationships among adult females and found that animals showed estrous-dependent variation in agonistic behavior. Following 2 weeks of daily dominance interactions, dominant and subordinate females were exposed to social defeat stress and c-Fos immunoreactivity was quantified in the MePD and MePV. A separate group of females received injection of a retrograde tracer into the bed nucleus of the stria terminalis (BNST) and we quantified stress-induced c-Fos immunoreactivity in a MePD/MePV-BNST pathway. Another group of animals received a social interaction test following social defeat and we quantified defeat-induced social avoidance as well as c-Fos immunoreactivity in the MePD and MePV. We expected that dominant individuals would show greater MePD and MePV activation during social defeat stress and less subsequent social avoidance compared to subordinates. Consistent with this prediction, female subordinates showed less defeat-induced c-Fos expression in the MePV compared to dominant counterparts and controls without a dominance status. However, unlike corresponding data in male hamsters, there was no status-dependent difference in activation of a MePD/MePV-BNST pathway in females. Also, subordinates show greater defeat-induced social avoidance as compared to dominants and controls. However, social interaction testing produced no status-dependent differences in MePD/MePV c-Fos expression. Overall, neural activity in the posterior regions of the medial amygdala contributes to status-dependent differences in the development, but not expression, of stress vulnerability in female hamsters.

Disclosures: J.R. Kelly: None. **C.J. Whitten:** None. **M.K. Hooker:** None. **J.N. Kearney:** None. **L.M. Hennon:** None. **M.S. Jenkins:** None. **M.C. Scarbrough:** None. **M.A. Cooper:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.13

Topic: F.03. Stress and the Brain

Support: NIH-Columbia University Individual Graduate Partnership Program
ZIA MH002970-02

Title: Dynorphin signaling in the central amygdala regulates multivalenced behavioral selection

Authors: *A. LIMOGES[1,2], H. A. TEJEDA[2];
[1]Biol. Sci., Columbia Univ., New York, NY; [2]NIMH, Bethesda, MD

Abstract: The central nucleus of the amygdala (CeA) regulates components of both positively and negatively valenced behaviors via specific cell types and circuits. Past studies on the peptide dynorphin (Dyn) and its cognate receptor, the kappa opioid receptor, within the CeA have highlighted a general role of this system in threat-related behaviors as well as drug-seeking behaviors. However, no studies have investigated the role of Dyn in motivational conflict. Here, we genetically deleted Dyn from the CeA of Dyn-flox mice and assessed the impact on reward, fear, and resolution of conflict between reward and fear. To test whether deletion of CeA Dyn reduces reward-seeking behavior, we trained mice in operant procedures. We observed no differences between groups, indicating that CeA Dyn is not engaged during reward learning. Next, we tested whether CeA Dyn deletion promotes fear learning. CeA Dyn deletion did not robustly modulate fear expression. Finally, to test whether CeA Dyn is critical for reward seeking in the face of threats, we employed a modified platform-mediated conflict task. In this task, tones predicted footshocks, and light cues predicted reward availability. The trials consisted of a mixture of nonoverlapping, partially overlapping, and entirely overlapping cue presentations. Mice also had access to a “safe” platform opposite the reward port, which allowed them to avoid shocks at the expense of rewards. Interestingly, mice with Dyn deletion in the CeA showed enhanced reward-seeking and reduced time on the safety platform relative to controls, suggesting that Dyn signaling limits reward seeking in the face of threats. Ongoing experiments suggest that selective deletion of Dyn from somatostatin-expressing CeA neurons phenocopies the global CeA Dyn deletion described above. Further, we are investigating activity dynamics of CeA Dyn neurons during motivational conflict. Together, our work suggests that Dyn produced by CeA neurons may be preferentially released in situations where appetitive behaviors must be suppressed to avoid threats. The findings of this study may inform our understanding of various psychiatric disorders driven by impaired Dyn signaling or CeA function.

Disclosures: A. Limoges: None. **H.A. Tejeda:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.14

Topic: F.03. Stress and the Brain

Support: NIMH Grant R21MH114182
The PSC-CUNY Awards program

Title: The Contribution of Associative Learning to Stress-Induced Social Avoidance Behavior

Authors: *J. LEE[1], S. HANIF[2], A. AUBRY[3], N. S. BURGHARDT[1,2];
[1]Psychology Program, Behavioral and Cognitive Neurosci., The Grad. Center, CUNY, New York, NY; [2]Dept. of Psychology, Hunter College, The City Univ. of New York, New York, NY; [3]Dept. of Neurosci. and Friedman Brain Inst., Icahn Sch. of Med. at Mt. Sinai, New York, NY

Abstract: Chronic social defeat stress (CSDS) leads to social avoidance, which is often interpreted as depressive-like behavior. This is supported by the finding that stressed C57/BL6J mice avoid CD-1 mice, the strain used to induce CSDS, and mice of the same strain. However, we find that stressed 129Sv/Ev mice only avoid CD-1 mice and not a conspecific, indicating that motivation to socialize is intact. We hypothesize that in this strain, fear conditioning occurs during each social defeat session, such that the CD-1 serves as a conditioned stimulus and the CD-1 attack serves as an unconditioned stimulus, leading to the formation of a fear memory that is retrieved during the social interaction test. If true, then social avoidance should be subject to extinction and second-order fear conditioning, both of which are commonly tested following traditional models of Pavlovian fear conditioning. We tested extinction of social avoidance by exposing defeated mice to 5 social interaction tests/day for 7 consecutive days. A novel mouse was used as the social target for each test. Consistent with extinction learning, we found that interaction time with the CD-1 significantly increased across experimental days (n=10; Day 1 vs. Day 7, $p< 0.01$). In contrast, when the social target was a 129Sv/Ev, interaction time remained high throughout testing (n=9; Day 1 vs. Day 7, $p= 0.62$). In a follow-up experiment, we found that defeated mice avoid a Swiss Webster mouse, but not the scent of a CD-1 (n=20; $p< 0.05$), indicating association of specific visual cues (color) with threat during CSDS. We next tested whether pairing an aggressive CD-1 with a tone (4Khz, 70dB) produces second-order conditioning. On each day of defeat, the last 10 seconds of a tone overlapped with the introduction of a CD-1. At test, the same tone was played when mice entered the interaction zone of an empty wire-mesh enclosure. Defeated mice (n=14) spent significantly less time with the tone-paired cup than non-defeated controls (n=16), indicating that CSDS can elicit avoidance behavior in the absence of a social target ($p< 0.05$). In addition, avoidance of a CD-1 upregulated c-Fos in the basolateral nucleus of the amygdala (defeated n=5, non-defeated n=5; $p< 0.001$), consistent with the known role of this region in retrieval of a conditioned fear memory. Collectively, these results indicate that CSDS may be useful for studying the formation of fear memories involving a social cue.

Disclosures: J. Lee: None. **S. Hanif:** None. **A. Aubry:** None. **N.S. Burghardt:** None.

Poster

552. Stress-Modulated Pathways: Amygdala

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 552.15

Topic: F.03. Stress and the Brain

Title: Psychoactive Treatments in a Mouse Model of PTSD

Authors: *W. DYER[1], N. THOMAS[1], N. MARION[1], A. PRUS[2], A. LACROSSE[2]; [1]Northern Michigan Univ. - Neurosci. Major, Marquette, MI; [2]Psychological Sci., Northern Michigan Univ., Marquette, MI

Abstract: Post-TraumaticStress Disorder (PTSD) can be characterized as a complex mental disorder by itscomplications in cognitive functioning and behavioral deficits often onset by atraumatic event in one's life. Due to the limited efficacy of therapeutic interventions,the current treatment for this disorder includes psychotherapy, exposure-basedtherapy, and medications such as selective serotonin reuptake inhibitors(SSRIs) cultivated for the treatment of anxiety disorders. In recent years,different psychoactive substances with known psychotropic and hallucinogeniceffects have been reviewed as novel treatments for acute stress disorders.Within the field of behavioral neuroscience, fear eliciting paradigms are usedto evoke and represent the acute stress symptoms that facilitate post-traumaticstress disorder. Within this study, sixteen C57/BL adult male mice were exposedto aversive shock stimuli within a novel operant chamber context. Four hoursafter the aversive stimuli, half of the mice were injected with Ketamine30mg/kg intraperitoneally and the other half received a saline injection.Twenty-four hours after the aversive stimuli, the mice were placed back intothe chamber to monitor for symptoms of distress. The mice who received thesaline injection associated the chamber with the aversive stimulus andexhibited freezing behaviors. Additionally, all the mice were placed into anopen field apparatus to assess for symptoms of generalized anxiety disorder.The mice within the Ketamine group showed a reduction in fear generalization.This research concluded that Ketamine serves as a promising therapeuticpsychoactive treatment for anxiety and acute stress-related disorders.

Disclosures: W. Dyer: None. **N. Thomas:** None. **N. Marion:** None. **A. Prus:** None. **A. LaCrosse:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.01

Topic: F.04. Neuroimmunology

Support: Lions Forskningfond postdoc 2021

Title: Lateral habenula regulates peripheral and central immune responses in aversive states

Authors: *S. CASTANY[1], O. BARBA[1,2], J. WISKERKE[1], D. ENGBLOM[1];
[1]Linköping university, Linkoping Univ., Linköping, Sweden; [2]Bordeaux neurocampus, Bordeaux, France

Abstract: Mood can have a great impact on our physical health. Interestingly, depressed patients have increased vulnerability to develop diseases than healthy population. Then, it is highly possible that aversive states such as depression may trigger immune function dysregulation. So far, little is known about which brain regions are involved in immune disfunction and which are the mechanisms. Lateral habenula (LHb) has been postulated to be the "anti-reward" center and its hyperactivity has a key role in the pathophysiology of depression. Therefore, the aim of the present study is to investigate whether lateral habenula participates in the dysregulation of the immune system observed in depressive states. To do that, we used optogenetics to selectively activate the neurons in the Lhb and we checked whether this was enough to trigger an aversive state in C57/bl6 WT males and female mice. Then, we subjected them to a chronic protocol of stimulation (Day 1: 20min, Day2 :40 min, Day 3: 70min at 20Hz-10λ 3s ON7 min OFF). At the end of the protocol, we challenged them with LPS to study the innate immune reactivity in the periphery (eg, spleen, liver, plasma) and centrally (eg brain). Lhb activation induced a robust aversive state, as ChR2-Lhb injected mice decreased time spent in the light paired chamber in the real time place preference paradigm (rtCPP). The aversion induced by Lhb activation was associated with an increase expression of proinflammatory cytokines (IL6, IL1β, TNFα, IFN-γ) in the spleen after LPS challenge, specifically in females. We also studied microglia reactivity in the insular cortex, an area that encodes interoceptive information and it is known to sense peripheral immunity changes. Lhb activation increased microglia reactivity to LPS only in females. Altogether, this data indicates that the specific stimulation of aversive circuitry, in particular the lateral habenula, induces a dysregulation of the innate immune system response both peripherally and centrally after an immune challenge. These findings may contribute to elucidate why depressive states lead so often to increased risk of comorbidities (e.g. cardiovascular problems, autoimmune diseases, or infections).

Disclosures: S. Castany: None. **O. Barba:** None. **J. Wiskerke:** None. **D. Engblom:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.02

Topic: F.04. Neuroimmunology

Support: Faculty start-up funds to LKF
NIH grant R01AG062716 to LKF

Title: Time-restricted feeding reduces neuroinflammation and improves social behavior in aged mice

Authors: *L. M. INCE, J. DARLING, A. PRABHAKAR, R. CHEN, E. R. CARLSON, K. MANEM, S. SRIDHAR, A. A. DABAK, L. K. FONKEN;
Col. of Pharm., Univ. of Texas at Austin, Austin, TX

Abstract: Inflammation is dysregulated with aging, resulting in a chronic low-grade activation of the immune system ('inflammaging'). In the brain, aging is associated with increased microglial reactivity, which correlates with impaired cognition and social withdrawal. Disruption of the circadian system, which drives rhythms of ~24 h throughout the body, also enhances inflammation. In young rodents, the circadian system suppresses microglial inflammatory responses during the active/dark phase, but this time-of-day effect is lost in aged rodents, resulting in tonic elevation of pro-inflammatory cytokine expression. Thus, disruption of circadian rhythms in aging may potentiate neuroinflammation and lead to changes in cognition and social behavior. We hypothesize that measures to bolster circadian rhythms in aging will reduce neuroinflammation and increase social behavior. To test this hypothesis, we employed a time-restricted feeding (TRF, food access limited to the dark/active phase) protocol in aged mice (18 months old) for 6 weeks and compared physiology and behavior to that of *ad lib*-fed age-matched and young (3 months old) controls. There was no effect of TRF on total daily food consumption or on body mass compared to aged *ad lib* mice, indicating that TRF did not serve as a caloric restriction. After six weeks of the feeding paradigm, aged mice significantly reduced time seeking a juvenile conspecific in the juvenile social exploration test (JSE) compared to young controls. TRF ameliorated reductions in social behavior in aged mice, indicating that TRF increased sociability in aged animals. Furthermore, TRF reduced an age-associated increase in hippocampal inflammation, reducing expression of interleukin-6 (*Il6*) during the active phase. These data indicate that TRF improves social behavior in aging, potentially via reducing neuroinflammation. Further work will focus on assessing how TRF modulates age-associated inflammation in the brain and peripheral tissues, and whether the beneficial effects of TRF extend into reduced pathology in rodent models of neurodegenerative disease.

Disclosures: L.M. Ince: None. **J. Darling:** None. **A. Prabhakar:** None. **R. Chen:** None. **E.R. Carlson:** None. **K. Manem:** None. **S. Sridhar:** None. **A.A. Dabak:** None. **L.K. Fonken:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.03

Topic: F.04. Neuroimmunology

Support: Faculty Start-up Funds
NIAAA grant T32 AA007471

Title: Microglia depletion ameliorates neuroinflammation and behavioral changes in Rev-Erba knockout mice

Authors: *R. CHEN, A. WEITZNER, B. ROUTH, K. BELL, J. STRATKER, L. FONKEN; Div. of Pharmacol. and Toxicology, the Univ. of Texas at Austin, Austin, TX

Abstract: The circadian system is an evolutionarily adaptive system that synchronizes biological and physiological activities within the body to the approximately 24 hr oscillations on Earth. At the molecular level, circadian core clock genes act as transcriptional factors that regulate the rhythmic expression of other genes involved in sleep, cognition, mood, and immune function. Nuclear receptor Rev-Erbα/β are transcriptional repressors directly regulated by core clock gene BMAL1 and inhibit expression of *Bmal1* and other gene targets. Global deletion of Rev-Erbα (RKO) promotes NF-κB signaling and activates the brain-resident immune cell microglia. RKO mice have been previously characterized to have a mania-like phenotype and an impaired memory function; however, the mechanisms by which changes in behavior arise remain unclear. Here we hypothesize that microglia mediate RKO-induced neuroinflammation and altered mood. To deplete microglia, RKO mice on a C57BL6/J background and their wild-type littermates were fed a diet containing CSF1R inhibitor PLX5622 (1200 ppm) or a standard lab diet as control. Affective behaviors and cognitive function were tested after 3 weeks of treatment, and neuroinflammatory outcomes were subsequently assessed in the hippocampus. RKO-induced hyperactivity in an open field and elevated plus maze test and increases in activity were ameliorated by microglia depletion in male but not female RKO mice. RKO induced differential anxiety-like phenotype in an open field and an elevated plus maze. Microglia depletion ameliorated anxiety-like behaviors in RKO females in an open field but did not alter risk-taking behaviors in an elevated plus maze. Moreover, microglia depletion ameliorated increases in pro-inflammatory cytokine expression (e.g., IL-1β and TNFα) in the hippocampus of male but not female RKO mice. Taken together, we showed that microglia were involved in Rev-Erbα-mediated neuroinflammation and mood regulation. These findings support that Rev-Erbα may serve as the link among the circadian system, the immune system, and mood regulation.

Disclosures: R. Chen: None. **A. Weitzner:** None. **B. Routh:** None. **K. Bell:** None. **J. Stratker:** None. **L. Fonken:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.04

Topic: F.04. Neuroimmunology

Support: NIH-NCI Cancer Center Support Grant P30 CA016059
R00DA045795 (NIDA)
P30DA033934 (NIDA)
VCU SoM Starup Funds

Title: Zfp189-mediated transcription regulates social behavior via activation of immunity genes in the prefrontal cortex

Authors: *N. L. TRUBY[1], G. SILVA[1], R. K. KIM[2], J. PICONE[2], R. L. NEVE[3], X. CUI[1], P. J. HAMILTON[4];
[1]Virginia Commonwealth Univ., Virginia Commonwealth Univ., Richmond, VA; [2]Virginia Commonwealth Univ. Neurosci. Grad. Program, Virginia Commonwealth Univ. Neurosci. Grad. Program, Richmond, VA; [3]Gene Delivery Technol. Core, Massachusetts Gen. Hosp., Boston, MA; [4]Virginia Commonwealth Univ. Hlth. Syst., Virginia Commonwealth Univ. Hlth. Syst., Richmond, VA

Abstract: Earlier work has correlated the expression of *Zfp*189 within mouse prefrontal cortical neurons (PFC) with enhanced behavioral resilience to social stress. However, the exact mechanisms through which *Zfp*189 is able to mediate social behavior remains unclear. The *Zfp*189 gene product is a Krüppel associated box (KRAB) zinc finger transcription factor of unknown function. To directly interrogate the transcriptional function and gene targets of ZFP189, we reprogrammed the endogenous ZFP189^{WT} by replacing the repressive KRAB domain with an enhanced transcriptional activation domain (VP64-p65-Rta (ZFP189^{VPR}) or by removing the functional moiety entirely (ZFP189^{DN}). We demonstrate divergent transcriptional regulation at a luciferase target gene, in vitro. Upon packaging these ZFP189 variant constructs in herpes viral vectors (HSVs) and delivering to mouse PFC, we observe that mice infected with HSV-ZFP189^{VPR}, the synthetic transcription factor of inverted transcriptional function relative to the pro-resilient ZFP189^{WT}, demonstrate dramatically reduced social behaviors. The infected PFC tissues were then microdissected and subjected to RNA-sequencing (RNAseq), providing insight into which ZFP189-gene targets might be mediating these effects. Lastly, we investigated the consequences of altered ZFP189-mediated transcriptional function on dendritic spine density and morphology in cortical pyramidal neurons. We observe that viral expression of ZFP189^{WT} or the synthetic ZFP189^{VPR} results in mature dendritic spines on pyramidal neurons. However, only the synthetic ZFP189^{VPR} precipitates pronounced social withdrawal in these mice. In performing RNAseq with these tissues, we identify a divergent impact on genes relating to adaptive immune response and implicate interferon-regulatory factors as ZFP189-sensitive mediators of social behavior. Collectively this body of work indicates that ZFP189 regulates social behavior by functioning as a transcriptional repressor at key adaptive immune target genes in PFC cortical neurons, which drive dendritic spine maturity and pro-social behaviors.

Disclosures: **N.L. Truby:** None. **G. Silva:** None. **R.K. Kim:** None. **J. Picone:** None. **R.L. Neve:** None. **X. Cui:** None. **P.J. Hamilton:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.05

Topic: F.04. Neuroimmunology

Support: UM Pepper Center grant, AG024824
Michigan Institute for Clinical and Health Research (MICHR) grant,

UL1TR002240
Michigan Alzheimer Disease Center (MADRC) grant, P30 AG053760

Title: Covid-19-like inflammation causes persistent memory deficits in aging mice.

Authors: H. K. CHOI[1], P. L. KE-LIND[2], K. M. SCHUH[3], *N. C. TRONSON[4];
[1]Psychology, [2]Psych, Univ. of Michigan, Ann Arbor, MI; [3]Univ. of Michigan Psychology Grad. Program, Ann Arbor, MI; [4]Univ. of Mchigan, Ann Arbor, MI

Abstract: COVID-19 has affected more than 540 million individuals worldwide, and up to 40% of survivors experience post-acute covid sequelae (PASC, “long COVID”). The symptoms of long COVID include “brain fog” and cognitive impairments, and mood related symptoms such as depression or anxiety. SARS-COV-2 virus only rarely infects the brain, suggesting that other effects of COVID-19 cause changes including memory impairments. Inflammation during illness is known to modulate memory, cognition, and mood. Recent studies demonstrate that effects of inflammation on memory and neuroimmune priming can last months after resolution of immune signaling. Importantly, SARS-COV-2 is a single stranded RNA (ssRNA) virus triggers innate immune activation via Toll-Like Receptor (TLR) 7 and TLR8. In comparison to TLR4 and TLR3 activation are widely studied (with LPS and poly I:C, respectively), the effects of TLR7-triggered inflammation in the brain are not well studied. In this project we examined the hypothesis that TLR7-triggered immune challenge causes lasting changes in the brain that contribute to the cognitive impairments and “brain fog” observed in Long COVID. We used a subchronic immune challenge protocol, recently developed in our laboratory, to determine whether the TLR7 agonist R848 (400-1000μg/kg) causes memory impairments or anxiety- and depression-like phenotypes that emerge and persist at least 8 weeks after the 2 week immune challenge. We observed a clear dose-response of cytokine and chemokine elevations in the hippocampus of young and mid-aged male and female mice; and mild weight loss to successive doses of R848, particularly in males. Eight weeks after immune challenge, we observed impairments in hippocampal-dependent novel object recognition in both sexes, and decreased context fear conditioning only in male mice - a pattern that mimics that observed with subchronic poly I:C challenge. These effects on memory were not due to lingering elevations in neuroimmune activation or changes in locomotor activity. This work is an initial step towards understanding how inflammatory sequelae of COVID-19 and other ssRNA viral illnesses contribute to cognitive effects of post-viral syndromes, and may, contribute to increased risk for aging-relate cognitive decline and Alzheimer’s Disease in the decades to come.

Disclosures: H.K. Choi: None. **P.L. Ke-Lind:** None. **K.M. Schuh:** None. **N.C. Tronson:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.06

Topic: F.04. Neuroimmunology

Support: Eunice Kennedy Shriver National Institute of Child Health and Human Development of the National Institute of Health under award number R25HD090723-02.
University of Massachusetts Boston, Research Trust Fund Dissertation Grant

Title: Exploring the effects of trait anxiety-like behavior and neuroimmune substrates on the expression of AMPH sensitization in adolescent Long-Evans rats

Authors: *C. CALHOUN[1], S. ROY[2], B. RICH-PLOTKIN[2], L. PINA[2], A. ALVES[2], T. DONALDSON[2];
[1]Neurosci., Albert Einstein Col. of Med., New York City, NY; [2]Univ. of Massachusetts Boston, Boston, MA

Abstract: Repeated drug exposure during adolescence poses a developmental challenge that can permanently alter neurobehavioral responses to drugs and shift an individual into a vulnerable state to develop substance use disorders (SUDs). Neuroimmune substrates, such as toll-like receptor 4, modulate the progressive stages of learning that occur within SUDs. Activated TLR4 mounts a positive amplification cascade that converges on transcription factors to influence pro-inflammatory cytokine activity. The current study explores the roles of trait anxiety and the TLR4 cascade in adolescent amphetamine (AMPH) sensitization. Two cohorts of 15 adolescent male Long-Evans rats (N=30) screened for high (HAn) and low (LAn) anxiety-like behavior were systemically injected with AMPH (4 mg/kg/ml, IP) or isotonic saline (0.9% NaCl, IP) 1X a day, every other day, for a total of four days. Following a one-week withdrawal period, all animals were challenged with a low AMPH dose (1 mg/kg/ml, IP) and evaluated for AMPH-induced locomotor sensitization. Brain tissues were immunohistochemically stained for anti-TLR4, glial-derived neurotrophic factor (GDNF), nuclear factor kappa B (NF-κB), and tumor necrosis factor alpha (TNF-α) proteins. Positive cell counts were recorded within the medial prefrontal cortex (mPFC), caudate putamen (cPU), nucleus accumbens (NAcc), and basolateral amygdala (BLA). HAn rats traveled more distance than LAn rats over the course of the experiment; an effect that was further potentiated by AMPH exposure. To further delineate trait differences independent of drug effects, AMPH values were divided by SAL values to yield AMPH/SAL ratios for HAn and LAn groups that were used in analyses. Over time, HAn rats displayed a higher AMPH/SAL ratio than LAn rats for total distance traveled and stereotypies. In accordance, after a one-week withdrawal period, HAn animals displayed increased sensitized responses indicated by higher AMPH/SAL ratios for total distance traveled, rears, and stereotypies. Next, anti-TLR4 protein counts were not different between HAn and LAn animals, but TLR4-downstream molecules were. That is, HAn animals expressed more NFkB protein than LAn animals within the mPFC. Further, HAn SAL-treated animals expressed more TNFa than LAn drug-treated animals and HAn animals expressed more GDNF than LAn animals. The aforementioned results combine to highlight an increased susceptibility for extreme trait anxiety-like behavioral phenotypes to AMPH sensitization and neuroimmune substrates may modulate this effect.

Disclosures: C. Calhoun: None. **S. Roy:** None. **B. Rich-Plotkin:** None. **L. Pina:** None. **A. Alves:** None. **T. Donaldson:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.07

Topic: F.04. Neuroimmunology

Support: NIH Grant MH114049

Title: Contribution of microglia and basolateral amygdala IL-1 on formation of fear memories

Authors: A. KULP, B. LOWDEN, S. CHAUDHARI, C. RIDLEY, T. WESLEY, D. F. BARNARD, M. HAUCK, ***J. JOHNSON**;
Kent State Univ., Kent, OH

Abstract: Exposure to stress can activate microglia and cause an increase in brain interleukin-1beta (IL-1). In the hippocampus, low concentrations of IL-1 facilitate memory formation while high concentrations inhibit. The amygdala is extremely responsive to stress and the basolateral amygdala (BLA) is critical for associative learning. Here, we tested the hypothesis that activation of microglia and low-grade production of IL-1 in the BLA facilitates contextual fear memory. Fischer male rats were exposed to 2 footshocks over 5min in an operant box (conditioning paradigm). Conditioned animals euthanized 1h later showed significant increases in IL-1 mRNA in the BLA compared to unconditioned controls. Administration of minocycline, a microglia inhibitor, prior to fear conditioning impaired contextual fear memory as measured by a reduction in freezing behavior when placed back into the context one day later. Administration of the IL-1 receptor antagonist (IL-1RA) into the BLA prior to conditioning had no effect on the expression of contextual fear memory. Administration of IL-1 into the BLA prior to fear conditioning resulted in a dose-dependent impairment in contextual fear memory formation. Collectively, the results indicate that microglia are critical for normal formation of contextual fear memory, but IL-1β itself within the BLA dampens contextual fear memory formation.

Disclosures: A. Kulp: None. **B. Lowden:** None. **S. Chaudhari:** None. **C. Ridley:** None. **T. Wesley:** None. **D.F. Barnard:** None. **M. Hauck:** None. **J. Johnson:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.08

Topic: F.04. Neuroimmunology

Support: National Institute on Aging R15AG052935

Title: Sex-dependent associative learning deficits following systemic immune activation

Authors: *T. G. HOEFLER[1], R. K. PATEL[2], N. T. PIROZZI[2], K. L. COUSINS[2], R. A. KOHMAN[1];
[1]Psychology, [2]Biol., Univ. of North Carolina Wilmington, Wilmington, NC

Abstract: Sex differences in immune function produce differential susceptibility to select illnesses and alterations in the inflammatory response. Inflammation can impair learning and memory processes but whether these deficits vary between males and females has not been directly evaluated. However, independent papers by Sparkman et al. (2005a and 2005b) suggest potential sex differences in inflammation-induced cognitive deficits. While male mice showed spatial learning deficits following administration of the bacterial endotoxin lipopolysaccharide (LPS), a separate study with female mice found no evidence of LPS-induced spatial learning or memory deficits. To determine whether males and females show differential cognitive deficits following an immune challenge, adult C57BL6/J mice received an intraperitoneal injection of LPS (250 µg/kg) or saline four hours prior to testing in a two-way active avoidance conditioning task. We hypothesized that LPS administration would impair associative learning in male, but not female, mice. The results showed that LPS-treated males performed significantly fewer avoidance responses and had lower discrimination index scores compared to saline-treated males. However, LPS did not alter acquisition in females, as saline- and LPS-treated females showed similar rates of avoidance responses and discrimination scores across the five days of testing. Additional work is currently in progress to quantify cytokine levels among other factors to determine whether these sexually divergent effects relate to differences in the inflammatory response. Presently, the data indicate that males are more vulnerable to LPS-induced cognitive deficits compared to females.

Disclosures: T.G. Hoefler: None. **R.K. Patel:** None. **N.T. Pirozzi:** None. **K.L. Cousins:** None. **R.A. Kohman:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.09

Topic: F.04. Neuroimmunology

Support: NIH Grant R01MH114882-01
NIH Grant R01MH104559
NIH Grant R01MH127820
CIHR Postdoctoral Fellowship 201811MFE-414896-231226

Title: Chronic social defeat stress increases intestinal permeability and inflammation in mice

Authors: *K. L. CHAN[1], L. LI[4], L. F. PARISE[5], F. CATHOMAS[2], K. B. LECLAIR[6], Y. SHIMO[7], R. DURAND-DE CUTTOLI[2], A. V. AUBRY[8], H.-Y. LIN[2], C. YUAN[2], M. P. KASTER[2], J. WANG[2], S. J. RUSSO[3];
[1]Icahn Sch. of Med. at Mount Sinai, [3]Neurosci., [2]Icahn Sch. of Med. at Mount Sinai, New York, NY; [4]Dept. of Neurosci., Icahn Sch. of Med. At Mount Sinai Grad. Training Program In Neurosci., New York, NY; [5]Neurosci., Mt.Sinai Sch. of Med., New York, NY; [6]Neurosci., [7]Icahn Sch. of Med. At Mount Sinai, New York, NY; [8]Icahn Sch. of Med., Astoria, NY

Abstract: BACKGROUND: Major depressive disorder (MDD) represents the leading cause of disability, affecting over 300 million people worldwide. Largely characterized by behavioral symptoms, there is a need to identify biological changes associated with MDD. Emerging literature recognize a correlation between MDD and indices of inflammation, such as increased circulating leukocytes and cytokines. However, it is not fully known how this inflammation is initiated. Recently, several chronic inflammatory conditions have been associated with increased intestinal permeability or 'leaky gut'. We hypothesize that chronic stress compromises the gut barrier, leading to translocation of gut microbial byproducts into circulation, triggering the inflammation associated with depression-like behavior.
METHODS: To model depression-like behavior in mice, a 10-day chronic social defeat stress (CSDS) model was used. Social behavior was assessed by conducting a social interaction (SI) test. We subsequently measured gut inflammation by flow cytometry, and intestinal permeability by orally gavaging mice with FITC-Dextran.
RESULTS: Following CSDS, intestinal permeability and circulating bacterial endotoxin levels were elevated in stressed mice, with both measurements negatively correlating with individual SI ratio. Additionally, expression of several tight junctions was downregulated in the intestines from defeated mice. Evaluating gut inflammation, pro-inflammatory $IFN\gamma^+$ T cells were upregulated, and anti-inflammatory $IL4^+$ T cells were downregulated in the colon after CSDS. Using ITGβ7-deficient mice, which have impaired immune cell migration to the gut, we find that gut inflammation precedes permeability and behavioral defects during stress.
CONCLUSIONS: Collectively, these results reveal that CSDS induces intestinal inflammation and barrier breakdown, which may promote systemic inflammation associated with depression-like behavior.

Disclosures: K.L. Chan: None. **L. Li:** None. **L.F. Parise:** None. **F. Cathomas:** None. **K.B. LeClair:** None. **Y. Shimo:** None. **R. Durand-De Cuttoli:** None. **A.V. Aubry:** None. **H. Lin:** None. **C. Yuan:** None. **M.P. Kaster:** None. **J. Wang:** None. **S.J. Russo:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.10

Topic: F.04. Neuroimmunology

Support: SNF Early Postdoc Mobility (FK)
Novartis Foundation for Medical-Biological Research (FK)

Title: Inter-relationship of inflammatory biomarkers in bipolar disorder

Authors: *F. KLAUS[1], Z. WU[2], A. N. SUTHERLAND[1], B. SOONTORNNIYOMKIJ[1], L. T. EYLER[1];
[1]Dept. of Psychiatry, Univ. of California San Diego, La Jolla, CA; [2]Dept. of Physics, Univ. of California San Diego, La Jolla, CA

Abstract: Converging evidence suggests a dysregulation in peripheral inflammatory markers in Bipolar Disorder (BD). Alterations of immune-inflammatory pathways are associated with acute mood episodes and there may be a cumulative burden over time, with a chronic inflammation in later disease stages. A dysfunctional immune response leads to an overproduction of pro-inflammatory mediators and associated local and system-wide damage. However, often only alterations in individual biomarkers are investigated, which might not capture inter-and intra-individual variability in inflammatory markers. We therefore aimed to investigate the inter-relationship between peripheral inflammatory markers in bipolar disorder (BD) compared to healthy control participants (HC). We investigated data from 59 BD and 96 age-and gender matched HC participants (age range: 25-65 years). All participants were taking part in a 4-year longitudinal study of cognitive aging; data from the baseline visits, conducted over a 2-week period, were analyzed and means across the three baseline visit computed. Blood was drawn at the same time of day from each participant on three occasions one week apart, and 18 inflammatory markers in peripheral blood were analyzed: BDNF, CCL11, CCL26, IP10, MCP1, MDC, MIP1b, VEGF, Fractalkine, IFN**γ**, IL6, IL8, IL10, TNF**α**, SAA, ICAM1, VCAM1 (High sensitivity multiplex panels (MESO SCALE DISCOVERY®), lymphocyte, WBC counts and CRP (high sensitivity ELISA). Participants with CRP > 10mg/l or WBC> 11'000/ul were excluded. We calculated correlation matrices between all biomarkers. Differences (d) between the correlation matrices between the two groups were examined, which yielded significant differences in the inter-relationship between the various cytokines between HC and BD (X^2 (N = 150) = 72.3, $p < 0.001$). Pairwise relationships between IL8 and CCL11 (d=-2.85, p=0.002), IP10 and CCL11 (d=-2.45, p=0.007), SAA and IFN**γ** (d=2.65, p=0.004) showed the three biggest group differences such that there was a significant positive relationship in IL8 -CCL11 and IP10 -CCL11 in BD (r_s=0.49, p=0.0001, resp. r_s=0.29, p=0.03) but not in HC (r_s=0.04, p=0.67, resp r_s=-0.13, p=0.23), and a positive relationship in HC (r_s=0.31, p=0.003) but not in BD (r_s=-0.14, p=0.30) in SAA and IFN**γ**. These results provide evidence for possible differences in the inter-relationship between inflammatory biomarkers in bipolar disorder in comparison to HC participants, pointing towards a focus on the involvement of altered relationships between inflammatory markers.

Disclosures: F. Klaus: None. **Z. Wu:** None. **A.N. Sutherland:** None. **B. Soontornniyomkij:** None. **L.T. Eyler:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.11

Topic: F.04. Neuroimmunology

Support: This study was funded by a R01MH106553 grant to JMS.

Title: Effects of prenatal maternal immune activation with lipopolysaccharide on neonatal reflex behaviors, maternal care, and neural development in rats

Authors: *M. B. HALL, E. RODRIGUEZ, E. LEMANSKI, D. WILLIS, J. M. SCHWARZ; Psychological and Brain Sci., Univ. of Delaware, Newark, DE

Abstract: Epidemiological evidence suggests that maternal immune activation during gestation increases the risk for offspring to experience symptoms of developmental disorders later in life. Notably, males are more likely than females to be diagnosed with neurodevelopmental disorders, including autism spectrum disorder, schizophrenia, and general learning disabilities. The goal of this project is to elucidate how maternal immune activation (MIA) with lipopolysaccharide (LPS) may lead to changes in brain and behavioral processes in male and female offspring. Sprague-Dawley dams were injected with LPS (50ug/ml/kg, i.p.) or saline on embryonic day (E)15. After birth, the ontogeny of reflex behaviors - such as righting, cliff avoidance, eye opening, gait, posture, and forelimb/hindlimb grasping - were measured daily from P3-P21 in two male and two female (scores averaged for N=1/sex) offspring per litter. Moreover, maternal care toward pups was observed twice a day on postnatal days (P)1-5, P10, P15, and P20, and scored for behaviors such as arch-back nursing, passive nursing, licking pups, etc. In a separate cohort of offspring, pups were euthanized on P7, P15, or P21 and half brains were dissected to collect medial prefrontal cortex, amygdala, dorsal hippocampus, and ventral hippocampus. The effect of E15 LPS on cytokine expression in these brain regions, across postnatal development, was examined using quantitative real-time polymerase chain reaction (qRT-PCR). Results show that offspring who experienced E15 LPS acquired successful forelimb and hindlimb grasping reflexes significantly earlier in development than those that received E15 saline. There were no differences in acquisition of neonatal reflexes between males or females. The effects of E15 LPS on maternal care and on the expression of glial and neural markers in postnatal brains across development will be discussed. Future studies will examine the effects of MIA on maternal and fetal cytokine expression and on adult learning, social, and anxiety behaviors. These findings will provide us with a better understanding of how early-life environmental factors affect later-life brain and behavioral processes, and how they may be differently dysregulated in males and females.

Disclosures: M.B. Hall: None. **E. Rodriguez:** None. **E. Lemanski:** None. **D. Willis:** None. **J.M. Schwarz:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.12

Topic: F.03. Stress and the Brain

Support: Margolis Foundation Pilot Funds

Title: Housing rats at moderate altitude causes systemic inflammation and alters cognitive behavior

Authors: *S. KANEKAR[1,2], R. ETTARO[1], P. RENSHAW[1,3];
[1]Psychiatry, Univ. of Utah, Salt Lake City, UT; [3]Mirecc, [2]US department of Veterans Affairs, Salt Lake City, UT

Abstract: Risk for major depressive disorder (MDD) increases with living at altitude, and hypobaric hypoxia may play a role. Residents at 4,500ft show deficits in blood oxygen levels and brain energetics vs. those at sea level. We developed a sex-based animal model to study depression at altitude. Rats housed at moderate altitude (4,500ft, 10,000ft) exhibit low brain energetic markers and monoamines vs. those at sea level. Rats at altitude also show symptoms of anhedonia, depression and anxiety. MDD is linked to issues in decision-making, problem-solving and cognitive control. Furthermore, both MDD and hypoxia are linked to inflammation. In this study, we therefore asked whether housing at altitude may alter cognitive function and inflammatory status. Male (M) and female (F) rats were housed for 2wks at sea level conditions (hyperbaric chamber) or at 4,500ft (Salt Lake City, UT). Rats were tested for cognitive behavior in the novel object recognition test. On day 1, rats are habituated to the novel field. On day 2, the rat is placed in the box with 2 identical objects at 2 corners (sample trial). On day 3, the rat is placed in box with a novel object replacing one of the original (familiar) objects (choice trial). Time spent with each object, number of contacts made and latency to first contact are measured. Rodent serum was tested by ELISA for the inflammatory cytokine, interleukin 6 (IL6). Cognitive Function: In sample trial, rats did not differ with altitude in exploration, latency to first contact or frequency of contact with either object. In the choice trial, rats at sea level spent more time with the novel object (F-96sec, M-83sec) vs. the familiar one (F-88sec, M-63sec), as expected. However, rats at altitude spent less time exploring the novel object (F-54sec, M-31sec) vs. the familiar one (F-93sec, M-52sec). Time spent with the novel object was significantly lower in males at altitude vs. sea level (Student's t-test, $p=0.03$, $n=5$ea), and shows a trend to be lower in females at altitude ($p=0.06$). Rats at sea level show much shorter latency to first contact of novel object (M, F-2sec) vs. the familiar one (F-11sec, M-37sec). In contrast, rats at altitude show longer latency to first contact of both the novel (F-11sec, M-17sec) and familiar objects (F-17sec, M-22sec). Altitude groups do not differ in motor function and do not show signs of neophobia (all rats approach the novel object before the familiar one). Inflammation: Serum IL6 was significantly higher at altitude vs. at sea level in females (Student's t-test, $p=0.04$, $n=6$ea) and shows a similar trend in males ($p=0.07$). These data indicate that housing at moderate altitude may increase systemic inflammation and alter cognitive function.

Disclosures: S. Kanekar: None. **R. Ettaro:** None. **P. Renshaw:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.13

Topic: G.05. Mood Disorders

Support: Transpharmation Ltd

Title: Lipopolysaccharide (LPS) administration by intraperitoneal (i.p.) injection induces neuroinflammation and anhedonia-like behaviour in rodents

Authors: F. DUNPHY-DOHERTY[1], L. DAVISON[1], ***J. PRENDERVILLE**[2];
[1]Transpharmation Ltd, Dublin, Ireland; [2]Transpharmation, Ltd., Dublin, Ireland

Abstract: Neuroinflammation has been linked to the pathogenesis and treatment of psychiatric disorders, including major depressive disorder (MDD). Anhedonia is a core symptom of psychiatric disorders and people with MDD commonly show motivational impairments. Tasks measuring effort-based functions have been suggested as models for these motivational symptoms. LPS is a component of the outer membrane gram-negative bacteria and has been shown to induce inflammation and behavioural changes in mammals. The effect of peripheral LPS administration on central nervous system (CNS) inflammation in mice and motivation in rats was assessed. Male C57BL/6J mice (n=10 per group) received an i.p. injection of saline or LPS doses (0.25mg/kg). Plasma and brain were collected 4 h after LPS administration and cytokines (TNF-α, IL-6, IL-1β) were analysed using Meso Scale Discovery multiplexed assays. Male Sprague Dawley rats (n=10 per group) received an i.p. injection of saline or LPS (0.25mg/kg). Motivation was assessed in the progressive ratio (PR) task at 2, 24 and 48 h after administration. TNF-α, IL-6, IL-1β were significantly increased in mouse plasma and brain 4 h after LPS administration (Student's t test, $p<0.001$). In the rat PR task LPS induced a significant decrease in breakpoint and lever press at 2 ($p<0.001$) and 24 ($p<0.01$) h after administration in comparison to the saline group (one-way repeated measures ANOVA, Fisher's LSD). No significant difference was observed between the LPS group and the saline group 48 h after administration. The establishment of preclinical models of neuroinflammation-induced behavioural changes representative of depressive symptomatology is required to advance drug development. LPS administration by peripheral injection (i.p.) in mice induced a robust inflammatory response in plasma and the CNS. In rats, i.p. administration of LPS induced deficits in motivation in the PR task during the sickness behaviour period (2 h after administration). Motivational deficits were also observed 24 h after administration, a timepoint previously shown to be optimal for assessing depressive-like behaviour in rodents following an inflammatory stimulus. These data suggest that peripheral administration of LPS can be used to model neuroinflammation and anhedonia-like behavioural changes of relevance to MDD.

Disclosures: F. Dunphy-Doherty: A. Employment/Salary (full or part-time):; Transpharmation Ltd. **L. Davison:** A. Employment/Salary (full or part-time):; Formerly Transpharmation Ltd. **J. Prenderville:** A. Employment/Salary (full or part-time):; Transpharmation Ltd.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.14

Topic: G.05. Mood Disorders

Support: NIH R01-MH112356-01

Title: The effect of L. reuteri on social behavior is independent of the adaptive immune system

Authors: *I.-C. WANG[1], S. W. DOOLING[1], M. SGRITTA[2], A. ROCHA FARIA DUQUE[3], M. COSTA-MATTIOLI[1];
[1]Baylor Col. of Med., Houston, TX; [2]Baylor Col. Of Med., Houston, TX; [3]Dept. of Food and Nutr., Sao Paulo State Univ., Araraquara, Brazil

Abstract: Gut microbes can modulate almost all aspects of host physiology throughout life. As a result, specific microbial interventions are attracting considerable attention as potential therapeutic strategies for treating a variety of conditions. Nonetheless, little is known about the mechanisms through which many of these microbes work. Recently, we and others have found that the commensal bacteria *L. reuteri* reverses social deficits in several mouse models (genetic, environmental and idiopathic) for neurodevelopmental disorders in a vagus nerve-, oxytocin- and biopterin-dependent manner. Given that gut microbes can signal to the brain through the immune system and *L. reuteri* promotes wound healing via the adaptive immune response, we asked whether the prosocial effect mediated by *L. reuteri* also depends on adaptive immunity. Here, we found that the effects of *L. reuteri* on social behavior and related changes in synaptic function are independent of the mature adaptive immune system. Interestingly, these finding indicate that the same microbe (*L. reuteri*) can affect different host phenotypes through distinct mechanisms.

Disclosures: I. Wang: None. **S.W. Dooling:** None. **M. Sgritta:** None. **A. Rocha Faria Duque:** None. **M. Costa-Mattioli:** None.

Poster

553. Neuroimmune Function and Behavior

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 553.15

Topic: H.08. Learning and Memory

Support: NIH Grant DE17794
NIH Grant AR069861
HHMI

Title: Immune checkpoint PD-1 in hippocampal neurons regulates neuronal excitability, synaptic plasticity, and memory

Authors: *J. ZHAO[1], S. BANG[2], K. FURUTANI[3], A. MCGINNIS[1], C. JIANG[2], A. ROBERTS[2], C. R. DONNELLY[4], Q. HE[2], M.-C. KO[5], R. D. PALMITER[6], R.-R. JI[7]; [1]Duke Univ., Durham, NC; [3]Duke Univ. Med. Center, Ctr. for Translational Pain Medicine, Dept. of Anesthesiol., [4]Dept. of Anesthesiol., [2]Duke Univ. Med. Ctr., Durham, NC; [5]Wake Forest Univ. of Med., Winston Salem, NC; [6]Univ. of Washington, Seattle, WA; [7]Pain Res. Division, Anesthesiol., Med. Ctr., Durham, NC

Abstract: Programmed death protein 1 (PD-1) is an immune checkpoint that plays important roles in disease, e.g., cancer and Alzheimer's disease. Immunotherapy using monoclonal antibodies against PD-1 demonstrated improved survival in cancer patients through immune activation. We recently demonstrated that neurons in the PNS (e.g., dorsal root ganglion, DRG) and CNS (e.g., spinal cord) express *Pdcd1* mRNA and functional PD-1 protein. Notably, PD-1 functions to suppress neuronal excitability, as *Pdcd1*-deficient mice show hyperexcitability in DRG nociceptive sensory neurons, leading to pain hypersensitivity and impaired morphine analgesia in mice. Single-cell analysis revealed low expression of *Pdcd1* mRNA in excitatory cortical neurons. To determine the specific role of PD-1 in immune cells, glial cells, and neurons, we removed PD-1 function selectively in neurons or microglia by generating *Pdcd1* conditional knockout (cKO) mice. Using global and cKO mice, anti-PD-1 immunotherapy, and NHP brain slices, we show that functional PD-1 is expressed in mouse and primate hippocampal neurons and PD-1 inhibition improves cognition in physiological conditions. Mice lacking the *Pdcd1* gene encoding PD-1 exhibit enhanced long-term potentiation (LTP) and learning and memory. These behavioral and cellular changes can be recapitulated by selective deletion of *Pdcd1* in hippocampal excitatory neurons but not in microglia. Perfusion of mouse or nonhuman primate brain slices with anti-PD-1 antibody is sufficient to increase excitability in CA1 hippocampal neurons. Conversely, re-expression of *Pdcd1* in PD-1 deficient hippocampal neurons suppresses memory and LTP. These findings suggest that anti-PD-1 treatment has therapeutic potential to counteract cognitive decline.

Disclosures: J. Zhao: None. **S. Bang:** None. **K. Furutani:** None. **A. McGinnis:** None. **C. Jiang:** None. **A. Roberts:** None. **C.R. Donnelly:** None. **Q. He:** None. **M. Ko:** None. **R.D. Palmiter:** None. **R. Ji:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.01

Topic: F.07. Biological Rhythms and Sleep

Support: CONACYT-254264

Title: Chronic sleep restriction increases alcohol intake in male Wistar rats

Authors: *F. GARCIA-GARCIA, L. LÓPEZ-MUCIÑO, F. BRAVO-GONZALEZ, M. ACOSTA-HERNANDEZ, J. RODRIGUEZ-ALBA;
Inst. de Ciencias de la Salud, Univ. Veracruzana, Xalapa, Mexico

Abstract: The reduction of sleep hours is a risk factor for developing cardiovascular, metabolic, and psychiatric problems. Previous studies have shown that poor sleep quality is a factor that favors relapse in addicted patients. In rodents, sleep deprivation increases the preference for methylphenidate and cocaine self-administration. However, it is unknown if chronic sleep restriction induces alcohol intake and delta FosB as an addiction marker in the brain rewarding system. Therefore, the objective of the present study was to evaluate alcohol consumption in a chronic sleep restriction model and determine the expression of delta FosB in adult brain rats. For this purpose, male Wistar rats (300-350 g body weight) were used, which were divided into four experimental groups (n = 6 each group): control without manipulation, sleep restriction (RS) for seven days, RS and alcohol exposure, and a group with only alcohol exposure. At the end of the manipulation, the rats were sacrificed, and the brains processed for the immunohistochemical detection of delta FosB. The results showed that SR elicits alcohol consumption compared to unrestricted sleep rats, and a significant increase in the number of positive cells to delta FosB in the brain nuclei of the motivation/brain reward circuit. In conclusion, sleep restriction increases alcohol consumption, as well as the expression of biological markers associated with addiction. Thus, sleep could be present as a potential protective factor against the development of addiction.

Disclosures: F. Garcia-Garcia: None. **L. López-Muciño:** None. **F. Bravo-Gonzalez:** None. **M. Acosta-Hernandez:** None. **J. Rodriguez-Alba:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.02

Topic: F.07. Biological Rhythms and Sleep

Support: VA Biomedical Laboratory Research and Development Service Merit Award I01 BX001404 (R.B.)
VA Biomedical Laboratory Research and Development Service Merit Award I01 BX4500 (J.M.M)
VA CDA IK2 BX004905 (D.S.U.)
NIH R01 NS119227 (R.B.)
NIH K01 AG068366 (F.K.)

Title: Automated sleep scoring via machine learning for high-throughput analysis of mouse sleep/wake behavior

Authors: E. L. HODGES[1], T. J. SPRATT[1], C. M. DENNEHY[2], E. RODRIGUEZ-DIAZ[3], S. K. SINGH[3], D. D. AGUILAR[1], C. YANG[1], F. KATSUKI[1], J. M. MCNALLY[1], R. BASHEER[1], D. S. UYGUN[1];
[1]VA Boston Healthcare Syst. - Harvard Med. Sch., West Roxbury, MA; [2]Dept. of Neurosci., Stonehill Col., Easton, MA; [3]VA Boston Healthcare Syst. - Boston Univ., Boston, MA

Abstract: Manually sleep scoring high-resolution electroencephalographic and electromyographic (EEG+EMG) recordings from rodents is tedious and labor-intensive. Automated, algorithmic, or machine-learning (ML)-based approaches offer an appealing solution to this problem but have not been widely adopted. Furthermore, currently used file formats such as .csv and .edf are slow and poorly suited for modern high-throughput analysis pipelines. These bottlenecks ultimately limit the feasibility of much-needed longitudinal and high-powered sleep studies. To address this, we used a column-based filetype (.parquet) to consolidate our research group's large volume of manually scored EEG+EMG records and developed a ML pipeline capable of accurately and quickly classifying epochs into Wake, Non-Rapid-Eye-Movement (NREM) sleep, REM, or Artifact. We used a preexisting time-series feature extraction library (TSFEL, Barandas et al. 2020) supplemented with custom-designed features to extract 100 spectral, temporal, and statistical metrics from each epoch. We then calculated these features for 32 files of 24-hour EEG+EMG recordings and used TensorFlow to train a recurrent neural network consisting of long-short-term-memory and convolutional layers. Here we demonstrate that using this approach, we reduced the time necessary to score an entire experimental cohort from weeks to hours while maintaining high scoring accuracy. To validate ML-generated sleep scores, we compared them to human-assigned scores under baseline (BL) and sleep-deprived (SD) conditions. EEG+EMG was recorded in wildtype C57BL/6 [WT], choline acetyltransferase-Cre [ChAT-Cre], and vesicular glutamate transporter 2-Cre [vGluT2-Cre] mice (n=4 each genotype). First, baseline was recorded for 24 hours. During a follow-up 24-hour recording, mice were sleep-deprived via gentle handling for 4 hours after the onset of the inactive period (2 hours after lights-on, ZT2-6). The overall distribution of behavioral states in our cohorts was: Wake: 56.4%, NREM: 37.1%, REM: 5.75%, Artifact: 0.7%. Using our automated approach, the time required to score a 24-hour file was reduced from >300 minutes to ~12 minutes. We report that machine-generated scores matched human scores in 90.4±1.1% of epochs (468,533/ 518,400 total 4-second epochs). State-specific F1 scores were: Wake: 0.93±0.01, NREM: 0.92±0.01, REM: 0.56±0.04. These data suggest that, with additional training data and refinement of REM features, our ML-based automated sleep scoring method can replace manual scoring in mouse studies. Future work will include developing a browser-based user interface to visualize and review ML-generated scores.

Disclosures: E.L. Hodges: None. **T.J. Spratt:** None. **C.M. Dennehy:** None. **E. Rodriguez-Diaz:** None. **S.K. Singh:** None. **D.D. Aguilar:** None. **C. Yang:** None. **F. Katsuki:** None. **J.M. McNally:** None. **R. Basheer:** None. **D.S. Uygun:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.03

Topic: F.07. Biological Rhythms and Sleep

Support: NRF-2019M3C1B8090805
2022R1A2C2005062
NRF-2022R1I1A4063209

Title: Optogenetic activation of mPFC neurons increases sleep in Sprague-Dawley rats

Authors: *Y. LEE[1,2], Y. LEE[1], S. HWANG[1], H. LEE[4,5], S. JUN[1,2,3];
[1]Dept. of Electronic and Electrical Engin., [2]Grad. Program in Smart Factory, [3]Dept. of Brain and Cognitive Sci., Ewha Womans Univ., Seoul, Korea, Republic of; [4]Dept. of Neurol., Ewha Med. Res. Inst., Seoul, Korea, Republic of; [5]4Department of Med. Sci., Ewha Womans Univ. Sch. of Med., Seoul, Korea, Republic of

Abstract: Optogenetic activation of mPFC neurons increases sleep in Sprague-Dawley rats
Youjin Lee[1,2], Yena Lee[1], Seoyoung Hwang[1], Hyang Woon Lee[3,4], Sang Beom Jun[1,2,5*]
[1]Department of Electronic and Electrical Engineering, Ewha Womans University, Korea [2]Graduate Program in Smart Factory, Ewha Womans University, Korea[3]Department of Neurology, Ewha Medical Research Institute, Korea [4]Department of Medical Science, Ewha Womans University School of Medicine, Korea[5]Department of Brain and Cognitive Sciences, Ewha Womans University, Korea
*Abstract:*Sleep is essential not only for physical and mental recovery, but also for controlling physical functions such as growth and preservation of energy metabolism, or strengthening cognitive function and long-term memory. Lack of sleep can increase the risk of metabolic disorders, cardiovascular disease, and Alzheimer's disease. Therefore, there have been several attempts to improve the quality of sleep through brain stimulation. Medial prefrontal cortex (mPFC), thalamus reticular nucleus (TRN), and hippocampus are mainly studied as sleep-related brain regions, but the exact neural mechanisms for sleep and awake control have not yet been clearly identified. In this study, optogenetics was used to figure out sleep-awake mechanisms by selectively stimulating specific neural cells and circuits. As a target brain area of optogenetic stimulation, mPFC was chosen, in that mPFC forms a cortical-thalamic network, a major neural circuit involved in sleep-awake control, by oscillating neural signals with TRN and thalamus. AAV-CaMKIIα-hChR2(H134R)-EYFP was injected into the mPFC region of Sprague-Dawley rat for channel rhodopsin-2 (ChR2) expression. For optpogenetic stimulation, a 473 nm laser was irradiated with an intensity of 4.7 mW/mm^2. ChR2-expressed neurons in mPFC were activated by light at two different frequencies: 2 or 10 Hz (50 % duty cycle), and changes in sleep-awake proportion were compared. Optogenetic activation of mPFC increased overall sleep quantity. Especially, in terms of sleep-awake proportion, more changes were observed at 10 Hz than 2 Hz stimulation. The results are expected to be useful to find an effective neuromodulation approach for improving the quality of sleep.
Acknowledgment
This research was supported by Convergent Technology R&D Program for Human

Augmentation through the National Research Foundation of Korea (NRF) funded by the Ministry of Science and ICT (NRF-2019M3C1B8090805), the Korea government. (MSIT) (2022R1A2C2005062), the Ministry of Education(NRF-2022R1I1A4063209) and BK21-plus program of Ewha Womans University.

Disclosures: **Y. Lee:** None. **Y. Lee:** None. **S. Hwang:** None. **H. Lee:** None. **S. Jun:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.04

Topic: F.07. Biological Rhythms and Sleep

Support: JSPS KAKENHI 18J21517

Title: Sik3 regulates circadian rhythms and sleep need through distinct neuron types and brain regions.

Authors: *F. ASANO[1], T. FUJIYAMA[1], C. MIYOSHI[1], T. KITAZONO[1], S. J. KIM[1], K. IWASAKI[1], N. HOTTA-HIRASHIMA[1], M. KAKIZAKI[1], A. IKKYU[1], S. KANNO[1], S. NAKATA[1], N. ASAMA[1], T. TSUKAMOTO[1], A. HIRANO[1,2], S. MIZUNO[3], F. SUGIYAMA[3], S. TAKAHASHI[3], H. FUNATO[1,4], M. YANAGISAWA[1,5,6];
[1]Intl. Inst. for Integrative Sleep Med. (WPI-IIIS), Univ. of Tsukuba, Tsukuba, Japan; [2]Fac. of Med., Univ. of Tsukuba, Tsukuba, Japan; [3]Transborder Med. Res. Center, Univ. of Tsukuba, Tsukuba, Japan; [4]Dept. of Anat., Grad. Sch. of Medicine, Toho Univ., Ota-ku, Japan; [5]Dept. of Mol. Genet., Univ. of Texas Southwestern Med. Ctr., Dallas, TX; [6]Life Sci. Center, Tsukuba Advanced Res. Alliance, Univ. of Tsukuba, Tsukuba, Japan

Abstract: Sleep is a fundamental behavior conserved from vertebrates to invertebrates. Sleep/wakefulness is under the control of “two-processes”: sleep need and circadian rhythm. *Salt-inducible kinase 3* (*Sik3*) is a member of AMP-activated protein kinase family, implicated in both sleep need and circadian rhythm regulation. The gain-of-function mutation in *Sik3* gene, named *Sleepy*, increases NREM sleep amount and delta power during NREM sleep, which is a well-accepted index of sleep need. However, the role of endogenous SIK3 in sleep/wakefulness remains unknown. Almost all the systemic *Sik3* null mice died neonatally due to multiple abnormalities, including airway malformation, and survived few *Sik3* null mice exhibited severe growth retardation. Here, we generated *Sik3-ex3-flox* mice to induce conditional *Sik3*-deficiency. *Sik3-ex3-flox* mice were crossed with various types of neuronal Cre driver mice and assessed sleep/wakefulness and circadian rhythms of established neuronal *Sik3*-deficient mice. SIK3-deficiency in GABAergic neurons did not change NREM sleep time or delta power during NREM sleep but induced a longer circadian period. Crossing with other Cre driver mice revealed that SIK3 deficiency specifically in the suprachiasmatic nucleus resulted in a longer circadian period. We also found that SIK3 deficiency in glutamatergic neurons in the forebrain reduced

NREM sleep amount and delta power during NREM sleep. These results indicate that SIK3 regulates sleep need and circadian rhythms through distinct types of neurons and different brain regions.

Disclosures: **F. Asano:** None. **T. Fujiyama:** None. **C. Miyoshi:** None. **T. Kitazono:** None. **S.J. Kim:** None. **K. Iwasaki:** None. **N. Hotta-Hirashima:** None. **M. Kakizaki:** None. **A. Ikkyu:** None. **S. Kanno:** None. **S. Nakata:** None. **N. Asama:** None. **T. Tsukamoto:** None. **A. Hirano:** None. **S. Mizuno:** None. **F. Sugiyama:** None. **S. Takahashi:** None. **H. Funato:** None. **M. Yanagisawa:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.05

Topic: F.07. Biological Rhythms and Sleep

Support: Alfred P. Sloan Foundation, Frances & Kenneth Eisenberg Translational Research Award, NARSAD Young Investigator Grant # 27297

Title: Chronic stress-related alterations of dopaminergic signaling during sleep/wake states

Authors: *S. WANG[1], C. MARKUNAS[2], A. EBAN-ROTHSCHILD[3];
[1]Univ. of Michigan Psychology Grad. Program, Ann Arbor, MI; [2]Psychology, Univ. of Michigan, Ann Arbor, MI; [3]Univ. of Michigan, Ann Arbor, Ann Arbor, MI

Abstract: Proper sleep is essential for well-being yet is disrupted in many neuropsychiatric conditions, including major depression-a debilitating psychiatric disease among the leading causes of disability worldwide. Sleep disturbances are highly prevalent in individuals suffering from depression, and it is currently understood that the strong association between these stem from a common mechanism. Nonetheless, the neuronal underpinnings linking sleep and depression are not fully understood. Dysregulation of the ventral tegmental area (VTA) dopamine signaling system may underlie the association between sleep disturbances and chronic stress-a major risk factor for depression. VTA dopamine (DA) neurons have a central role in motivational processes as well as in sleep/wake regulation. Chronic stress provokes alterations in VTA-DA neurons activity during wakefulness; yet, whether it also results in alterations to VTA-DA activity during sleep is undetermined. We hypothesized that chronic stress induces alterations in VTA-DA signaling during sleep, resulting in sleep disturbances and exacerbating depression-like symptoms. To test our hypothesis, we used in-vivo calcium-dependent fiber photometry recordings from VTA-DA neurons in tandem with EEG-EMG and video recordings in male mice exposed to a 10-day long chronic social defeat stress (CSDS) paradigm. We subjected mice to ethologically relevant behavioral tests (measuring motivation to engage in social interactions, obtain food rewards and potential mates) to quantify depression-related symptoms. EEG-EMG, neuronal activity and behavioral data were collected prior to and

following exposure to the CSDS paradigm. As expected, a subset of mice were susceptible to the CSDS paradigm and displayed depression-related behaviors, while the other subset were resilient to the paradigm and behaved similarly to control mice. Susceptible mice, compared to both control and resilient mice, had fragmented NREM sleep, less EEG slow-wave activity during the dark phase, and altered VTA-DA population transients during both wakefulness and NREM sleep following CSDS. Susceptible mice also displayed altered population activity around wake-to-NREM sleep transitions. Lastly, CSDS exposure resulted in arousal-state specific alterations to DA release (via recordings of the florescent dopamine biosensor dLight1.1) at the nucleus accumbens and basolateral amygdala. Taken together, our findings provide a mechanistic framework for the participation of DA signaling in sleep disturbances and other depression-like symptoms following chronic stress.

Disclosures: S. Wang: None. **C. Markunas:** None. **A. Eban-Rothschild:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.06

Topic: F.07. Biological Rhythms and Sleep

Support: NIH Grant R01NS106032

Title: An oxytocin agonist increases cataplexy in orexin knockout mice

Authors: A. A. JOYAL[1,2], C. E. MAHONEY[1,2], T. E. SCAMMELL[1,2];
[1]Neurol., Beth Israel Deaconess Med. Ctr., Boston, MA; [2]Div. of Sleep Med., Harvard Med. Sch., Boston, MA

Abstract: Narcolepsy is a sleep disorder caused by the selective death of the orexin neurons. The loss of the orexin neurons destabilizes sleep/wake states and produces mixtures of states, including cataplexy, episodes of muscle paralysis during wake that is usually triggered by strong emotions. Patients with narcolepsy type 1 report that cataplexy most often occurs in positive social settings, such as laughing with friends or an exciting event. As oxytocin is essential for interpersonal interactions, we hypothesized that an oxytocin agonist would increase cataplexy in narcoleptic mice. We measured sleep/wake states and cataplexy in orexin knockout mice after injection of saline or the oxytocin receptor agonist, carbetocin. From 3 to 6 hours after lights off, carbetocin (4.2mg/kg, ip) nearly tripled the amount of cataplexy. We then blocked this increase with coadministration of the oxytocin receptor antagonist, L-368,899 (5mg/kg, ip). After coadministration of both agonist and antagonist, cataplexy returned to baseline levels. These results demonstrate that oxytocin signaling can increase cataplexy in mice, suggesting that oxytocin antagonism may be a viable therapeutic mechanism for narcolepsy.

Disclosures: A.A. Joyal: None. **C.E. Mahoney:** None. **T.E. Scammell:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.07

Topic: F.07. Biological Rhythms and Sleep

Support: NIH Grant R01 NS098813

Title: Narcolepsy symptomatology is dependent on the magnitude of Hypocretin/orexin neuron loss in Orexin-tTA; TetO-DTA mice

Authors: *Y. SUN[1], M. D. SCHWARTZ[1], J. HEU[1], R. TISDALE[1], S.-C. MA[1], M. HAIRE[1], H. COURTNEY[1], S. MORAIRTY[1], A. YAMANAKA[2], T. S. KILDUFF[1];
[1]SRI Intl., Menlo Park, CA; [2]Res. Inst. of Envrn. Medicine, Nagoya Univ., Res. Inst. of Envrn. Medicine, Nagoya Univ., Nagoya, Japan

Abstract: *Post mortem* data from humans with narcolepsy suggests that variable loss of hypocretin/orexin (Hcrt) neurons may underlie the differences in symptom severity seen in narcolepsy Type1 (NT1) and Type 2 (NT2). In the present study, we utilized *orexin-tTA;TetO-DTA* (Ox-DTA) mice, a conditional model of narcolepsy that allows ablation of Hcrt neurons through removal of doxycycline (DOX) from their diet, to induce variable amounts of Hcrt neuron loss depending on the duration of DOX removal. Furthermore, restoration of dietary DOX after a DOX(-) period (Re-DOX(+)) has previously been shown to arrest further Hcrt neuron degeneration. Six treatment groups were used in this study: **Group 1** (control): 7 weeks DOX(+) to suppress transgene expression and maintain an intact Hcrt neuron population; **Group 2**: 4 days DOX(-) to induce partial Hcrt neuron degeneration; **Group 3**: 4 days DOX(-) then Re-DOX(+) for 7 weeks; **Group 4**: 7 days DOX(-); **Group 5:** 7 days DOX(-) then Re-DOX(+) for 7 weeks; **Group 6**: 7 weeks DOX(-) to fully degenerate the Hcrt neurons. For each mouse, EEG/EMG recordings were conducted for 24-h prior to euthanasia, cardiac perfusion and brain extraction; the *in vivo* results presented below are based on analysis of the first 6-h of the dark phase (ZT12-18). *Post mortem* immunohistochemical analyses revealed 41±3.3% and 39±3.5% Hcrt neuron loss in Groups 2 and 3 compared to control Group 1. EEG/EMG recordings showed more bouts of both sleep and wake and shorter duration NREM bouts in Groups 2 and 3. Similar to the clinical manifestation of NT2, these indices of sleep/wake fragmentation occurred without cataplexy. Mice in Groups 4, 5 and 6 showed 58±4.1%, 70±3.6%, and 95±1.1% loss of Hcrt neurons, respectively, and exhibited cataplexy as well as sleep/wake fragmentation, mimicking the clinical symptoms of NT1. Moreover, there were significantly fewer cataplexy bouts in the partial degeneration Groups 4 (1±0.9, $p<0.001$) and 5 (5.3±1.8, $p<0.001$) than in the fully-degenerated Group 6 mice (18.4±5.1). Subcutaneous body temperature and activity also decreased as the Hcrt neurons degenerated. We conclude that the degree of sleep fragmentation, cataplexy severity and the decreased subcutaneous temperature and activity is directly related to the
magnitude of Hcrt neuron loss. Importantly, by manipulating the amount of Hcrt neuron loss in

Ox-DTA mice, we induced symptoms that resembled either NT1 or NT2 in humans.
Research supported by R01 NS098813 to TSK.

Disclosures: Y. Sun: None. **M.D. Schwartz:** None. **J. Heu:** None. **R. Tisdale:** None. **S. Ma:** None. **M. Haire:** None. **H. Courtney:** None. **S. Morairty:** None. **A. Yamanaka:** None. **T.S. Kilduff:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.08

Topic: F.07. Biological Rhythms and Sleep

Title: High-fat diet consumption alters circadian and behavioral responses to constant light

Authors: *A. T. MCFARLAND, M. E. BURNS, F. MEDEIROS CONTINI, S. M. SOARES, J. C. PRICE, M. J. LAMONTE, M. E. CUNNINGHAM, K. W. ADAMS, J. A. SEGGIO;
Biol. Sci., Bridgewater State Univ., Bridgewater, MA

Abstract: Previous work has illustrated a bidirectional relationship between circadian clock function and obesity. One here-to-stay form of circadian disruption is light-at-night or light pollution either through building and streetlights or through TVs and smart phone use at night. Light exposure during the night can produce negative behavioral and physiological effects in both diurnal and nocturnal animals. Lastly, high-fat diet consumption has been known to produce altered behavioral effects and modulate the circadian clock. This study investigated whether consumption of a high-fat diet can lead to altered behavioral, physiological, and circadian characteristics in male C57BL/6J mice. Mice were either given a high-fat diet or regular chow and then placed into either a standard light:dark cycle (LD) or constant light (LL). Their circadian rhythms, anxiety-like behaviors (via an open-field and light-dark box), and weight gain were observed. While all animals in LL exhibited period lengthening as expected, animals consuming a high-fat diet exhibited significantly reduced period lengthening compared to regular chow animals in LL. Additionally, LL increased novelty-induced activity and anxiety-like behaviors in regular chow mice, while high-fat diet mice exhibited no such changes, compared to their respective LD controls. High-fat diet also led to increased body mass and reduced activity in the open field regardless of lighting condition. These results indicate that diet and obesity can modulate behavioral responses to aberrant light exposure. This study also suggests that high-fat diet consumption and/or obesity can also regulate circadian responses to light.

Disclosures: A.T. McFarland: None. **M.E. Burns:** None. **F. Medeiros Contini:** None. **S.M. Soares:** None. **J.C. Price:** None. **M.J. Lamonte:** None. **M.E. Cunningham:** None. **K.W. Adams:** None. **J.A. Seggio:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.09

Topic: F.07. Biological Rhythms and Sleep

Support: Korea, MOTIE 20008980

Title: The extract of chrysanthemum morifolium and linarin enhanced the sleep duration in rodent models via Cl^- channel activation

Authors: M. KIM, *S. OH;
Ewha Womans Univ, Sch. of Med., Ewha Womans Univ, Sch. of Med., Seoul, Korea, Republic of

Abstract: Interest in sleep-promoting compounds derived from natural compounds is increasing owing to various side effects of prescription drugs for sleep disorders such as benzodiazepines. Dried Chrysanthemum morifolium flowers have traditionally been used in Korea for the treatment of insomnia. Thus, in this study, the sleep-promoting activity of *Chrysanthemum morifolium* extract and its active substance Linarin was assessed by measuring sleep latency time and duration in a pentobarbital-induced sleep mice model. Additionally, the effects of orally administered *Chrysanthemum morifolium* extract on rapid-eye-movement (REM) and non-REM sleep were analyzed by electroencephalography (EEG) in rats. In a dose-dependent manner, *Chrysanthemum morifolium* extract and Linarin showed longer sleep duration in the pentobarbital-induced sleep test compared to administered with pentobarbital alone groups at both hypnotic and subhypnotic doses. The low and high doses of *Chrysanthemum morifolium* extract administration significantly increased sleep quality, especially the relative power of low-frequency (delta) waves, compared with the normal group. Linarin and muscimol both increased chloride (Cl^-) uptake in the SH-SY5Y human cell line and the chloride influx was reduced by bicuculline. After administration of *Chrysanthemum morifolium* extract, the hippocampus, frontal cortex, and hypothalamus from rodents were collected and blotted for GAD (glutamic acid decarboxylase) and $GABA_A$ receptors subunit expression levels. The expression of GAD, α1- and β2-subunits of the $GABA_A$ receptor was modulated in the mouse brain. In conclusion, *Chrysanthemum morifolium* extract augments pentobarbital-induced sleep duration and enhances the sleep quality in EEG waves. These effects might be due to the activation of Cl^- channel.

Disclosures: M. Kim: None. **S. Oh:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.10

Topic: F.07. Biological Rhythms and Sleep

Support: NIH Grant 5P20GM103642
NSF Grant 1736026
NIH Grant 2R25NS080687
5R25GM061151-19
NIH Grant P20GM103475
NSF Grant 1633184
NSF Grant 2131647

Title: Gut microbes affect the onset of circadian rhythms and regulate gene expression in honey bee (Apis mellifera)

Authors: *Y. B. KORU[1], E. C. COURTNEY[2], E. AVILES-RIOS[1], Y. ORTIZ-ALVARADO[1], M. A. DOKE[1], A. MONTES-MERCADO[1], A. A. RUGGIERI[1], N. RODRIGUEZ[1], R. GIORDANO[3], R. K. DONTHU[4], J. LEON[1], A. GHEZZI[1], T. GIRAY[1], J. AGOSTO-RIVERA[1]; [1]Univ. of Puerto Rico, San Juan, PR; [2]Minerva Univ., San Francisco, CA; [3]Florida Intl. Univ., Miami, FL; [4]Know Your Bee, San Juan, PR

Abstract: Circadian rhythms are vital to the survival and behavior of honey bees, regulating key processes such as the age-dependent division of labor within colonies. Nurse bees in a colony, which take care of brood, lack circadian rhythms, whereas forager bees, which go outside to collect nectar, pollen, and water, have strict rhythmicity that allows them to predict optimal foraging times. In honey bees, neurological processes such as learning and memory are mediated by genetic, neuroplastic, and environmental factors, with recent studies focusing on the connection between gut bacteria and neural circuits. However, the effects of gut microbes on the development and regulation of circadian rhythm in honey bees has been understudied. We hypothesize that altering honey bee gut microbiota will influence ontogeny of circadian rhythm, and aim to elucidate this relationship using antibiotic treatment. Locomotor activity monitor (LAM) was used to measure rhythmicity in laboratory conditions, and transcription analysis of brain tissue was performed using deseq and limma to understand differential expression of important neurotrophic factors under antibiotic treatment. We find that antibiotic treatment results in the delay of the development of circadian rhythm, and identify 5 differentially expressed genes: Insulin growth factor-binding protein complex acid labile subunit (IGFALS), cys-loop liganda gated ion channel subunit, vegetative cell wall protein gp1, zinc finger CCCH domain-contaning protein and CUGBP Elav-like. Further research on the role of these genes could allow for an understanding of which specific signals provide a molecular mechanism for how gut bacteria influences the ontogeny of circadian rhythm.

Disclosures: Y.B. Koru: None. **E.C. Courtney:** None. **E. Aviles-Rios:** None. **Y. Ortiz-Alvarado:** None. **M.A. Doke:** None. **A. Montes-Mercado:** None. **A.A. Ruggieri:** None. **N. Rodriguez:** None. **R. Giordano:** None. **R.K. Donthu:** None. **J. Leon:** None. **A. Ghezzi:** None. **T. Giray:** None. **J. Agosto-Rivera:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.11

Topic: F.07. Biological Rhythms and Sleep

Support: Hertfordshire Knowledge Exchange Programme
Hertfordshire Local Enterprise Partnership
European Regional Development Fund

Title: EEG gamma power enables robust automated sleep-staging in rodents

Authors: *J. R. HUXTER[1], S. KANTOR[1], M. LANIGAN[1,2], L. GIGGINS[1], L. SILENIEKS[3], M. DUXON[1];
[1]Transpharmation Ltd., London, United Kingdom; [2]Sch. of Life and Med. Sci., Univ. of Hertfordshire, Hatfield, United Kingdom; [3]InterVivo Solutions Inc, Toronto, ON, Canada

Abstract: Sleep disturbances are common in neuropsychiatric and neurodegenerative diseases. Consequently, changes in sleep architecture and changes in EEG spectral power specific to different sleep-stages are valuable tools in translational preclinical research. Historically, algorithms for rodent sleep-stage classification rely on changes in muscle-tone and EEG power in the delta (1-4 Hz) and theta (4-10 Hz) bands. However, these outputs typically require refinement by human visual scoring, making the process laborious and prone to human error. We aimed to identify a simple but robust sleep-stage classification system which better correlates with the results of human visual scoring and makes manual curation redundant. We recorded fronto-parietal EEG, EMG (nuchal muscle) and locomotor activity from male Sprague Dawley rats (495-586g, N=7) for 22 hours, starting at 2.5 hours after lights-on. Ten-second epochs were visually scored by an expert as either Wake, REM sleep or non-REM sleep. K-means clustering and regression tree analysis revealed, unexpectedly, that 30-100Hz gamma oscillations were powerful predictors of the visual classifications, and particularly effective for distinguishing REM sleep from non-REM sleep. Further, by adopting a 2-stage median-based automated scoring algorithm, we were able to replicate human visual scoring with >90% accuracy. Compared with delta/theta-based automatic scoring there was significantly improved accuracy in assigning sleep-stages to individual epochs (Paired $t(6)=132.9$, $p<.0001$), and a large increase from 30% to 98% accuracy for REM sleep in particular. Compared with visual scoring, the algorithm reduced processing time from hours to seconds, while removing inter-rater variability and potential human error.

Disclosures: **J.R. Huxter:** A. Employment/Salary (full or part-time):; Transpharmation. **S. Kantor:** A. Employment/Salary (full or part-time):; Transpharmation Ltd. **M. Lanigan:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Transpharmation Ltd. **L. Giggins:** A. Employment/Salary (full or part-time):; Transpharmation Ltd. **L. Silenieks:** A. Employment/Salary (full or part-time):; InterVivo Solutions Inc. **M. Duxon:** A. Employment/Salary (full or part-time):; Transpharmation Ltd..

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.12

Topic: F.07. Biological Rhythms and Sleep

Support: NRF Grant 2018M3C7A1056293, Korea
NRF Grant 2022R1A2C1005532, Korea

Title: Enhanced resistance and recovery of stress from insufficient sleep by deficiency of cereblon (CRBN)

Authors: J.-H. JUNG, J. KIM, U. AKBER, J.-W. BAEK, T. KIM, *C.-S. PARK;
Gwangju Inst. of Sci. and Technol. (GIST), Gwangju Inst. of Sci. and Technol., Gwangju, Korea, Republic of

Abstract: Insufficient sleep or sleep deprivation (SD) is known to result in brain malfunctions, including neurological disorders and impairment of cognitive function. Cereblon (CRBN) is a multi-functional protein and a substrate receptor in $CRL4^{CRBN}$ E3 ligase complex. Thus, CRBN regulates the proteostasis of various target proteins via ubiquitination and subsequent degradation. In our recent study, we showed that CRBN regulates DNAJ protein, a co-chaperon, attenuating tau aggregation and AMP-activated protein kinase (AMPK) sensing energy states via ubiquitin-proteasome system. We wondered whether CRBN affects sleep behavior via targeting key proteins related to sleep regulation. To investigate the role of CRBN in sleep-wake behavior and SD, we initially restricted wild-type ($Crbn^{+/+}$) and CRBN knockout ($Crbn^{-/-}$) mice to sleep for 6 hours. We observed that the protein level of CRBN decreased after SD in $Crbn^{+/+}$ mice, while those of several neuropathological proteins and chaperones increased. However, the induction of neuropathological proteins by SD in $Crbn^{-/-}$ mice was much smaller and the levels were even less than in the basal state of $Crbn^{+/+}$ mice. We then recorded the electroencephalography of $Crbn^{+/+}$ and $Crbn^{-/-}$ mice during baseline, SD, and recovery sleep (RS). In baseline, 24-hours sleep-wake profiles did not differ between $Crbn^{+/+}$ and $Crbn^{-/-}$ mice in basal state, whereas $Crbn^{-/-}$ mice showed slightly increased non-rapid eye movement (NREM) continuity comparing to $Crbn^{+/+}$ mice. Consistent with the fact that AMPK is activated highly and constitutively in $Crbn^{-/-}$ mice during SD, a high NREM continuity and an increased low delta power (0.5~2.0Hz) were observed in RS in these mice. Taken together, our results show that the loss of CRBN in the brain contributes mice to get resistance to SD-induced stress.

Disclosures: J. Jung: None. **J. Kim:** None. **U. Akber:** None. **J. Baek:** None. **T. Kim:** None. **C. Park:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.13

Topic: F.07. Biological Rhythms and Sleep

Support: Natural Sciences and Engineering Research Council of Canada (NSERC RGPIN-2021-03909, B. A. K.; Undergraduate Student Research Award, S. S.)
Canada Research Chair (B. A. K.)
Canadian Foundation for Innovation (CFI-JELF 41428, B. A. K.)

Title: The effects of sleep fragmentation on trial-unique non-matching to location (TUNL) performance in mice

Authors: *S. SAW, T. YILDIRIM, R. MISTLBERGER, B. KENT;
Psychology, Simon Fraser Univ., Burnaby, BC, Canada

Abstract: Pattern separation is a mechanism by which the hippocampus keeps similar spatial memories distinct. Sleep and circadian rhythm disruption can impair hippocampus-dependent learning and memory in rodents, and these procedures markedly suppress adult hippocampal neurogenesis, a form of brain plasticity that is important for pattern separation. In this study, we are examining how sleep disruption and circadian rhythm shifts affect performance on the trial-unique non-matching to location (TUNL) task, which is a touchscreen-based cognitive test designed to assess pattern separation. Touchscreen-based cognitive tests in rodents are semi-automated and more standardized, less stressful, and have a higher translational potential to human clinical research than traditional behavioural tests for rodents. In the TUNL task, mice must remember the spatial location of a square stimulus on a screen, then choose that stimulus at a new location when both the old and new locations are presented. We trained mice (c57BL/6; n= 22; 12F) on the TUNL task at a fixed time each day during the first 6 h of the dark period. We evaluated 1) the effect of daily training on timing of the circadian rest-activity cycle, and 2) how performance was affected by a. test time, b. a 6-hour phase delay of the light-dark (LD) cycle, and c. seven days of sleep fragmentation. Hippocampal neurogenesis following sleep fragmentation was measured using BrdU immunolabeling. Data collection and analysis are ongoing. Preliminary results indicate that the activity rhythms of the mice are modified by the training and feeding schedules, with daily activity rising and peaking just prior to the daily feeding time (ZT19) each night. There is also evidence that the daily onset of nocturnal activity is shifted by the TUNL testing time, suggesting that the reward-based cognitive testing can alter the phase angle of entrainment to the LD cycle. For the LD phase delay of 6 hours, we hypothesize that this shift will be associated with decreased accuracy on the TUNL task. We also hypothesize that the seven days of sleep fragmentation will be associated with reduced accuracy on the TUNL task and lower levels of hippocampal neurogenesis in comparison to mice that had no sleep disruptions. These findings will inform future research evaluating whether tasks assessing pattern separation are sensitive to sleep and circadian disruption.

Disclosures: S. Saw: None. **T. Yildirim:** None. **R. Mistlberger:** None. **B. Kent:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.14

Topic: F.07. Biological Rhythms and Sleep

Support: Programa UNAM-DGAPA-PAPIIT IN231620.

Title: Sleep and sleep deprivation in crayfish: study of brain and cardiorespiratory electrical activity

Authors: *M. OSORIO-PALACIOS, L. MONTIEL-TREJO, I. OLIVER-DOMÍNGUEZ, R. AGUAYO-SOLÍS, J. HERNÁNDEZ-FALCÓN, K. MENDOZA-ÁNGELES;
Univ. Nacional Autónoma de México, Ciudad de México, Mexico

Abstract: In vertebrate and invertebrate animals, sleep is essential for the maintenance of life itself. For example, in complex organisms such as human beings, sleep deprivation elicits changes in the electroencephalogram structure and dysregulation of cardiorespiratory activity. In rats, after two to three weeks of sleep deprivation, animals lose weight despite a great increase in food intake, if deprivation continues, they finally die. In invertebrates as crayfish, 24 hours of sleep deprivation are enough to cause death. Previous results show that 1 hour of sleep deprivation modifies the dynamic of brain and cardiorespiratory electrical activity in crayfish. Here, the main goal was to analyze physiological time series like EEG, EKG and respiratory electrical activity of adult crayfish *Procambarus clarkii* by time-frequency domain analysis in order to assess the effect of sleep deprivation. We used wavelet transform and the first four statistical moments of the distribution to analyze brain and cardiorespiratory electrical activity, and recorded during 8 continuous hours in two different conditions: 1) control and 2) after one hour of sleep deprivation. Our results show that: (1) brain electrical activity from control sleeping animals showed a decrease in power at 30 Hz. Sleep deprived animals presented lower power in all EEG frequencies even when they were allowed to sleep, (2) regarding cardiorespiratory activity, as deprivation of sleep evolves, skewness with respect to the median tends to zero (increased symmetry), standard deviation decreases (higher rigidity), kurtosis also vanishes (data becomes more concentrated around the mean), and the distribution becomes more Gaussian. In conclusion, the analysis of brain and cardiorespiratory electrical activity in the time and frequency domain shows that power and all moments of the distribution are relevant parameters that allow significant differentiations between both conditions, control and after one hour sleep deprivation.

Disclosures: M. Osorio-Palacios: None. **L. Montiel-Trejo:** None. **I. Oliver-Domínguez:** None. **R. Aguayo-Solís:** None. **J. Hernández-Falcón:** None. **K. Mendoza-Ángeles:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.15

Topic: F.07. Biological Rhythms and Sleep

Support: KAKENHI 17H06095
KAKENHI 22H04918

Title: Involvement of microglia in sleep/wake regulation in baseline conditions and under acute social defeat stress in mice

Authors: *K. MIYANISHI[1], H. FUNATO[1,2], M. YANAGISAWA[1,3,4];
[1]Yanagisawa / Funato Lab., IIIS / Univ. of Tsukuba, Tsukuba, Japan; [2]Dept. of Anat., Fac. of Medicine, Toho Univ., Tokyo, Japan; [3]Dept. of Mol. Genet., Univ. of Texas Southwestern Med. Ctr., Dallas, TX; [4]Life Sci. Ctr., Tsukuba Advanced Res. Alliance, Univ. of Tsukuba, Tsukuba, Japan

Abstract: Microglia, tissue-resident macrophages in the central nervous system (CNS), account for 10-15% of all cells in the adult brain. They play crucial roles not only in immune challenge, such as neurodegenerative and neuroinflammatory diseases, but also in healthy conditions of the CNS. We previously reported circadian changes in the morphology and function of microglia. In the prefrontal cortex, microglia displayed hyper-ramified morphology and increased phagocytosis of synapses during sleep. These results suggest that the diurnal variations in microglial activity may contribute to sleep/wakefulness. However, whether and how microglia regulate sleep/wake behavior remains to be elucidated. Here, to examine the hypothesis that microglia regulate sleep/wake behavior, we employed CNS-penetrant selective colony-stimulating factor-1 receptor inhibitor, PLX5622 (PLX), which eliminates microglia in mice. EEG/EMG recordings were performed after mice were fed with the PLX-formulated chow for 14 days. We found that oral administration of PLX decreased wake amount and increased NREM sleep amount during the dark phase in baseline conditions. To further investigate the role of microglia in sleep/wakefulness under acute social defeat stress (ASDS), which triggers microglial activation as previously reported, we examined whether ASDS alters the effect of microglial depletion with PLX on sleep/wake behavior. Following the previous studies, C57BL/6N mice were briefly encountered with highly aggressive ICR mice 4 times during 1-hour ASDS session to avoid severe physical injury of defeated mice. We exposed mice with or without PLX-treatment to ASDS session during ZT11-12 and then performed EEG/EMG recordings. We found that PLX-treatment further exaggerated an ASDS-induced reduction in wake amount, suggesting that microglia may play a potentially protective role against ASDS. Taken together, our study indicated the possibility that microglia are involved not only in the regulation of sleep/wake behavior in baseline conditions especially the dark phase, but also in changes in sleep architecture in response to mental stress.

Disclosures: K. Miyanishi: None. **H. Funato:** None. **M. Yanagisawa:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.16

Topic: F.07. Biological Rhythms and Sleep

Support: NIH T32

Title: Identifying the role of sleep deprivation in anti-tumor immunity

Authors: *N. FRANCIS, J. BORNIGER;
Cold Spring Harbor Lab., cold spring harbor, NY

Abstract: Patients and survivors of breast cancer frequently experience debilitating sleep problems that are associated with reduced quality of life and increased mortality. Chronic disruption of the sleep/wake cycle promotes both spontaneous breast cancer development and accelerates tumor progression and metastatic spread in animal models. Despite the prevalence of disrupted sleep in cancer patient populations, a gap in knowledge remains regarding the molecular mechanisms of sleep disruption-induced disease progression. Some evidence suggests that sleep/wake states are fundamentally coupled to the immune system, and disruptions in arousal states can alter the immune composition of the tumor microenvironment. We hypothesized that sleep deprivation altered breast cancer progression and anti-tumor immunity. We predicted to observe alterations in immune cells within the tumor microenvironment and to progress breast cancer. In the current study, tumor-bearing breast cancer and healthy mice underwent chronic total sleep deprivation (6-8 hrs per day for 3-5 wks). Mice were assessed for changes in plasma cytokine levels (ELISA proteome array) and changes in immune populations within the tumor and other tissues (flow cytometry). Preliminary results suggest that sleep deprivation increases the concentrations of circulating chemokines and cytokines that are associated with adverse outcomes (increased metastasis and increased mortality) in breast cancer. Additionally, levels of dendritic cells, total T cells, and various T-cell populations were altered when tumors and spleens from sleep- deprived tumor-bearing mice were assessed. Taken together, the altered chemokines/cytokines in the plasma and immune cell populations in the tumor and spleen appears to be associated with the senescence secretory associated phenotype (SASP). Therefore, sleep deprivation could be altering immune cell trafficking through chemokine signaling leading to adverse outcomes in breast cancer.

Disclosures: **N. Francis:** None. **J. Borniger:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.17

Topic: F.07. Biological Rhythms and Sleep

Title: Serotonergic psychedelic 5-MeO-DMT induces sleep-like cortical activity during wakefulness in mice

Authors: *B. BREANT[1], J. PRIUS-MENGUAL[1], T. SHARP[2], D. M. BANNERMAN[3], V. V. VYAZOVSKIY[1];
[1]Dept. of Physiology, Anat. and Genet., [2]Dept. of Pharmacol., [3]Dept. of Exptl. Psychology, Univ. of Oxford, Oxford, United Kingdom

Abstract: *Introduction*: The traditional view that the serotonergic system plays an important role in subcortical control of global sleep-wake states is supported by observations that administration of serotonergic psychedelics suppresses rapid eye movement (REM) sleep and results in increased sleep fragmentation. However, the possibility that potentiating the serotonergic system through psychedelics results in an occurrence of altered states of vigilance has received less attention. We hypothesise that the serotonergic system plays a role in controlling the quality rather than the quantity of specific sleep-wake states thus producing a dissociated state of vigilance. The aim of this study is to characterise the effects of a short-lasting psychedelic compound, 5-methoxy-N,N-dimethyltryptamine (5-MeO-DMT), on brain activity and sleep-wake states in laboratory mice.
Methods: We performed chronic frontal and occipital EEG and EMG recordings in freely behaving laboratory mice (C57BL6, n = 15). 4-hour sleep deprivation was performed on a subset of these animals (n = 8). In a subset of animals, local field potentials (n = 4) and multiunit activity (n = 2) were recorded from the visual cortex. Each animal received an IP injection of 5-methoxy-N,N-dimethyltryptamine (5 MeO-DMT, 5 mg/kg in a 1mg/mL saline solution) at either the beginning of the light period or after sleep deprivation. Vigilance states were manually scored in 4s epochs using SleepSign, and electrophysiology data was analysed with Matlab.
Results: We observed that after 5-MeO-DMT administration, cortical activity in awake, moving animals was characterised by occurrence of neuronal population OFF-periods, resembling NREM sleep. These observations were supported by EEG spectral analysis showing a reduction in theta-frequency power (interindividual range: - 3.82 % to - 73.47 %) and an increased spectral power in slow frequency range (9.11 % to 48.60 %) in the first 30 minutes following the injection. The effects were short-lasting and largely dissipated 1 hour after the injection. The latency to REM sleep was delayed (4.53 minutes to 84.267 minutes).
Conclusion: Our data support the notion that the effects of 5-MeO-DMT are short-lasting but significantly impact sleep-wake cycle. The main effect on vigilance states consisted in an acute suppression of REM sleep. Reduced theta activity and increased slow wave activity during waking suggest that 5-MeO-DMT induces a dissociated state of vigilance, having features of both wakefulness and sleep. This project was supported by a BBSRC Scholarship. The Compound was provided by Beckley Psytech.

Disclosures: B. Breant: None. **J. Prius-Mengual:** None. **T. Sharp:** None. **D.M. Bannerman:** None. **V.V. Vyazovskiy:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.18

Topic: F.07. Biological Rhythms and Sleep

Support: College of Science, CSU East Bay
NIH 7R15GM125073-02
NSF IOS 2042873

Title: Distinct neuromodulatory input pathways to mushroom body regulate sleep in Drosophila

Authors: M. REYES[1], S. BUCHERT[2], J. LOW[2], A. NGUYEN[3], *D. SITARAMAN[1];
[1]California State Univ., California State Univ. East Bay, hayward, CA; [2]California State Univ., Hayward, CA; [3]Univ. of San Diego, San Diego, CA

Abstract: A key feature of sleep-wake regulation is the ability to rapidly transition from one state to the other and persist in that state. One current model in mammals relies on mutually inhibitory interactions between sleep and wake-promoting neuronal populations that implement a bistable flip-flop circuit. Each half of this circuit (sleep- and wake-promoting neurons) strongly inhibits the other via interneurons thereby creating a self-reinforcing behavioral switch preventing intermediate states. Once the wake microcircuit is activated it remains unclear how these neurons persist in an active state to support extended periods of arousal. Evidence from multiple animal models shows that the arousal phase is associated with concomitant and persistent activity of specific neuromodulators primarily dopamine and noerpinephrine but it is not clear if their function is permissive to promote wakefulness, sleep homeostasis or state transitions.We recently identified two distinct synaptic microcircuits within mushroom body in Drosophila where sleep-promoting KCs increase sleep by preferentially activating a class of cholinergic ONs and wake-promoting KCs decrease sleep by preferentially activating a class of glutamatergic ONs. In addition to the core sleep and wake synaptic microcircuits within the MB we have also identified 5 classes of dopaminergic neurons (DANs) and octopaminergic neurons (VPMs) that project to the MB and regulate sleep. Using genetic, physiological, and behavioral approaches we find that the molecular signaling and circuit connectivity within these sleep networks regulates both persistence of wakefulness and sleep need. We will present these results and discuss potential mechanisms of sleep regulation by aminergic inputs to the mushroom body

Disclosures: **M. Reyes:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Center for Student Research, LSAMP. **S. Buchert:** None. **J. Low:** None. **A. Nguyen:** None. **D. Sitaraman:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.19

Topic: F.07. Biological Rhythms and Sleep

Support: PRONACES-CONACYT grant 194171
VIEP-BUAP 2021-2022 to CA in Neuroendocrinología (BUAP-CA-288)
CONACYT No. 926368

Title: Alcohol effects on anxiety and sleep-wake cycle in a rat model of anxiety

Authors: *A. FIERRO-ROJAS[1], C. CORTÉS[1], G. GARCÍA-AGUILAR[2], J. R. EGUIBAR[3]; [1]Behavioral Neurophysiol. and Motor Control laboratory, Benemérita Univ. Autónoma De Puebla, Puebla, Mexico; [2]Fac. of Psychology, Benemérita Univ. Autónoma de Puebla, Puebla, Mexico; [3]Intl. Office, Benemerita Univ. Autonoma De Puebla, Puebla, Pue., Mexico

Abstract: Anxiety is known to produce aversive symptoms like sleep disruption and fear of novel stimuli when exposed to new environments. Subjects with anxiety tend to increase their alcohol intake to diminish these aversive symptoms with respect to subjects with lower anxiety responses. We have selectively bred two sublines from Sprague-Dawley rats that differs in their spontaneous yawning frequency. The high-yawning (HY) rats have a mean of 20 yawns/h whereas low-yawning (LY) rats have a mean of just 2 yawns/h. Additionally, LY rats have shown anxious behaviors when evaluated in different standard psychophysiological tests. The aim of this study was to assess the effect of alcohol intake on anxious behavior in the elevated plus maze and on the organization of sleep-wake cycle. We used eight male rats of each high-yawning (HY) and low-yawning (LY) sublines at 3 months of age. All subjects maintained in standard conditions with free access to purified water and food pellets. Subjects implanted for electroencephalographic (EEG), electromyographic (EMG) and electrooculographic (EOG) recordings to characterize sleep-wake stages. We obtained a basal sleep-wake recording followed by a second recording sleep-wake recording (AL1) after a period of 7-day alcohol habituation with a solution of 9.6% ethanol as the only source of hydration. A third sleep-wake (AL2) recording obtained after a 3-week period of alcohol preference by using a two-bottle choice paradigm with free access to ethanol solution and purified water. Additionally, anxiety was evaluated in the elevated plus maze (EPM) before and after alcohol exposure. LY rats had higher alcohol intake with respect to HY rats ($P<0.05$) and showed an increase of the number of crossings of the open arms in the EPM ($P<0.05$). Alcohol intake produced a general decrease on wake duration ($P<0.05$), and a concomitantly increase in the slow-wave sleep (SWS) and rapid eye movement (REM) sleep ($P<0.05$) in both sublines. However, there was an increase in the mean duration of SWS episodes only in LY rats ($P<0.05$), but a decrease of SWS and REM sleep in their resting phase. In conclusion, LY rat is a model of higher anxiety because they consumed more alcohol than HY rats and were more susceptible on anxious effects on behavior and on the sleep-wake cycle.

Disclosures: A. Fierro-Rojas: None. **C. Cortés:** None. **G. García-Aguilar:** None. **J.R. Eguibar:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.20

Topic: F.07. Biological Rhythms and Sleep

Title: Strain specific behavioral responses to constant light between male C57BL/6J andB6.Cg-Lep^{ob}/J mice

Authors: *F. MEDEIROS CONTINI, M. E. BURNS, M. J. LAMONTE, J. C. PRICE, S. M. SOARES, A. T. MCFARLAND, C. T. WARING, J. M. MICHAUD, J. A. SEGGIO;
Biol. Sci., Bridgewater State Univ., Bridgewater, MA

Abstract: Light-at-night exposure, either through smart-devices or shift-work, has been known to lead to obesogenic and abnormal behavioral outcomes. Previous work has also shown that leptin can modulate several neurobehavioral outcomes, as leptin KO mice exhibit altered behavioral responses to a wide variety of behavioral assays that measure anxiety-like behaviors, learning and memory, and depressive-like behaviors. This study investigated whether leptin KO mice exhibit worsened behavioral outcomes when placed into constant room-level lighting (LL) compared to animals with intact leptin signaling. Male C57BL6/J (B6) and B6.Cg-*Lep*^{ob}/J (OB) were placed into circadian rhythm monitoring home-cages and placed into either a 12:12 LD cycle (LD) or LL. Novelty-induced activity, anxiety-like behaviors, glucose tolerance, and blood triglycerides were measured. B6 mice exhibited increased period lengthening in LL, increased circadian robustness, and locomotor activity compared to OB mice. While B6 mice exhibited increased novelty-induced activity in an open-field in LL, OB mice did not, although no differences in anxiety-like behaviors were found in the elevated-plus maze. While LL increased body weight of animals, LL did not further exacerbate glucose levels or triglyceride levels in either mouse strain. Serum leptin levels were also elevated in B6 mice experiencing LL compared to B6 mice in LD. These results indicate that leptin may regulate circadian and behavioral responses to altered light exposure, even when the metabolic effects are somewhat muted.

Disclosures: F. Medeiros Contini: None. **M.E. Burns:** None. **M.J. Lamonte:** None. **J.C. Price:** None. **S.M. Soares:** None. **A.T. McFarland:** None. **C.T. Waring:** None. **J.M. Michaud:** None. **J.A. Seggio:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.21

Topic: F.07. Biological Rhythms and Sleep

Support: CIHR FDN-148478
NSERC RGPIN-2020-06717

Healthy Brains Healthy Lives
Alzheimer Canada
Canada Research Chair Tier 1

Title: Deep brain theta-band optogenetic stimulation of the septohippocampal fibres in fornix improves memory performance in an Alzheimer's disease mouse model

Authors: E. VICO VARELA, ***J. E. S. CARMICHAEL**, F. MANSEAU, G. ETTER, J. CHOI, S. WILLIAMS;
Psychiatry, McGill University, Douglas Res. Ctr., Montreal, QC, Canada

Abstract: Electrical stimulation of the fornix has been proposed as a therapeutic intervention to reverse memory loss in Alzheimer's disease (AD). The fornix is a white matter bundle containing axons (cholinergic, GABAergic, and glutamatergic fibres) connecting the hippocampus to the medial septum, the anterior thalamic nuclei, and the mammillary bodies. It is widely known that inactivating the fornix leads to memory impairments in rodents and humans, but recently acute or chronic activation of the fornix at the theta frequency (5-10Hz) has been shown to be a viable intervention to improve memory. To determine if fornix stimulation is sufficient to rescue memory impairments in a mouse model of Alzheimer's disease we applied focal 5Hz optogenetic stimulation to the septohippocampal fibres. Using the hAPP-J20 AD mouse model (4-6months old) we show that targeted 5Hz optical stimulation of the GABAergic septohippocampal fibres results in a significant increase in theta band power during both wake and REM sleep in the hippocampus. Continuous 4-hour 5Hz fornix stimulation between the encoding and recall phases of both novel-object-location and passive avoidance memory tasks rescued J20 performance to a level comparable with healthy controls. During REM sleep and to a lesser extent non-REM sleep, J20 mice displayed large-amplitude interictal spikes (IIS) lasting 6-9ms. Continuous 4-hour 5Hz optical fornix stimulation drastically reduced the occurrence of these IISs. We find that the decreased occurrence of IIS during REM sleep was correlated with subsequent memory performance suggesting a possible causal mechanism. Together these data demonstrate that focal theta-frequency stimulation of the fornix is effective at rescuing memory performance and reducing the occurrence of pathological IISs in the J20 AD mouse model.

Disclosures: E. Vico Varela: None. **J.E.S. Carmichael:** None. **F. Manseau:** None. **G. Etter:** None. **J. Choi:** None. **S. Williams:** None.

Poster

554. Autonomic Regulation of Sleep: Behavior II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 554.22

Topic: F.07. Biological Rhythms and Sleep

Support: CIHR FDN-148478
Canada Research Chair
HBHL

Title: Replay of trajectory events during REM sleep across different contexts

Authors: *J. CHOI[1], J. CARMICHAEL[2], G. ETTER[2], S. WILLIAMS[2];
[1]Integrated Program in Neurosci., McGill University, Douglas Res. Ctr., Montreal, QC, Canada;
[2]Dept Psychiatry, McGill University, Douglas Res. Center,, Montreal, QC, Canada

Abstract: Sleep is critical for memory consolidation and can be broken down into rapid eye movement (REM) and slow wave sleep (SWS) phases. During SWS the neural activity patterns from awake experiences are replayed at a compressed timescale. Disrupting these replay events impairs memory performance, suggesting a possible mechanism for memory consolidation during SWS. Recently the theta rhythms in the hippocampus during rapid eye movement (REM) sleep have been shown to be critical for consolidating contextual information, however the underlying mechanisms of consolidation during REM sleep under various contexts is unknown. Here, we combined *in vivo* calcium imaging and electrophysiology to record large neuronal populations in dorsal CA1 during task and REM sleep (n=5 mice, average 585-1255 cells/subject). We used a conventional linear track (LT) and novel half anxiety track (HAT) which is a modified version of linear track with the walls removed on half of the track. The task was recorded in novel, familiar, and anxiogenic contexts. To detect the replay events, bayesian decoding was implemented using the whole cell population and estimated positions were reconstructed during REM sleep using tuning curves from the track sessions. A line fitting method was then applied to decoded positions to find continuous linear trajectory events. With Bayesian decoding analysis, we were able to detect linear trajectory events during REM sleep (~6 events/min). The number of detected replay events significantly exceeded the chance level ($p<0.05$). The quality of replay events (R^2 value) was significantly higher in novel compared to familiar contexts ($p<0.01$). The locations of replay trajectories were biased to the open arm area when the open arm was switched to new location in HAT. In this study, we identified structured replay of previous experiences during REM sleep using the whole cell population instead of place cells alone. In addition, we showed that the quality and content of replay events can be affected by contextual novelty with a bias to the open arm component.

Disclosures: J. Choi: None. **J. Carmichael:** None. **G. Etter:** None. **S. Williams:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.01

Topic: G.01. Fear and Aversive Learning and Memory

Support: NRF Grant 2020R1A2C2014830
MIST Grant 2021M3E5D2A01023887
WISET Grant 2021-418

Title: Immediate extinction deficit with chasing stress as an unconditioned stimulus in rats

Authors: *K.-I. JO, Y. SEONG, I.-H. BAEK, J. JEONG, J.-S. CHOI;
Sch. of Psychology, Korea Univ., Seoul, Korea, Republic of

Abstract: Immediate extinction deficit (IED) refers to lack of extinction effect when the interval between Pavlovian fear conditioning and extinction is relatively short. A conventional Pavlovian conditioning protocol involves paired presentation of auditory conditioned stimulus (CS) and footshock unconditioned stimulus (US). Here we tested whether IED persists following Pavlovian conditioning with a naturalistic US, threat posed by a fast-moving robot, called Chaser. On day 1, all the rats were adapted to a donut-shaped maze (60 cm outer diameter, 18 cm in width, 42 cm high) for 10 min then in a square box (51 cm x 51 cm x 51 cm, W x D x H) for additional 10 min. On day 2, the rats were placed in the maze (Context A) and conditioned with five paired presentations of a tone CS, (2 kHz, 5 s, 80 dB) and chasing by Chaser for 2.5 s as the US. They were divided into 4 groups of 7 each and received either immediate extinction (15 min), delayed extinction (24 hr), immediate no-extinction, or delayed no-extinction in a different context (Context B, the square box). The extinction groups received 30 CS-only presentations. The no extinction groups spent the same amount of time without any CS. On day 4, all four groups received a retention test composed of 5 CS-only presentations in Context B. On day 5, they received a renewal test in Context A. Fear response was measured by freezing using an automated video analysis. The results show that the delayed extinction group froze less than all other groups during the retention test. On the other hand, the immediate extinction group was not different from the no-extinction control indicating that IED was reliably produced when the US was psychological experience such as chasing. There was no statistical difference among groups on the renewal test. These results suggest that IED is a robust phenomenon observed across different types of stress events.

Disclosures: K. Jo: None. **Y. Seong:** None. **I. Baek:** None. **J. Jeong:** None. **J. Choi:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.02

Topic: G.01. Fear and Aversive Learning and Memory

Support: PAPIIT-DGAPA 201420
CONACYT 1146193

Title: Effect of stressors in adolescent rats during long-term sugar consumption & aversive memory

Authors: *I. MORENO MANZANARES, M. I. MIRANDA;
Inst. de Neurobiología, Univ. Nacional Autónoma de México., Queretaro, Mexico

Abstract: Human adolescents tend to vary the quantity and quality of the food they consume depending on the degree of stress experienced. They prefer hyper-palatable foods with higher fat and sugar content when feeling under greater psychological demand. In order to understand how these food preferences are established, we used 42 adolescent male Wistar rats (37 days after birth at the beginning of the experiment) divided into three groups: control (C)(n=12), restrain stress (RS) (n= 16) and foot shock stress (FS)(n=14). All groups had ad libitum access for 21 days of standard lab food, water, and 10% sucrose solution. On days 3, 6, 9, 12, 15, and 18, the RS group received 30 minutes of mild restrain and the FS group one electrical foot shock of 1mA/1s. Every day the weight of the rat, water and sugar consumption, and food were registered. After 21 days of sugar consumption, all rats were subjected to conditioned taste aversive (CTA) and tested for aversive memory extinction. At the end of the experiments, although all groups showed a marked preference for drinking sugar solution, no significant differences were found between groups in body weight, water consumption, sugar solution, or food.Similarly, no significant changes were observed during CTA acquisition, retrieval, or aversive memory extinction sessions. Although the number of studies evaluating the chronic consumption of sugar in adolescent rats is insufficient, there is enough evidence in adults that can be used as a reference to evaluate the behavior we found. Our results disagree with reported studies in adult rats in which the weight of stressed rats is significantly lower than that of stress-free rats. Further studies in adult rats will help elucidate the no effect found in this study.

Disclosures: I. Moreno Manzanares: None. **M.I. Miranda:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.03

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01NS094571
R35NS122181

Title: Investigating the role of mWAKE in the central amygdala

Authors: *J. XIONG[1,2], Q. LIU[3], B. J. BELL[3], M. N. WU[2,3];
[1]1Biochemistry, Cell. and Mol. Biol. Program, [2]Neurosci., [3]Neurol., Johns Hopkins Univ., Baltimore, MD

Abstract: To optimize their fitness, animals organize their physiology and behaviors according to a 24 hr daily rhythm. These rhythms are generated by a core circadian clock, formed at the molecular level by a transcriptional-translational feedback loop. In mammals, the central circadian pacemaker is housed in the suprachiasmatic nucleus (SCN). However, these molecular

clocks are not only enriched in the SCN, but also found in other regions of the body (e.g., the liver) and other regions of the brain. Recent studies have investigated the function and regulation of local clocks in the liver and gut, but the role of local brain oscillators is poorly understood. Our lab discovered a clock output molecule in Drosophila named WIDE AWAKE (WAKE), which mediates the circadian timing of sleep and arousal. Acting in a cell-autonomous manner, mWAKE upregulates specific inhibitory channels and receptors at night to suppress the excitability of arousal-promoting clock neurons at that time. There is a single mammalian homolog of WAKE (mWAKE/ANKFN1), which is expressed in the mouse SCN as well as additional brain regions which have been suggested to house local clocks, such as the central nucleus of the amygdala (CeA) and the lateral amygdala (LA). Ongoing work in our lab suggests that mWAKE promotes rhythmic behaviors in mice by suppressing the excitability of specific neural circuits at night in multiple brain regions, including SCN, LA and dorsomedial hypothalamus (DMH). Here, we begin our investigations of mWAKE and mWAKE+ neurons in the CeA. The CeA plays a critical role in processing fear learning, which is under circadian control, and so we hypothesize that mWAKE and mWAKE+ neurons may regulate rhythms of fear learning. We find that mWAKE labels a cell population in the CeA that is distinct from well-described neuronal clusters expressing somatostatin (SOM) or PKCδ, which suggests that mWAKE+ neurons may constitute a novel neural circuit. Using an $mWake^{Cre}$ line coupled with stereotaxic viral injections, we are currently conducting projection analyses and behavioral assays to determine the role of mWAKE in the CeA. To further investigate the function of mWAKE+ sub-circuits, we recently generated an $mWAKE^{Flp}$ mouse line in which a FLPo sequence is inserted at the end of the last exon of the gene. Using this line in intersectional approaches should enable the functional manipulation of sub-clusters of mWAKE+ neurons and the characterization of circuit mechanisms mediating circadian-related behaviors in the CeA.

Disclosures: J. Xiong: None. **Q. Liu:** None. **B.J. Bell:** None. **M.N. Wu:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.04

Topic: G.01. Fear and Aversive Learning and Memory

Support: NINDS Grant T32NS105602
NIH Grant R01MH122742
Internal funding from UW-Madison School of pharmacy/Medicine and Public Health

Title: Sex dependent modulation of threat assessment by psilocybin

Authors: *J. RAZIDLO[1], J. R. NERAD[2], N. T. JONES[3], Z. ZAHID[4], M. I. BANKS[5], C. J. WENTHUR[6];
[1]Univ. of Wisconsin-Madison, Univ. of Wisconsin - Madison, Neurosci. Training Program,

Madison, WI; [2]Sch. of Pharm., [3]Mol. and Cell. Pharmacol. Program, Univ. of Wisconsin - Madison, Madison, WI; [4]Anesthesiol. and Pharm., Univ. of Wisconsin Madison, Madison, WI; [5]Dept. of Anesthesiol., Univ. of Wisconsin, Madison, WI; [6]Pharm., Univ. of Wisconsin - Madison. Psychoactive Pharmaceut. Investigation Program, Madison, WI

Abstract: Psychiatric disorders such as Major Depressive Disorder (MDD) have become an increasingly prevalent issue and are compounded by the limited efficacy of current treatment options. To combat these disorders, researchers have focused their attention on novel therapeutic approaches. One promising treatment is the serotonergic $5HT_{2A}$ agonist and classical psychedelic, psilocybin. This compound has been shown to exert rapid antidepressant effects through mechanisms that have yet to be fully understood. Psilocybin has been observed to induce an acute stress response, as measured by elevated glucocorticoid release after administration. We propose that the therapeutic effects of psilocybin are due to a transient enhancement of neural plasticity in the ventral hippocampal medial-prefrontal-cortex (HPC-mPFC) pathway and subsequent memory formation produced by this acute stress. Impaired plasticity in the HPC-mPFC pathway is observed in patients with MDD, and individuals have reported decreased depressive symptoms after taking psilocybin. These improvements in mood have been reported to last weeks to months. Despite this, there are no studies to date that assess the duration of this critical window. To study this window of plasticity at the behavioral level, we used C57BL/6J (male and female) mice and the associative learning tasks of both cued and contextual fear conditioning. Mice were administered a single intraperitoneal (IP) injection of saline or psilocybin (3 mg/kg) 4h prior to fear conditioning (FC). In both cued and contextual FC there was no difference in freezing behavior in fear acquisition, nor fear extinction between mice that received saline or psilocybin. Interestingly, male mice that received psilocybin showed reduced fear renewal (unpaired t-test, $p=0.0135$), which was not seen in females. What is even more striking is that females that received psilocybin showed increased fear generalization when introduced to a novel context when compared to saline animals (unpaired t-test, $p=0.0409$). There were no behavioral differences between treatment groups or sexes in our contextual FC experiments. Future aims will focus on altering the timing of drug administration relative to FC as well as altering the dose of psilocybin.

Disclosures: **J. Razidlo:** None. **J.R. Nerad:** None. **N.T. Jones:** None. **Z. Zahid:** None. **M.I. Banks:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); VCENNA, Inc.. F. Consulting Fees (e.g., advisory boards); VCENNA, Inc. **C.J. Wenthur:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Psilera. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Usona.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.05

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH Grant R01MH125615
NSF Grant IIS-1908299 III

Title: Differential Aversive and Appetitive Conditioning in Artificial Neural Networks

Authors: *S. LEEM, A. KEIL, P. LIU, M. DING, R. FANG;
Univ. of Florida, Univ. of Florida, Gainesville, FL

Abstract: Artificial neural networks (ANNs), especially deep convolutional neural networks (DCNNs), are excellent models of the ventral visual stream in the human and the nonhuman primate brain. For visual object recognition, DCNNs perform on par with human observers. Aiming to augment the capability of DCNNs to model more complex cognitive functions such as learning, memory, and emotion, we have recently developed an ANN model, referred to as VCA or visual cortex amygdala model, which is capable of labeling hedonic valence in natural images varying in affective content. The current study further tested the hypothesis that the VCA model can associate the aversive (or appetitive) value of an unconditioned stimulus (US) with an initially neutral conditioned stimulus (CS). Gabor patches parametrized by orientation were used as CS and were paired with images from the International Affective Picture System (IAPS) serving as US in a differential Pavlovian conditioning procedure: Two Gabor patches with orthogonal orientations were repeatedly (100 times) presented to the VCA model together with unpleasant and pleasant images, respectively. The model parameters were updated upon each presentation to minimize the difference between the model predicted valence and the normative valence of each IAPS image. We found that (1) the initially neutral cues, when tested alone, began to acquire the emotional quality of the paired conditioned images as learning progressed and (2) this property parametrically generalized to other Gabor patches not seen by the model during the learning process. Similar results were found when varying the contrast or the spatial frequency of the Gabor patches albeit that the learning rate for conditioning contrast or frequency was slower than that for orientation. These simulations demonstrate that the VCA model generates results comparable to human behavioral Pavlovian conditioning experiments and, more generally, suggest that the VCA model can be used in conjunction with empirical studies to deepen our understanding of the neural basis of affective processing and associative learning in the human brain.

Disclosures: **S. Leem:** None. **A. Keil:** None. **P. Liu:** None. **M. Ding:** None. **R. Fang:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.06

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH Grant 8K00 DA055493-03
NIH Grant R01 DA047981

Title: Sex and estrous cycle effects in stress-enhanced fear learning

Authors: *S. A. LOPEZ, K. M. LATTAL;
Oregon Hlth. & Sci. Univ., Portland, OR

Abstract: Traumatic events have long-lasting effects on future behavior. For example, individuals diagnosed with post-traumatic stress disorder (PTSD) are hyper-responsive to otherwise ordinary stimuli (e.g., loud noises) long after the trauma. For women, the likelihood of developing PTSD following trauma is greater, the comorbid progression to substance use disorder (SUD) is more rapid, and although less clear, PTSD-SUD comorbidity rates can be higher. Yet, much of the preclinical work assessing the neurobiology of stress-related psychopathology has focused on males. To date, sex differences in fear learning have been reported but are inconsistent; and understanding of the influence of ovarian hormones in this context is further limited. Here, we characterized male and female rat behavior across the stress-enhanced fear learning (SEFL) paradigm. Rats underwent a single massive shock event, mild stress exposure, and a context fear conditioning, or SEFL test. Using this stress procedure, it has been previously demonstrated that exposing male rats to a battery of foot shocks in one context potentiates their fear response (i.e., freezing) to a mild stressor (i.e., single foot shock) in a different context that persists long after the initial massive foot shock experience. SEFL has a measurable memory component and captures behavioral outcomes resembling PTSD, making it instrumental in increasing our understanding of PTSD neural substrates. While previous work using SEFL procedures suggests the persistent effects of "trauma" on mild stress responsivity holds true in female rats, these effects have not been thoroughly characterized, and the effects of estrous cycle remain unknown. We found that females with a history of "trauma" (4 or 15 shocks) experienced in one context have higher freezing levels during the SEFL test, relative to the no shock controls. As seen in males, this SEFL effect is long-lasting, captured even thirty days following exposure to a mild stressor (a single footshock). We also evaluated several different behaviors during conditioning and testing, including rearing, jumping, and darting. Although freezing levels across each stage of the SEFL paradigm were not significantly different between males and females, females displayed unique escape-like behaviors the day of “trauma” and mild stress exposure. In addition, monitoring of the estrous cycle revealed the influence of cycle phase the day of “trauma” on future fear learning. These findings are fundamental to future work investigating the neurobiology of stress-related psychopathologies and in determining any sex differences contributing to female susceptibility.

Disclosures: S.A. Lopez: None. **K.M. Lattal:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.07

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH Grant R01MH122561

Title: Investigation of diverse context-specific behavioral responses during fear extinction

Authors: *Q.-S. E. LE, A. GOODMAN, R. VUTUKURI, C. D. BORKAR, D. HEREFORD, A. RESENDEZ, J. P. FADOK;
Tulane Univ., Tulane Univ., New Orleans, LA

Abstract: Severe trauma can leave lasting detrimental effects, such as persistent fear memories and maladaptive responses that are often difficult to extinguish. Given that women experience greater difficulty in treatment for fear disorders and are twice as likely to develop post-traumatic stress disorder, we investigated sex as a factor during fear extinction acquisition and recall. We used a modified Pavlovian conditioning paradigm pairing footshocks with a serial compound stimulus (SCS) consisting of distinct auditory tone and white noise (WN) cues in adult male and female mice (C57BL/6J, N=16 each sex, 3-6 months). We previously used this SCS fear conditioning paradigm to elicit distinct freezing and flight behaviors to the SCS's tone and WN portions, respectively (Fadok et al., 2017, Borkar et al., 2020). To investigate sex differences in behavioral responses during fear extinction, after fear conditioning, we presented the SCS alone in the conditioning context for two extinction sessions of 16 trials each, then tested extinction recall in the conditioning context and in a neutral context. Throughout the paradigm, we recorded freezing, flight, escape jumps, tail rattling, grooming, and rearing behaviors to assess changes in the mouse ethogram. We observed significant changes in defensive behaviors during and after fear extinction in both males and females. During extinction, freezing to tone decreased, flight and escape jumping during WN decreased, and freezing during WN increased. Additionally, tail rattling and rearing frequencies decreased after fear extinction. However, we did not find sex differences in these behavioral changes. We did find that females displayed greater inter-trial freezing compared to males during the first extinction session, and males groomed for longer than females during extinction recall in the conditioning context. Within the neutral context, grooming frequency increased, and tail rattling did not occur. Unpaired control mice (C57BL/6J, N=10 each sex, 3-4 months) did not display SCS-evoked freezing or flight behaviors during extinction sessions, nor did they show similar changes after extinction. Overall, our results highlight distinct behavioral profiles to fear extinction that warrant further analysis. Given our previous work showing that central amygdala somatostatin- and corticotropin-releasing factor-positive cells drive distinct SCS-evoked freezing and flight behavior profiles (Fadok et al., 2017), we are currently investigating circuits involving these cells and their influence on behavioral changes during and after extinction of SCS-evoked fear.

Disclosures: Q.E. Le: None. **A. Goodman:** None. **R. Vutukuri:** None. **C.D. Borkar:** None. **D. Hereford:** None. **A. Resendez:** None. **J.P. Fadok:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.08

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01-MH117791

Title: More than freezing: temporal ethograms reveal a fear-conditioned cue to orchestrate diverse behaviors

Authors: *A. CHU, C. B. MICHEL, N. T. GORDON, K. E. HANRAHAN, A. M. DUBOIS, D. C. WILLIAMS, M. A. MCDANNALD;
Psychology and Neurosci., Boston Col., Chestnut Hill, MA

Abstract: In Pavlovian fear conditioning, a neutral cue is paired with an aversive stimulus, such as foot shock. One result of this predictive relationship is the fear-conditioned cue will elicit defensive behavior. The most commonly measured behavior is freezing, a 'passive' behavior defined by the absence of movement. Recent studies have shown that a fear-conditioned cue can elicit 'active' defensive behaviors, characterized by increased movement. The goal of Experiment 1 was to construct ethograms of rat behavior spanning passive defensive, active defensive, and reward-related behavior across Pavlovian fear discrimination. We tested 24 rats (12 females) in a discriminative conditioned suppression procedure. Rats were trained to nose poke for a food reward, then received 16 fear discrimination sessions in which three auditory cues predicted unique foot shock probabilities: danger (p=1), uncertainty (p=0.25), and safety (p=0). Poke-reward and cue-shock contingencies were independent. We captured images at a rate of 5 frames/s before and during cue presentation (5 s pre-cue, 10 s during cue) and quantified 10 behaviors. Temporal ethograms reveal the fear-conditioned, danger cue to orchestrate a suite of behaviors. By the end of discrimination, danger elicited a mixture of passive (freezing), active defensive behaviors (locomotion, jumping, and rearing) while suppressing reward-related behaviors. Discrimination consisted of restricting these behaviors to the danger cue, as analysis of early discrimination showed all three cues to initially orchestrate these same behaviors. Freezing was not the dominant danger-elicited behavior and its expression differed between sexes. Finally, danger suppression of reward-related behaviors was not a byproduct of freezing, as there was no relationship between these two behaviors across subjects. Building on our results, Experiment 2 aims to investigate specific roles for ventral tegmental area (VTA) dopamine neurons, and their terminals in the basolateral amygdala (BLA), in the acquisition and expression of active defensive behavior. 24 rats (12 females) received sham or 6-OHDA infusions into the VTA (deleting dopamine neurons) or BLA (deleting dopamine fibers). Following recovery, all rats underwent fear discrimination, frame capture, and behavior analysis as in Experiment 1. Full analysis and results of Experiment 2 will be presented. Our hand-scoring efforts reveal fine organization of behavioral responding and highlight the need for assessing active defensive behaviors alongside freezing. Our findings are further paving the way for investigations into the neural basis of active defensive behaviors.

Disclosures: A. Chu: None. **C.B. Michel:** None. **N.T. Gordon:** None. **K.E. Hanrahan:** None. **A.M. DuBois:** None. **D.C. Williams:** None. **M.A. McDannald:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.09

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH Award 1U01NS122082-01

Title: Basolateral amygdala oscillations enable association between neutral stimuli and threat during fear conditioning in a biophysical network model

Authors: *A. CATTANI[1], D. ARNOLD[2], M. M. MCCARTHY[1], N. J. KOPELL[1];
[1]Boston Univ., Boston, MA; [2]USC, Los Angeles, CA

Abstract: In the fear conditioning (FC) paradigm, a neutral stimulus (conditioned stimulus, CS, such as a tone) evokes a fear response through a learned association with an aversive stimulus (unconditioned stimulus, US, such as a mild shock). The basolateral amygdala (BLA) is thought to be the main site where the association takes place through structural synaptic plasticity involving, but not restricted to, fear-encoding projection neurons. Evidence from rodent research suggests that FC confers to BLA increased power at low theta (~4 Hz) (Davis et al., 2017) and gamma (Courtin et al., 2013) frequencies detected in local field potential (LFP) recordings. On the contrary, high theta (~8 Hz) remains mostly unaffected by FC, and it is thought to be associated with fear extinction (Davis et al., 2017).Here, we aim to shed new light on the role of inhibitory interneurons in generating BLA oscillatory patterns and to elucidate the functional role of rhythms in FC. We used a biophysically detailed model of the BLA circuit (see Fig. 1) that contains excitatory projection neurons, as well as interneurons (VIP, SOM, and PV), which are responsible in isolation for low theta, fast theta, and gamma rhythms, respectively. We found that all the interneuron types projecting directly or indirectly to the projection neurons, are needed for FC. Interneurons and their rhythms have differential effects on projection neuron spiking activity. First, interneurons cooperate during VIP low theta peak phases to enable the correct fine-timing between projection neurons activated by CS (ECS) and fear-encoding projection neurons (F). Furthermore, interneurons cooperate to pause the activity of ECS and F during VIP low-theta troughs. This alternation between fine-timing and pauses at low-theta allows potentiation of the synaptic conductance between ECS and F via spike-timing dependent plasticity (STDP), thus establishing the association between CS and fear. The lack of any single subtype of interneuron leads systematically to synaptic depression, which translates into a shortfall of association. Finally, by modeling the LFP of our circuit activity, we found an increase of low-theta frequencies during the onset of CS presentation in the fear-conditioned network, thus corroborating experimental results in rodents.These results suggest that BLA interneurons, and the rhythms they generate, may serve as network elements critical to the instantiation of FC. Extending our BLA network to include also fear extinction circuits, this work may suggest targets for therapies aimed at preventing pathological fear conditioning and renewal.

Disclosures: A. Cattani: None. **D. Arnold:** None. **M.M. McCarthy:** None. **N.J. Kopell:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.10

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01-MH117791

Title: Probing the lower limits of probabilistic threat learning to reveal roles for the ventral pallidum

Authors: *E. RUSSELL, M. MCDANNALD;
Boston Col., Chestnut Hill, MA

Abstract: Probing the lower limits of probabilistic threat learning to reveal roles for the ventral pallidum
Emma L. Russell & Michael A. McDannaldBoston College, USA
A fundamental task of the brain is to assign valence to events in our environment. Rewarding events should elicit approach and consumption, while threatening events should elicit defensive behaviors. The ventral pallidum is anatomically poised to contribute to reward and threat behavior. In reward settings, ventral pallidum neurons scale firing increases according to palatability. In threat settings, ventral pallidum neurons scale firing decreases according to foot shock probability. Firing decreases are greatest to a danger cue predicting 100% foot shock, intermediate to an uncertainty cue (25%), but absent to a safety cue (0%). The goal of the current study was to reveal roles for the ventral pallidum in threat learning. To do this, Experiment 1 sought to establish discrimination procedures that would isolate the two main behavioral components of discrimination: selectively acquiring ‘fear’ to a threat cue, and reducing fear to a neutral cue. Forty-seven Long Evans rats (23 females) were mildly food-deprived and trained to nose poke for food pellets. Rats were then assigned to one of three groups and received Pavlovian fear discrimination. For each group, a threat cue probabilistically predicted foot shock on 10% (n=15), 20% (n=16), or 30% (n=16) of trials, while a neutral cue never predicted foot shock. 10 threat and 10 neutral trials were given per session, meaning 10% rats received a single foot shock per session. All groups showed complete discrimination. However, 10% rats selectively acquired moderate nose poke suppression to the threat cue. 20% and 30% rats initially generalized suppression to both cues, before selectively showing high levels of suppression to the threat cue. Experiment 2 used a dual viral approach (AAV-eSYN-EGFP-T2A-iCre + AAV-flex-taCasp3-TEVp) to delete ventral pallidum neurons in an experimental group, while a control group had neurons left intact. Delete and Control rats were then assigned to the 10% or 30% foot shock group and Pavlovian fear discrimination given. A full analysis of Experiment 2 will be presented. The viral approach in Experiment 2 is setting up pathway-specific manipulations of ventral pallidum outputs to the basolateral amygdala or ventral tegmental area. In total, these experiments are working to pinpoint roles for the ventral pallidum in threat learning.Keywords: fear conditioning, discrimination, shock, casp3, ventral pallidum

Disclosures: **E. Russell:** None. **M. McDannald:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.11

Topic: G.01. Fear and Aversive Learning and Memory

Title: Effects of partial reinforcement on fear extinction learning in mice

Authors: *Y. FUKUNAGA, C. J. SU, V. A. CAZARES;
Psychology and Neurosci. Program, Williams Col., Williamstown, MA

Abstract: Anxiety-related disorders are characterized by a dysregulated response to potential threat, including generalized and/or persistent avoidance of feared stimuli. The processes underlying maladaptive threat or fear learning can be studied using fear conditioning and extinction. Fear conditioning is an associative learning process by which organisms learn when environmental stimuli (conditioned stimuli, CS) are predictive of aversive outcomes (unconditioned stimulus, US). Fear extinction is the learning process by which organisms learn to reduce defensive responses when the CS no longer predicts the US. These studies are focused on understanding a paradoxical finding showing that intermittent CS-US pairings during fear conditioning can lead to resistance to extinction learning, relative to a conditioning schedule following a 1:1 CS-US frequency ratio. This effect has been termed the partial reinforcement extinction effect (PREE) and has been largely studied in operant and reward-based learning. However, significantly less is known about the PREE in fear learning. Our primary objective was to investigate the effects of partial reinforcement in fear acquisition, fear memory consolidation, and recall in mice. Furthermore, we investigated whether the effects of partial reinforcement are mediated by the number of CS presentations or CS duration. To achieve this, C57BL/6J mice were conditioned to an auditory CS paired with a mild footshock (US) for two days. Following conditioning, extinction was carried out for three consecutive days, where each day one session of extinction consisted of 12 unreinforced CS presentations. Finally, to assess the effects of partial reinforcement on fear extinction memory, we measured fear recall at 48 hours and 30 days later, given that previous studies show that the passage of time leads to the spontaneous recovery of fear. Our preliminary results demonstrated that partial reinforcement (0.5, CS:US) enhanced fear acquisition and fear recall on extinction day 1. In contrast to previous studies, our results showed no effect on extinction acquisition, consolidation or fear recall (48 hours and 30 days tone test). Taken together, the present study suggests a small or minimal role of PREE on fear extinction learning in contrast to PREE previously reported in the context of appetitive learning.

Disclosures: **Y. Fukunaga:** None. **C.J. Su:** None. **V.A. Cazares:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.12

Topic: G.01. Fear and Aversive Learning and Memory

Support: National Research Foundation of Korea (MEST 2020R1A2C2102134)

Title: Activation of PBN CGRP neurons promotes the arrangement of defensive behavior appropriate to the context

Authors: *H. CHO, G. PYEON, I.-H. BAEK, Y. JO;
Sch. of Psychology, Korea Univ., Seoul, Korea, Republic of

Abstract: Fear is expressed by a wide spectrum of behaviors and it is crucial to orchestrate defensive behaviors that match distinct threat circumstances. Freezing is highly effective in response to repeated electric footshock by an invisible predator, whereas darting, in which an animal rapidly moves forward in an attempt to flee from perceived danger, is most appropriate when chased by a predator. The neurons expressing calcitonin gene-related peptide (CGRP) in the parabrachial nucleus (PBN) are known as a general alarm signal which respond to aversive stimuli and alert the forebrain of ongoing or potential threats. To determine whether $CGRP^{PBN}$ neurons relay aversive information and contribute to defensive response, we bilaterally injected adeno-associated virus carrying Cre-dependent channelrhodopsin and implanted fiber-optic cannulae over PBN of $Calca^{cre/+}$ mice. We activated $CGRP^{PBN}$ neurons for 30 s and confirmed that repeated 30 Hz stimulation of $CGRP^{PBN}$ neurons evoked robust time-locked freezing behavior in mice. We investigated whether activation of $CGRP^{PBN}$ neurons promote adaptive defensive behavior that fits the context using a naturalistic threat paradigm. Chasing threat paradigm mimics an imminent threat situation where mice are placed in a circular track and chased by a predator-like robot. The ecological setting allows the observation of darts, which were defined as rapid movements preceded by immobility. We subjected mice to threat conditioning in which a tone (10 s, 70 dB) was paired with a predator-like robot chasing (3 s, 0.58 m/s). We activated $CGRP^{PBN}$ neurons of $Calca^{cre/+}$ mice (n = 5) during the chasing threat and observed a significant decrease in freezing compared to the control group (n = 7) when the tone was presented alone the next day. Instead, $CGRP^{PBN}$-activated mice showed greater darting compared to the control group. Followed by an extinction training, the control group displayed decreased darting whereas darting by $CGRP^{PBN}$-activated mice was maintained and significantly greater compared to the control group. Indeed $CGRP^{PBN}$-activated mice showed significantly less freezing compared to the control group. We discovered that mice that received $CGRP^{PBN}$ stimulation learned to dart in response to the cue associated with the chasing threat and showed impaired extinction learning. Our findings suggest that $CGRP^{PBN}$ neurons are the core alarm system in which the enhanced alarm signal by $CGRP^{PBN}$ neurons elicited behavioral responses appropriate to distinct threat stimuli.

Disclosures: H. Cho: None. **G. Pyeon:** None. **I. Baek:** None. **Y. Jo:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.13

Topic: G.01. Fear and Aversive Learning and Memory

Support: Leopoldina Grant LPDS2021-05
VICI Grant 453-15-009

Title: Investigating the mechanistic role of painful self-experience in emotional contagion: an effect of autoconditioning?

Authors: *J. PACKHEISER[1], E. SOYMAN[2], E. PARADISO[1], E. RAMAAKER[1], N. SAHIN[1], S. MURALIDHARAN[1], M. WÖHR[3], V. GAZZOLA[1], C. KEYSERS[1];
[1]Social Brain Lab., Netherlands Inst. for Neurosci., Amsterdam, Netherlands; [2]Kadir Has Univ., Istanbul, Turkey; [3]KU Leuven, KU Leuven, Leuven, Belgium

Abstract: Emotional contagion refers to the transmission of emotions from one conspecific to another. Previous research in rodents has demonstrated that the self-experience of footshocks enhances how much an observer is affected by the emotional state of a conspecific in pain or distress. We hypothesized auditory auto-conditioning to contribute to this enhancement: during the observer's own experience of shocks, the animal associates its own audible nocifensive responses, its pain-squeaks in particular, with the negative affective state induced by the shock. When the animal later witnesses a cage mate receive shocks and hears it squeak, the previously strengthened connection between fear and squeaks could be a mechanism eliciting the enhanced fearful response in the observer. As hypothesized, in a first study, we found pre-exposure to shocks to increase freezing and distress calls upon the playback of pain-squeaks. Freezing was also increased during the playbacks of phase-scrambled squeaks, but distress calls were more frequent during the playback of regular squeaks. Core to the notion of autoconditioning is that the effect of pre-exposure is due to the pairing of a pain-state with hearing one's own pain squeaks. In a second study, we therefore compared the response to squeak playbacks after animals had been pre-exposed to pairings of a CO2 laser with a squeak playback against three control groups that were pre-exposed to the CO2 laser alone, to squeak playbacks alone or to neither of these conditions. We however could not find any differences in freezing or distress calls among all experimental groups. In summary, we demonstrate the sufficiency of pain-calls to trigger fear in a way that critically depends on the nature of an animals prior experience and discuss why the pairing of a CO2 laser with pain squeaks cannot substitute footshock pre-exposure.

Disclosures: J. Packheiser: None. **E. Soyman:** None. **E. Paradiso:** None. **E. Ramaaker:** None. **N. Sahin:** None. **S. Muralidharan:** None. **M. Wöhr:** None. **V. Gazzola:** None. **C. Keysers:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.14

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIAAA R01AA024434 (K.R.K.)
NIAAA F32AA29595 (J.S.H.)
NIGMS R25GM125500 (N.L.)

Title: Operant responses in to appetitive and aversive odors in Drosophila

Authors: *N. LE[1], J. S. HERNANDEZ[2], N. J. MEI[2], R. AZANCHI[2], K. R. KAUN[2];
[1]Post-baccalaureate Res. Educ. Program, [2]Neurosci., Brown Univ., Providence, RI

Abstract: Animals learn to perform appropriate responses in complex, natural environments by altering their behavior in response to positive or negative outcomes. The underlying neural basis for this operant conditioning is not well understood in current model organisms due to the variability in behavioral outcomes stemming from the same experience. *Drosophila melanogaster* is an enticing model to study the neural basis of operant behavior because of its well-defined memory circuits, well-characterized behavioral responses, and the ability to study a large number of animals. To measure operant memory in *Drosophila*, we developed an automated runway assay in which flies enter a defined region of interest to consequently release an appetitive or aversive odor. We examined male and female 3-5 day old wild-type Canton-S *Drosophila* self-administering appetitive (apple cider vinegar or 3% ethanol) and aversive (benzaldehyde or geosmin) odors across 3 days. In order to track individuality in behavior, flies were housed individually and allowed to self-administer the odor for 15 minutes each day. We compared the operant behaviors of flies when self-administering appetitive and aversive odors using computer vision and machine learning methods. We found that both male and female flies show individual variation in their responses to an odor, such that some flies spent the majority of their time self-administering the odor, and others spent most of their time avoiding the odor. This was true for both appetitive and aversive odors. Female flies that found 3% ethanol appetitive self-administered the odor longer than male flies that found 3% ethanol appetitive. No sex differences were found in 3% ethanol self-administration in flies that did not find 3% ethanol appetitive. Similarly, no sex differences were found in flies in self-administration for aversive odors. Ultimately, these findings can be used to predict behaviors demonstrated in learning both appetitive and aversive outcomes.

Disclosures: N. Le: None. **J.S. Hernandez:** None. **N.J. Mei:** None. **R. Azanchi:** None. **K.R. Kaun:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.15

Topic: G.01. Fear and Aversive Learning and Memory

Support: NRF-2021R1A6A3A01086526
NRF-2022R1A2C2009265
NRF-2022M3E5E8018421
NRF-2018M3C7A1024736
NRF-2021R1C1C1006607

Title: Mouse model of fear generalization using restraint stress

Authors: *S. PARK[1], Y. HUH[2,3], J. CHO[1];
[1]Brain and Cognitive Sci., Scranton College, Ewha Womans Univ., Seoul, Korea, Republic of; [2]Dept. of Med. Sci., Catholic Kwandong Univ., Gangneung-si, Korea, Republic of; [3]Intl. St. Mary's Hosp., Translational Brain Res. Center, Catholic Kwandong Univ., Incheon, Korea, Republic of

Abstract: The main symptom of anxiety disorders, as in post-traumatic stress disorder (PTSD), is overgeneralization, marked by widespread distress responses to harmless stimuli or situations. Animal models have been used as a valuable tool to study detailed mechanisms or disorders. A mouse model of generalized fear, therefore, will likely provide insight into understanding and treating anxiety disorders. Here we report a model that may be used as a mouse model of generalized fear. In this model, we modified a shock-probe defensive burying behavioral test and used restraint stress in place of an electric shock. We found that mice that experienced restraint stress displayed elevated anxiety-like behaviors and defensive burying of a restrainer-resembling object, which is not the restrainer used to induce restraint stress, similar to behaviors of generalization. The degree of generalization did not generalize to extend to a neutral object, which had lower resemblance to the restrainer. More experiments need to be carried out to confirm the extent of generalization, but our preliminary results suggest that this model may be used for studying fear generalization.

Disclosures: S. Park: None. **Y. Huh:** None. **J. Cho:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.16

Topic: G.01. Fear and Aversive Learning and Memory

Support: Hirschl/Weill-Caulier Trust Award
McKnight Memory and Cognitive Disorders Award
McKnight Doupe Fellowship
Klingenstein-Simons Fellowship
Brain Research Foundation Award
NARSAD Young Investigator Award
Botanical Center Pilot Award

Title: Unbiased identification of a novel hypothalamic nucleus that regulates persistent fear and anxiety states after severe stress

Authors: *A. LABANCA[1], Z. T. PENNINGTON[2], A. C. W. SMITH[3], Z. DONG[4], P. SOMPOLPONG[4], R. L. CLEM[5], P. J. KENNY[6], D. J. CAI[7];
[2]Dept. of Neurosci., [1]Icahn Sch. of Med. At Mount Sinai Grad. Training Program In Neurosci., New York, NY; [3]The Med. Univ. of South Carolina, Charleston, SC; [4]Icahn Sch. of Med. at Mount Sinai, New York, NY; [5]Neurosci., Mount Sinai Sch. of Med., New York, NY; [6]Neurosci., Icahn Sch. of Med. At Mount Sinai, New York, NY; [7]Mount Sinai, New York, NY

Abstract: Acute severe stress is able to produce lasting and dramatic enhancements of fear and anxiety, promoting a range of psychiatric illnesses. How this sensitization is instantiated in the nervous system remains unclear. To address this issue, we first utilized unbiased whole-brain immediate early gene imaging of mice to examine how a severe stressor is able to augment subsequent behavioral responses to a novel stressor a week later. In addition to observing broad cortical hypoactivation in previously stressed animals relative to previously unstressed animals, hyperactivation of several subcortical regions was found, including the anterior hypothalamic nucleus (AHN). Notably, despite the AHN's rich interconnections with stress circuitry, its role in stress has been little explored. To assess the causal contribution of the AHN to persistent fear and anxiety states after severe stress, we inactivated this structure during an acute severe stressor to assess both its acute and long-term consequences. AHN inactivation reduced fear responses during the severe stressor; moreover, multiple measures of fear and anxiety were reduced a week later when the AHN was online (memory of the severe stressor, anxiety-like behavior, and responses to a novel stressor). To delineate the cell population involved, we separately tested the effects of inhibiting GABAergic and glutamatergic cells within the AHN and found that GABAergic, and not glutamatergic, AHN neurons support stress behaviors. Lastly, we have begun single photon calcium imaging of the AHN and found that the AHN responds to acute stressors, and ongoing work is examining how GABAergic AHN neural activity is modified by the prior experience of severe stress. By understanding the contribution of this previously underappreciated nucleus and cell population to stress behaviors, potentially novel targets for stress-induced pathology may be uncovered.

Disclosures: A. LaBanca: None. **Z.T. Pennington:** None. **A.C.W. Smith:** None. **Z. Dong:** None. **P. Sompolpong:** None. **R.L. Clem:** None. **P.J. Kenny:** None. **D.J. Cai:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.17

Topic: G.01. Fear and Aversive Learning and Memory

Support: Hirschl/Weill-Caulier Trust Award
McKnight Memory and Cognitive Disorders Award
Klingenstein-Simons Fellowship
McKnight Doupe Fellowship
Klingenstein-Simons Fellowship
Brain Research Foundation Award
NARSAD Young Investigator Award

Title: Plasticity in dissociable neural circuits supports the divergent consequences of severe stress

Authors: *Z. PENNINGTON, A. LABANCA, Z. DONG, P. SOMPOLPONG, S. KOSARUSSAVADI, D. J. CAI;
Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Acute severe stress is able to produce an array of changes in fear and anxiety-like behavior, including defensive freezing to stress-associated stimuli, heightened reactions to novel aversive events, and increases in anxiety-like behavior. Whether these changes arise from plasticity in a common circuit, or if distinct circuits support the various consequences of severe stress, remains unknown. To address this question, we combined pharmacologic, *in vivo* imaging, and ensemble tagging strategies with a model in which mice are given an acute severe stressor and a week later are exposed to a battery of fear and anxiety-like behavior tests. Firstly, we found that blocking protein synthesis induced by severe stress in the basolateral amygdala (BLA) and ventral hippocampus (vHC) have doubly-dissociable impacts on subsequent fear and anxiety-like behavior. Blockade of protein synthesis in the BLA profoundly reduced associative freezing in the stressor context and mitigated heightened responses to a novel stressor, but had no impact on the anxiety-like behavior of the same animals. Conversely, blockade of protein synthesis in the vHC attenuated stress-induced changes in anxiety-like behavior but had no impact on responses to novel stressors and had minimal impact on associative freezing. Secondly, using chemogenetics, we demonstrated that neural activity in BLA and vHC have similar dissociable contributions to the expression of these post-stress phenotypes. Thirdly, in an attempt to isolate ensembles of BLA and vHC neurons supporting these changes, we utilized activity-dependent tagging of stress-reactive cells and looked at their reactivation in response to subsequent events. We found stress reactive ensembles in the BLA (but not the vHC) are more likely to be reactivated by novel stressors, suggesting that stress-induced plasticity specifically within this cell population supports the heightened response to novel stressors. Collectively, these results indicate that plasticity in separate BLA and vHC circuits support distinct impacts of stress on behavior, and ongoing research aims to elaborate on the unique role of stress-reactive ensembles. Critically, these results suggest that the impacts of severe stress are biologically divergent. If true, this might indicate that interventions geared towards one these post-stress phenotypes (e.g., associative memories) in conditions like PTSD may not affect other post-stress phenotypes (e.g., anxiety).

Disclosures: **Z. Pennington:** None. **A. LaBanca:** None. **Z. Dong:** None. **P. Sompolpong:** None. **S. Kosarussavadi:** None. **D.J. Cai:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.18

Topic: G.01. Fear and Aversive Learning and Memory

Title: Threat controllability modulates neural reactivity during Pavlovian fear conditioning

Authors: *A. BOLARAM[1], J. B. PURCELL[1], H. E. DARK[1], D. K. GREY[1], A. J. KNIGHT[2], D. C. KNIGHT[1];
[1]Dept. of Psychology, [2]Physical Med. & Rehabil., The Univ. of Alabama at Birmingham, Birmingham, AL

Abstract: Pavlovian conditioning procedures are often used to study the neural processes that underlie fear learning and memory. Although prior work suggests the ability to control threats (e.g., unconditioned stimuli; UCS) impacts emotional function, the neural processes that underlie fear conditioning to controllable and uncontrollable threats has received less attention. Therefore, the present study assessed the functional magnetic resonance imaging (fMRI) signal of participants (N=35, [27 female], Mean Age = 24.85) who completed a Pavlovian fear conditioning procedure designed to investigate the conditioned neural response to warning (CS+) and safety (CS-) signals under controllable and uncontrollable threat (UCS) conditions. Two visual stimuli (green and yellow squares) served as conditioned stimuli (CSs; 8 second duration) and a loud, 100 dB white noise served as the UCS (0.5-6.0 second duration). One CS (CS+; 20 trials) was always (100% pairing rate) paired with the UCS. The other CS (CS-; 20 trials) was never paired with the UCS (0% pairing rate). Stimuli were presented during both Controllable and Uncontrollable threat (UCS) conditions. During Controllable trials participants could terminate the UCS (Controllable threat) by a button press. During Uncontrollable trials the button press did not terminate the UCS (Uncontrollable threat). Instead, the duration of the uncontrollable UCS was yoked with the duration of a prior participant's controllable UCS to ensure UCS durations were equivalent across Controllable and Uncontrollable conditions. Neuroimaging data were collected using a 3T Siemens Prisma scanner. High resolution T1-weighted anatomical images were obtained in the sagittal plane using an MPRAGE sequence. Whole brain fMRI data were acquired using a gradient echo echoplanar image sequence. Multivariate ANOVA revealed a significant main effect of CS type (i.e., CS+ vs CS-) in the dorsomedial prefrontal cortex (dmPFC), dorsolateral prefrontal cortex (dlPFC), ventromedial prefrontal cortex, hippocampus, insula, and inferior parietal lobule. A significant main effect of UCS Controllability (i.e., Controllable vs Uncontrollable) was observed in ventrolateral prefrontal cortex (vlPFC), dlPFC, and hippocampus. A CS type x UCS Controllability interaction was observed within the dmPFC, dlPFC, insula, amygdala, and parahippocampal gyrus. The present findings suggest that threat controllability alters the conditioned neural

response within brain regions that support fear learning, expression, and regulation processes, which may have important implications for emotion-related disorders.

Disclosures: **A. Bolaram:** None. **J.B. Purcell:** None. **H.E. Dark:** None. **D.K. Grey:** None. **A.J. Knight:** None. **D.C. Knight:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.19

Topic: G.01. Fear and Aversive Learning and Memory

Support: CRC 1280, project number 316803389
Marie Skłodowska-Curie Innovative Training Network CEN (Cerebellum and Emotional Networks)

Title: Effects of dopaminergic and anti-dopaminergic drugs on fear extinction learning in healthy human participants.

Authors: *A. DOUBLIEZ[1], L. MÜNTEFERING[1], K. KÖSTER[1], E. NIO[1], N. DIEKMANN[3], S. CHENG[3], C. J. MERZ[4], A. THIEME[1], B. ALBAYRAK[2], S. A. NICKSIRAT[1], F. ERDLENBRUCH[1], G. BATSIKADZE[1], T. M. ERNST[1], D. TIMMANN[1];
[1]Dept. of Neurol. and C-TNBS, [2]Dept. of Pediatrics I and C-TNBS, Essen Univ. Hospital, Univ. of Duisburg-Essen, Essen, Germany; [3]Inst. for Neural Computation, [4]Dept. of Cognitive Psychology, Inst. of Cognitive Neurosci., Ruhr Univ. Bochum, Bochum, Germany

Abstract: The ability to extinguish learned fear responses is crucial for adaptive behavior. The mesolimbic dopaminergic system originating in the ventral tegmental area has been proposed to contribute to fear extinction learning because of its critical role in reward learning. The unexpected omission of aversive unconditioned stimuli (US) is considered as rewarding (outcome better than expected) and to drive extinction learning (see e.g. Kalisch et al., Trends Cogn Sci. 2019;23:274-277). We tested the hypothesis that extinction learning is facilitated by dopaminergic drugs and impeded by anti-dopaminergic drugs. The effects of dopamine agonists (levodopa (100 mg) and bromocriptine (1.25 mg)) and antagonists (tiapride (100 mg) and haloperidol (3 mg)) on fear extinction learning were compared to placebo in 150 young and healthy human participants. A three-day differential fear conditioning paradigm was performed with pupil size and skin conductance responses (SCRs) being recorded. Fear acquisition training was performed on day 1, extinction training on day 2, and recall was tested on day 3. The conditioned stimuli (CS+, CS-) consisted of two geometric figures. A short electrical stimulation was used as the aversive US. One of the four drugs or placebo was administered prior to the extinction phase on day 2. Serum drug levels were measured at the end of each experimental day. At the time of abstract submission, data acquisition applying levodopa, tiapride or placebo prior extinction has been completed, and pupil size has been fully analyzed. During fear acquisition

training, pupil size was significantly larger in CS+ compared to CS- trials with no significant difference between groups (non-parametric ANOVA, PROC Mixed procedure), showing that the CS+/US association has been successfully learned. On day 2, there was a significant decline of pupil size comparing early and late extinction training in each of the three groups. Decline of pupil size was less in the levodopa group compared to the placebo group (block x group interaction: $p = 0.038$). Different to early extinction, there was no difference in pupil size comparing CS+ and CS- in late extinction with no difference between groups. During recall, all groups showed spontaneous recovery. The tiapride group exhibited significantly less decline of pupil size towards the CS+ in late recall compared to early recall than the placebo group (CS x block x group interaction: $p = 0.024$). Thus, the application of an anti-dopaminergic drug prior extinction had on inhibitory effect on the consolidation of extinction memory. Findings further support the hypothesis that the dopamine system is involved in fear extinction learning.

Disclosures: **A. Doubliez:** None. **L. Müntefering:** None. **K. Köster:** None. **E. Nio:** None. **N. Diekmann:** None. **S. Cheng:** None. **C.J. Merz:** None. **A. Thieme:** None. **B. Albayrak:** None. **S.A. Nicksirat:** None. **F. Erdlenbruch:** None. **G. Batsikadze:** None. **T.M. Ernst:** None. **D. Timmann:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.20

Topic: G.01. Fear and Aversive Learning and Memory

Support: The Vulnerable Brain Project vbp.life
James S. McDonnell Foundation
NCATS Grant TL1TR001447

Title: Human threat learning is associated with gut microbiota composition

Authors: *J. P. OYARZUN[1], T. M. KUNTZ[2], Y. STUSSI[1], O. T. KARAMAN[1], S. VRANOS[1], B. L. CALLAGHAN[3], C. HUTTENHOWER[2], J. E. LEDOUX[4], E. A. PHELPS[1];
[1]Harvard Univ., Cambridge, MA; [2]Biostatistics, Harvard T.H. Chan Sch. of Publ. Hlth., Boston, MA; [3]Psychology, UCLA, Los Angeles, CA; [4]Ctr. for Neural Sci., New York Univ., New York, NY

Abstract: Rodent studies have shown that the gut microbiota can influence threat and safety learning, which has been linked to anxiety phenotypes. In humans, it has been demonstrated that microbiota composition varies with anxiety disorders, but evidence showing an association with threat learning is lacking. Here, we tested whether individual variability in threat and safety learning was related to gut microbiota composition in healthy adults. We found that threat but not safety learning varies with individuals' microbiome composition. Our results provide evidence that the gut microbiota is associated with excitatory threat learning across species.

Disclosures: **J.P. Oyarzun:** None. **T.M. Kuntz:** None. **Y. Stussi:** None. **O.T. Karaman:** None. **S. Vranos:** None. **B.L. Callaghan:** None. **C. Huttenhower:** None. **J.E. LeDoux:** None. **E.A. Phelps:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.21

Topic: G.01. Fear and Aversive Learning and Memory

Support: H2020-MSCA-IF-2019

Title: How we fear what we see - MEG study

Authors: *K. J. SWIDER[1], A. W. M. EVERS[2], S. MORATTI[1];
[1]Facultad de Psicología; CIF: Q-2818014-I, Complutense Univ. of Madrid, Madrid, Spain; [2]Fac. of Social and Behavioral Sci., Leiden Univ., Leiden, Netherlands

Abstract: Background: Fear conditioning is recognised as one of the base mechanisms of the maintenance of chronic musculoskeletal pain that is pharmacological difficult to treat. Placebo effect that undoubtedly may influence pain perception can be induced via classical conditioning (CC). Placebo manipulation during fear conditioning may help us understand the neural mechanism of learning and has a great potential for alternative therapies. In the present study, next to placebo effect, we investigate the neural oscillations associated with sensory processing in visual cortex for fear irrelevant and relevant cues.
Methods: Magnetoencephalography (MEG) recording were collected from 30 healthy volunteers (11 male; between 18 and 36 years). The participants were asked to pay attention to the visual stimulus presented on the screen (4 Gabor patches; 15Hz luminance) and to rate, using scale (0-10), their sensation to the electric stimuli (US) delivered to their forearm. The experiment started with habituation and ended with extinction phase, where only Gabor stimuli were presented. During the acquisition, participants from hidden conditioning group (CCH) learned the association between two fear relevant visual stimuli that predicted high-pain (CS+P) and low-pain (CS+M) electric stimuli application; and two fear irrelevant conditions that predicted tactile (CS-T) and no stimuli application (CS-0). In CCH group placebo manipulation was introduced during the testing phase. Without the participants' knowledge, after CS+P and CS+M presentation the same intensity of pain stimuli was delivered. In open conditioning group (CCO), before each experimental phase, the participants were always informed about CS-US contingency. One phase consisted of 10 trials for each CS type.
Results: Preliminary analysis of aversive conditioning process examined stimulus-driven neuromagnetic activity using steady state visual evoked fields (ssVEF). For CCH group we observed increased cortical source space effect (ssVEFs general response: CS+P-CS-0 and CS+M-CS-0 differences) in acquisition compared to both habituation and extinction phase. For CCO analogous effects were observed, however only for CS+M-CS-0 difference. Crucially,

between group comparisons indicate higher ssVEFs general cortical source space effect for CS+M-CS-0 effect for CCH group. There were no differences between acquisition and testing phase for both groups.
Conclusion: We replicated previous findings of increased ssVEFs response in conditioning phase, were the CS gains its relevance. We also indicated that the type of conditioning (open vs hidden) may influence the magnitude of effect.

Disclosures: K.J. Swider: None. **A.W.M. Evers:** None. **S. Moratti:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.22

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIMH Grant R15MH116337

Title: Impact of acute stress, sex, and childhood maltreatment on fear learning and fear generalization in a fear-potentiated startle paradigm

Authors: *K. M. BOAZ[1], C. N. CORDES[1], C. L. PFISTER[1], T. D. NIESE[1], S. L. PARKER[1], K. E. LONG[1], M. L. STANEK[1], B. R. RORABAUGH[2], S. D. NORRHOLM[3], P. R. ZOLADZ[1]; [1]Psychology and Neuroscience, The Sch. of Hlth. and Behavioral Sci., Ohio Northern Univ., Ada, OH; [2]Pharmaceut. Sci., Marshall Univ., Huntington, WV; [3]Psychiatry and Behavioral Neurosciences, Wayne State Univ., Detroit, MI

Abstract: Many researchers approach the etiology of trauma-, stressor-, and anxiety-related mental disorders from the perspective of classical conditioning processes gone awry. According to this view, abnormal associative relationships between conditioned and unconditioned stimuli may underlie pathological anxiety and result in unusually intense fear memories or fear memories that cannot be properly extinguished. Recent work has expanded on this view by showing that many psychological disorders involving pathological anxiety are associated with an exaggerated form of stimulus generalization, leading individuals with such disorders to respond with fear and anxiety to a variety of contexts and cues that should not be threatening. It is well-known that stress, biological sex, childhood maltreatment, and certain dispositional factors can increase one's susceptibility for pathological anxiety and significantly impact fear learning; thus, it is possible that these factors, alone or in combination, contribute to clinical anxiety by influencing fear generalization processes. In the present study, 478 healthy undergraduate students were exposed to the socially-evaluated cold pressor test immediately or 30 min prior to learning to associate one geometrical shape, but not another, with an aversive stimulus in a fear-potentiated startle paradigm. The next day, participants were tested for fear generalization by measuring their fear responses to a variety of stimuli that were similar to, but different from, the shapes observed on Day 1. Objective and subjective measures of stress were collected on Day 1,

and childhood maltreatment was quantified with the Childhood Trauma Questionnaire. The results revealed that, across both stress time points, greater heart rate and greater cortisol levels in response to stress were associated with weaker fear acquisition and a flatter generalization gradient. These effects were influenced by participant sex and trait anxiety. We also found evidence to suggest that greater childhood maltreatment was associated with impaired fear acquisition in males but enhanced fear acquisition in females. These findings reveal a complex interaction between acute stress, biological sex, childhood maltreatment, dispositional anxiety, and fear learning that may lend insight into the etiology of certain stress-related psychological disorders.

Disclosures: **K.M. Boaz:** None. **C.N. Cordes:** None. **C.L. Pfister:** None. **T.D. Niese:** None. **S.L. Parker:** None. **K.E. Long:** None. **M.L. Stanek:** None. **B.R. Rorabaugh:** None. **S.D. Norrholm:** None. **P.R. Zoladz:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NIMH Grant R15MH116337.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.23

Topic: G.01. Fear and Aversive Learning and Memory

Support: ZIAMH002781-15
NCT00018057

Title: Associations among physiology, brain morphometry, and anxiety in human threat learning: divergent evidence from electrodermal and electromyography indices of defensive responding

Authors: *P. NEWSOME, Jr.[1], S. G. RUIZ[2], A. L. GOLD[3], D. S. PINE[1], R. ABEND[1]; [1]Emotion and Develop. Br., NIH, Natl. Inst. of Mental Hlth. (NIMH), Bethesda, MD; [2]Dept. of Psychology, Yale Univ., New Haven, CT; [3]Dept. of Psychiatry and Human Behavior, Warren Alpert Med. Sch. of Brown Univ., Providence, RI

Abstract: Background: Predicting aversive outcomes through associative learning is supported by conserved neural and peripheral mechanisms that govern how we handle threat. While theories in translational and developmental neuroscience link perturbed threat learning processes to pathological anxiety and its treatment, empirical work in anxiety patients yield mixed findings. This may be because different indices capture distinct aspects of conditioned threat anticipation. Here, we compare skin conductance response (SCR), indexing cue-based threat responses, and fear-potentiated startle (FPS), indexing threat-induced state, in capturing anxiety effects on threat learning, across age. We also examine whether subjective fear to conditioned

cues tracks with FPS and SCR. Lastly, we explore associations among these psychophysiological indices and morphometry of conserved subcortical brain structures. **Methods:** Anxiety patients and healthy comparisons (N=306; 178 females; 8-50 years) completed a well-validated Pavlovian threat learning task involving conditioned threat and safety cues. FPS, SCR, and subjective fear to cues indexed differential conditioning and extinction. Of note, novel FPS quantification was employed to overcome limitations with commonly employed approaches. Lastly, we explored associations between psychophysiology and gray matter volume (GMV) in subcortical structures, including amygdala and hippocampus. **Results:** Differential conditioning was observed in patients and comparisons; however, there was comparable differential conditioning between groups for both FPS and SCR (Group x Cue: p=.82, η_p^2<0.01 and p=.16, η_p^2=0.01, respectively). While SCR indicated comparable extinction between groups (p=.29, η_p^2<0.01), FPS revealed attenuated extinction in patients (p=.040, η_p^2=0.01). Regression analyses further confirmed FPS and SCR correlate in conditioning (r=0.18, p=.002), but not extinction (r<0.01, p=.90). Subjective fear of conditioned cues tracked with SCR but not FPS. SCR magnitude diminished with age (p<.001), while FPS indices were age-invariant. While our prior work reported age-moderated links between SCR and hippocampal GMV, such associations were not found for FPS. **Conclusion:** Attenuated extinction learning in anxiety patients was revealed with FPS but not SCR, suggesting abnormally persistent conditioned threat states. FPS did not vary with GMV, indicating SCR and FPS may retain distinct neurobiological functions. These findings provide novel insight on how impaired extinction learning manifests in pathological anxiety and could guide continued research on mechanisms of anxiety and its treatment.

Disclosures: P. Newsome: None. **S.G. Ruiz:** None. **A.L. Gold:** None. **D.S. Pine:** None. **R. Abend:** None.

Poster

555. Fear and Aversive Learning and Memory: Acquisition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 555.24

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01MH114961

Title: The Contributions of Apparatus Size to Novel Object Recognition in Juvenile and Adult Rats

Authors: *L. BARON, L. H. ATCHINSON, A. POULOS;
State Univ. of New York, Albany, Albany, NY

Abstract: Most behavioral testing procedures, like those in novel object recognition, were established mainly for adult male rodents, which can be problematic when attempting to assay behavior in developing rodents. There is significant variability between studies regarding

apparatus size, especially when using these assays in juvenile animals. For example, some studies use the exact apparatus dimensions for juveniles as adults, while others scale apparatus dimensions to those of adult mice. Many behavioral assays rely on the rodent's natural aversion to unfamiliar or unguarded/open spaces. It stands to reason that a larger arena would elicit more anxiety-like responses from a smaller animal. Thus, this experiment aimed to evaluate the validity and necessity of scaling apparatus size for a novel object recognition test (NOR) based on the average weights of adult (Postnatal days [P] 93-94) and juvenile (P24-25) male and female Long-Evans rats. We measured the time each animal spent investigating a novel and a familiar object and determined the percentage of total exploration time spent exploring the novel object. 60% or more exploration time spent at the novel object indicated a novel preference. We hypothesized that adult and juvenile animals would perform similarly in the full (100 x 100 cm) and scaled (51 x 51cm) arenas, respectively. We found an interaction between age and arena-size ($P=0.005$). Sex did not reach significance as a main effect or within an interaction. Within each group, 70% of juvenile animals showed a novel object preference in the scaled arena, while only 40% of subjects exhibited novel object preference in the standard oversized arena. In adult animals, 67% of subjects showed a novel object preference in the larger, traditional-sized arena compared to 55% of subjects in the scaled-down arena. These results confirm that arena size is an important factor that should be standardized and carefully considered when investigating novel object preference learning in juvenile rats. Overall, the best cross-comparative results were observed when arena size was relative to the animal size. This information provides a new lens to view previous studies and sets a standard for future studies using juvenile rats.

Disclosures: L. Baron: None. **L.H. Atchinson:** None. **A. Poulos:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.01

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH R00-MH105549
NIH-R01-MH120136
Rising STARs Award from the University of Texas System
NARSAD Young Investigator Grant

Title: Neural correlates of ultrasonic vocalizations in the prelimbic cortex of rats

Authors: *N. ELINSON-WATSON, G. AQUINO-MIRANDA, F. H. DO MONTE;
Neurobio. and Anat., Univ. of Texas Hlth. Sci. Center, Houston, Houston, TX

Abstract: Rodents emit ultrasonic vocalizations (USVs) to establish communications with their conspecifics. In rats, USVs can be divided into two main types: i) 22 KHz aversive calls that communicate negative emotional states, and ii) 50 KHz appetitive calls that communicate

positive emotional states. It has been shown that the emission of aversive vs. appetitive calls by an emitter rat can bidirectionally regulate the behavioral response of a receiver rat. However, which brain regions encode both types of USVs and how different frequencies of vocalization result in opposite behavioral outcomes remains unknown. One candidate region is the prelimbic subregion of the prefrontal cortex (PL), a structure implicated in the regulation of social behaviors and decision-making processes. To explore whether PL neurons encode USVs of different valences, male Long-Evans rats with single-unit recording electrodes implanted in PL were exposed to pre-recorded aversive and appetitive USV playbacks during the same session. A 22 KHz artificial sound was used as a control stimulus. An offline analysis of 474 neurons recorded from 19 rats revealed two subpopulations of PL cells that responded exclusively to either aversive USVs (14.4%, 7.4% excited and 7% inhibited) or appetitive USVs (11.8%, 4.6% excited and 7.2% inhibited), indicating that PL neurons can discriminate appetitive and aversive USVs. Interestingly, ~65% of PL neurons that changed their firing rates in response to 22 KHz USVs did not respond to 22 KHz artificial sound, suggesting that distinct populations of cells in PL encode social calls vs. ordinary sounds of the same frequency. Next, to check whether aversive and appetitive USV playbacks can affect animal's behavior, a different group of rats previously trained to press a lever for sucrose during the presentation of a light cue was exposed to the USV playbacks (22 KHz or 50 KHz) or the artificial 22 KHz sound either 10 s before or 10 s during the sucrose cue presentation. We found that the presentation of aversive or appetitive USV playbacks did not change cued sucrose-seeking responses. The playback of aversive or appetitive USVs also did not affect cued sucrose-seeking responses in separate groups of rats that had previously observed a conspecific receiving electrical foot shocks or received the foot shocks themselves. Moreover, exposure to USV playbacks did not alter motor activity assessed in an open field arena. Together, our results establish a role for PL neurons in the discrimination of social cues of different valences, and reveals that the communicative function of distinct USVs may require a richer social context to elicit behavioral changes in the receiver animals.

Disclosures: **N. Elinson-Watson:** None. **G. Aquino-Miranda:** None. **F.H. Do Monte:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.02

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH R00-MH105549
NIH-R01-MH120136
NARSAD Young Investigator
University of Texas Rising STARs Award

Title: Conveyance of reward cue information from the prelimbic cortex to the paraventricular nucleus of the thalamus during approach-avoidance conflict

Authors: *G. AQUINO-MIRANDA, V. CHUONG, X. ZHANG, D. S. ENGELKE, F. H. DO MONTE;
Neurobio. and Anat., Univ. of Texas Hlth. Sci. Ctr. At Houston, Houston, TX

Abstract: The paraventricular nucleus of the thalamus (PVT) has been implicated in the regulation of reward-approach vs. threat-avoidance responses during situations of conflict. PVT neurons change their firing rates in response to food cues and these changes are largely suppressed during motivational conflict. However, which inputs convey food cue information to PVT neurons remain unknown. A potential candidate is the prelimbic prefrontal cortex (PL), which responds to food cues and projects densely to the PVT. To explore this question, male adult Long-Evans rats with single-unit recording electrodes implanted in PL were exposed to an approach-food vs. avoid-predator odor conflict model. Rats were initially trained to press a lever for sucrose during the presentation of audiovisual cues. During the test session, animals were exposed to three phases: (i) reward phase, only food cues presented; (ii) cat odor phase, only a fear-inducing cat odor presented, and (iii) conflict phase, food cues concomitantly presented with cat odor. Compared to reward phase, rats displayed increased defensive behaviors and reduced food-seeking responses during conflict. PL recordings (404 neurons from 29 rats) revealed changes in the spontaneous firing rate across the phases with an increase in the percentage of neurons showing inhibition and a reduction in the percentage of neurons showing excitation during conflict. Tracking the activity of the same cells across the phases, we found that most PL neurons changed their firing rates in more than one phase, and ~70% of these cells responded in opposite directions, suggesting valence encoding. After aligning PL activity to the onset of the food cues, we observed a similar proportion of responsive neurons during the reward and conflict phases (~20%). Surprisingly, only a small fraction of these cells responded in both phases (~7%; Fisher Exact Test, $p < 0.05$), suggesting that distinct subpopulations of food cue responsive neurons are recruited during conflict. Using a combination of recordings and optogenetics for the photoidentification of PL neurons that project to PVT (PL^{PVT}, n = 63 neurons), we found that the spontaneous activity of PL^{PVT} neurons resembled those of the entire PL population across the phases. However, in contrast to non-identified PL neurons, the number and the magnitude of excitatory food cue responses in PL^{PVT} neurons were the same between the reward and conflict phases. Together, our results demonstrate that activity in PL neurons correlates with changes in behavioral decision during motivational conflict and suggest that PL^{PVT} neurons convey food cue information to PVT independently of the defensive state of the animals.

Disclosures: G. Aquino-Miranda: None. **V. Chuong:** None. **X. Zhang:** None. **D.S. Engelke:** None. **F.H. Do Monte:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.03

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH Grant 738 R01-NS102490
NIH Grant R00-MH105549
NIH Grant R01-MH120136
Brain & Behavior Research Foundation Grant (NARSAD Young Investigator)
Rising STARs Award from UT System

Title: Enhancing associative learning in rats with a computationally designed training protocol

Authors: *C. E. CHO, X. O. ZHANG, Y. ZHANG, D. S. ENGELKE, P. SMOLEN, J. H. BYRNE, F. H. DO MONTE;
Neurobio. and Anat., The Univ. of Texas Hlth. Sci. Ctr. at Houston, Houston, TX

Abstract: Associative learning requires the activation of protein kinases with distinct temporal dynamics. Learning protocols with computationally designed intertrial intervals (ITIs) that maximize the interaction between fast-activated protein kinase A (PKA) and slow-activated extracellular signal-regulated kinase (ERK) enhance nonassociative learning in *Aplysia*. Here, we examined whether a computationally designed protocol based on PKA and ERK dynamics in rat hippocampus would enhance associative learning in mammals. We simulated ~1000 training protocols with varying ITIs and identified an optimal protocol predicted to induce stronger learning than those with fixed ITIs. Male adult Long-Evans rats were exposed to an auditory fear conditioning paradigm where conditioned stimuli (CS, 3 kHz tone, 30 s) co-terminated with unconditioned stimuli (US, footshock, 0.7 mA, 0.5 s). Rats in the full conditioning (FC, n = 14) and partial conditioning (PC, n = 12) groups received 8 or 4 CS-US pairings with fixed ITIs of 4.5 min respectively, whereas rats in the optimal partial conditioning (OPC, n = 14) group received 4 CS-US pairings with ITIs of 8, 8 and 16 min. The next day, FC and OPC groups exhibited reduced locomotion speed compared to the PC group, suggesting stronger fear memory retrieval. FC and OPC groups also showed impaired extinction learning characterized by sustained freezing compared to the PC group. In a separate experiment, we compared the OPC protocol with an equally spaced partial conditioning (SPC, n = 10) protocol consisting of 4 CS-US pairings with ITIs of 11 min and 10 s. Rats in the OPC group showed stronger fear acquisition and retrieval, suggesting that the memory facilitating effects of the optimal protocol are specific for the irregular intervals and not the training duration. Next, we examined whether the optimized ITIs would enhance fear extinction. Conditioned suppression of reward-seeking behavior was used as an additional measure of fear memory as it is more sensitive than freezing during extinction. Fear conditioned rats were assigned to three groups: full extinction (FE, n = 12) and partial extinction (PE, n = 11) groups received 12 and 4 CS trials with 2.5 min ITIs respectively, whereas the optimal partial extinction (OPE, n = 13) group received 4 CS trials with ITIs of 8, 8 and 16 min. Compared to FE and PE groups, the OPE group showed increased lever pressing during the pre-CS periods in a spontaneous recovery test, suggesting enhanced extinction of contextual fear memory. Together, our findings demonstrate enhanced associative learning in mammals with a behavioral protocol designed using a computational model of memory-related signaling pathways.

Disclosures: C.E. Cho: None. **X.O. Zhang:** None. **Y. Zhang:** None. **D.S. Engelke:** None. **P. Smolen:** None. **J.H. Byrne:** None. **F.H. Do Monte:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.04

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH R00-MH105549
NIH-R01-MH120136
Rising STARs Award from the University of Texas System
NARSAD Young Investigator Grant

Title: Neural signatures of risk-taking vs. risk-avoiding behaviors in the prefrontal cortex of morphine-conditioned rats

Authors: *C. B. QUAVE[1], A. M. VASQUEZ[1,2], E. P. BORA[1,3], C. L. CHIDOMERE[1,4], G. AQUINO-MIRANDA[1], D. S. ENGELKE[1], F. H. DO MONTE[1];
[1]Dept. of Neurobio. & Anat., The Univ. of Texas Hlth. Sci. Ctr. at Houston, Houston, TX; [2]Dept. of Neurosci., Rice Univ., Houston, TX; [3]The Univ. of Texas at Austin, Austin, TX; [4]Dept. of Biomed. Engin., Univ. of Houston, Houston, TX

Abstract: Opioid use disorder is associated with impaired risk-related decision-making. However, it is unclear how repeated opioid exposure affects the brain to alter risk taking under motivational conflict. In a modified conditioned place preference protocol, rats injected with either saline or morphine were exposed to the side of the apparatus preferred least at baseline. Conditioning occurred over 10 alternating days (5 pairings in each side). Two days after conditioning, rats underwent a preference test immediately followed by a conflict test in which an aversive stimulus (cat saliva) was introduced in the side of the chamber previously paired with morphine injections. In the preference test, morphine-treated rats spent more time in the drug-paired side of the apparatus than did saline-treated rats. In the conflict test, saline group rats avoided the side of the apparatus containing cat odor. In contrast, rats in the morphine group continued to prefer the previously drug-paired side despite the presence of cat odor, demonstrating increased risk-taking behavior. K-means clustering uncovered two subsets of morphine-treated rats that exhibited either: *i)* enhanced place preference and persistent drug seeking during conflict (*risk-takers, RT*), or *ii)* moderate place preference and suppressed drug seeking during conflict (*risk-avoiders, RA*). Single-unit recordings from neurons in the prelimbic (PL) cortex, a region involved in decision-making and strategy shifting, revealed decreased firing rates upon acute morphine exposure. In contrast, on the final drug conditioning day, morphine failed to suppress neuronal firing rates, suggesting that PL neurons undergo adaptation to repeated morphine exposure. Recordings during the conflict test identified distinct populations of PL neurons that were either excited or inhibited when rats entered the side of the apparatus that contained cat odor. Interestingly, RT rats showed a lower proportion of neurons excited during paired side entries as compared to saline-treated or RA rats, suggesting an attenuated neuronal response to threat in the drug-paired context. Additionally, while cells inhibited during paired side entries were unaffected during neutral side entries in saline-treated and RA rats, this discrimination in PL firing rates across sides did not occur in RT rats. Taken together, our results

suggest that a loss of PL inhibition after opioid conditioning is associated with the formation of contextual reward memory. Furthermore, suppression of excitatory responses to threat and impaired inhibitory signaling of neutral vs. threat contexts in PL may underlie increased risk taking following opioid exposure.

Disclosures: C.B. Quave: None. **A.M. Vasquez:** None. **E.P. Bora:** None. **C.L. Chidomere:** None. **G. Aquino-Miranda:** None. **D.S. Engelke:** None. **F.H. Do Monte:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.05

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH grant R00-MH105549
R01-MH120136
Rising STARs Award from UT System
IBRO Collaborative Research Grant
NARSAD Young Investigator

Title: Prelimbic cortex neurons encode changes in cued food-seeking behavior under distinct internal states

Authors: *X. O. ZHANG, C. E. CHO, G. AQUINO-MIRANDA, N. ELINSON-WATSON, F. H. DO MONTE;
Dept. of Neurobio. and Anat., Univ. of Texas Hlth. Sci. Ctr. at Houston, Houston, TX

Abstract: Flexibly adjusting foraging behavior based on internal metabolic needs and environmental threats is crucial for animal survival. However, the neural mechanisms underlying the transition in cued food-seeking behaviors under distinct metabolic and threat states remain unclear. Neurons in the prelimbic (PL) subregion of the medial prefrontal cortex change their activity in response to food-associated cues. To test if PL responses to food cues vary according to animals' metabolic states (hungry vs. satiated) or threat states (safe vs. threatened), we used a miniature fluorescent microscope to record calcium transients in freely behaving rats (n = 548 neurons, 6 rats). Adult male Long-Evans rats were initially trained to press a lever for sucrose upon the presentation of an audiovisual cue. During the metabolic state test, rats were presented with 12 food cues followed by a 50 min sucrose *ad libitum* period to induce satiation, and subsequently exposed to 12 additional food cues. During the threat state test, rats were presented with12 food cues in a safe arena followed by a 10 min period of predator odor (cat saliva) exposure to induce fear, and subsequently presented with 12 additional food cues. Rats showed a reduction in food seeking from hungry to satiated states, as well as from safe to threatened states. After aligning the calcium transients of all PL neurons to the onset of the food cues, we observed no differences in the averaged PL activity under hungry vs. satiated states but a significant

increase from safe to threatened states, suggesting that PL neurons are preferentially recruited during more salient internal states. In contrast, the number of food-cue responsive neurons and the magnitude of their responses remained the same across the distinct states. Interestingly, tracking the activity of the same cells across the session revealed that different PL neurons respond to food cues during distinct internal states. PL neurons showing excitatory or inhibitory responses to food cues when rats were in the hungry or safe states exhibited an overall reduction in their responses during the satiated or threatened states. In parallel, when rats transitioned to satiated or threatened states, separate groups of non-responsive cells emerged to display either excitatory or inhibitory responses to food cues. Taken together, our results suggest that the recruitment of distinct PL neuron subpopulations when animals transition between metabolic or threat states may serve to adjust behavioral responses to environmental food cues. Our findings may help to understand maladaptive food-seeking behavior in patients with eating- and anxiety-related disorders.

Disclosures: X.O. Zhang: None. **C.E. Cho:** None. **G. Aquino-Miranda:** None. **N. Elinson-Watson:** None. **F.H. Do Monte:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.06

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIMH T32MH073526
NIMH K01MH116264
Brain and Behavior Foundation
Whitehall Foundation
Brain Research Foundation
UCLA DGSOM
Klingenstein-Simons Foundation

Title: Neural signatures of amnesic and persistent memories

Authors: *B. JIN[1], L. HOLDEN-WINGATE[2], L. OHANIAN[2], B. LE[3], L. DENARDO[4]; [1]Molecular, Cell. and Integrative Physiol., [2]Integrative Biol. and Physiol., [3]Psychobiology, [4]Physiol., UCLA, Los Angeles, CA

Abstract: Episodic memories formed early in life are rapidly forgotten, while those made later in life can last a lifetime. The organization of memory circuits is well studied in the adult brain, and fearful memories are stored across a network of cortical and limbic structures. However, how memories are represented in the developing brain and whether there exists a neural signature of fleeting memory remain poorly understand. To address these questions, we used TRAP2 (Targeted Recombination in Active Populations) mice in combination with brain

clearing and light sheet fluorescence microscopy to visualize the brain-wide activated neurons during fear memory retrieval in mice at different stages of development. We quantified neuronal populations activated by recent (1 day) fear memory retrieval at key developmental time points, including infant (P17), pre-adolescent (P25), and young adult (P60) mice, which are characterized by amnesic and lasting memories respectively. Across all ages, the sensory cortices expressed more TRAPed cells in fear-conditioned mice than non-shock controls, but did not significantly differ by age among condition-matched groups. However, network analyses revealed age-specific neural signatures of memory. To infer changes in functional connectivity, we examined correlations between numbers of TRAPed cells across brain regions. Pre-adolescent brains were marked by decreased co-activation among cortical and thalamic regions, while adult brains displayed increased co-activation of cortical and subcortical areas, including between retrohippocampal cortical areas and the hippocampus. To further understand how activity in these regions related to memory retrieval, we examined correlations between TRAP patterns and behavioral characteristics of individual animals. Areas with high correlations with freezing measures changed with age. In P17 mice, freezing behavior was correlated with numbers of TRAPed cells in amygdalar areas. On the other hand, freezing was correlated with TRAPed cell numbers across several cortical association areas in adults. Interestingly, freezing in pre-adolescent mice was correlated to regions that were associated with non-freezing behaviors in adult mice. This age-dependent shift coincides with the transition from shorter-lasting memories in infancy to longer-lasting memories in pre-adolescent and adult mice, highlighting potential mechanisms that underlie the emergence of persistent fearful memories across development.

Disclosures: B. Jin: None. **L. Holden-Wingate:** None. **L. Ohanian:** None. **B. Le:** None. **L. DeNardo:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.07

Topic: G.01. Fear and Aversive Learning and Memory

Support: Whitehall Foundation Research Grant (LAD)
Klingenstein-Simons Fellowship in Neuroscience (LAD)
NIH K01MH116264 (LAD)
NIH K08MH116125 (SAW)

Title: Development of a Preclinical Rodent Model of Transcranial Magnetic Stimulation

Authors: *A. S. ENOS[1], M. W. GONGWER[1], A. Q. KASHAY[2], J. RILEY[1], A. QI[2], C. M. GOODPASTER[1], L. A. DENARDO[1], S. A. WILKE[2];
[1]Physiol., [2]Psychiatry, Univ. of California Los Angeles, Los Angeles, CA

Abstract: Repetitive Transcranial Magnetic Stimulation (rTMS) is an FDA approved therapy for patients with treatment-resistant depression and obsessive-compulsive disorder and also shows promise for numerous other psychiatric and neurological disorders. During clinical rTMS treatment, focal electromagnetic stimulation is thought to exert therapeutic effects by driving plasticity in neural circuits. Despite its common use, clinical effects are variable and the underlying neural circuit changes induced by rTMS treatment are poorly understood, limiting the rational design of more precise and effective treatment protocols. Much of what we know about the mechanisms of rTMS stems from studies of the human motor system, but prefrontal cortex (PFC) is the primary clinical target. Progress has been hindered by lack of a preclinical rodent model with strong face validity for how clinical rTMS is delivered. Scaling TMS coils to the rodent brain has resulted in focal stimulation that is too weak to elicit action potentials. Instead, larger coils are often used that stimulate the entire rodent brain. To overcome these hurdles, we are developing a new preclinical rodent model that will allow us to study circuit and behavioral differences in mice following chronic rTMS targeting the medial prefrontal cortex (mPFC). We obtained a novel rodent TMS electromagnetic coil that has comparable power and precision to human stimulators. To enable delivery of chronic rTMS that mimics human protocols, we developed a way to prevent the magnet from overheating during pulse sequences. We generated a liquid cooling system that pumps a supercooled solution of 30% calcium chloride through a copper coil encircling the magnet. With this system, we can perform several 30-minute rTMS sessions in a day, allowing us to study chronic effects of rTMS on neural circuits and behavior in cohorts of mice. We delivered 5-day rTMS or sham treatment on chronically stressed mice, followed immediately by behavioral testing and perfusion for histological analyses. We identify behavioral changes in rodent anxiety assays as well as changes in the expression of histological markers of neural activity. Our findings provide the foundation for further studies probing long-term molecular and synaptic changes induced by chronic rTMS targeting PFC. This mechanistic understanding will be critical as we optimize this noninvasive therapy for treatment of both depression and other neurological and psychiatric disorders.

Disclosures: **A.S. Enos:** None. **M.W. Gongwer:** None. **A.Q. Kashay:** None. **J. Riley:** None. **A. Qi:** None. **C.M. Goodpaster:** None. **L.A. DeNardo:** None. **S.A. Wilke:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.08

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH K01MH116264
Whitehall Foundation Research Grant
Klingenstein-Simons Foundation Grant

Title: Prefrontal control of remote fear memory via temporal association cortex

Authors: *Z. ZEIDLER[1], M. SEONG[2], M. SHARI[2], L. A. DENARDO[1];
[1]Physiol., [2]UCLA, Los Angeles, CA

Abstract: Memories of an emotional experience can last a lifetime, yet the same memory can involve new neurons and circuits across time. Growing evidence indicates that as memories age, they are reorganized into a distributed cortical network. While several independent cortical regions participate in remote memory, our understanding of their interactions at the cellular level are lacking. Time-dependent reorganization of memory has clinical implications. Remote memories of trauma are more treatment resistant than recent memories, linking memory evolution with memory resilience. In an auditory fear memory paradigm, we investigated how the prelimbic (PL) region of the prefrontal cortex interacts with the temporal association area (TeA), a cortical region involved in processing the salience of auditory information. To test whether PL-TeA projections have a unique role in remote, but not recent, memory, we optogenetically silenced PL-to-TeA axons during recent or remote memory. Inhibition of PL-TeA projections caused a selective decrease in memory strength during remote memory recall, without affecting recent memory recall, indicating that the role of the PL-TeA memory retrieval emerges over time. In ongoing experiments, we are examining how memories are encoded in PL-TeA circuits across time. Using adult mice, we infected PL neurons with GCaMP8m and used fiber photometry to measure activity arising from their axons in TeA during auditory fear conditioning, recent memory, and remote memory recall. We investigated task-related signals across memory timepoints, exploring differences between recent and remote recall. We next used freely-moving 1-photon calcium imaging to monitor either PL neurons, or using an intersectional retroviral-Cre based approach, PL-TeA projection neurons, during the same timepoints. We identified task and behavior encoding in both populations, comparing overall PL activity to specific PL-TeA activity at recent and remote memory timepoints. Overall, these results identify a novel circuit in remote fear memory and suggest principles of memory organization across time.

Disclosures: Z. Zeidler: None. **M. Seong:** None. **M. Shari:** None. **L.A. DeNardo:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.09

Topic: G.01. Fear and Aversive Learning and Memory

Support: NSERC Postgraduate Doctoral Award

Title: Prefrontal cortex circuit maturation and its role in adaptive threat avoidance in mice

Authors: *C. B. KLUNE[1], C. M. GOODPASTER[1], R. CHEN[1], N. S. JONES[1], L. A. DENARDO[2];
[2]Physiol., [1]UCLA, Los Angeles, CA

Abstract: The medial prefrontal cortex (mPFC) acts through its connections with the nucleus accumbens (NAc) and basolateral amygdala (BLA) to promote adaptive threat responses. To establish these mature cognitive functions, mPFC circuits undergo a prolonged development during which experience may feedback on the brain to promote adaptive responding. However, how the behavioral functions of mPFC circuits mature across development is poorly understood. Our study investigates the maturation of learned threat avoidance in mice and the contributions of mPFC-BLA and mPFC-NAc projections over development. Using platform-mediated avoidance (PMA), in which a fear-conditioned tone prompts rodents to navigate to a safety platform, we found that juvenile (postnatal day (P)23), adolescent (P35) and adult (P60+) mice were all able to learn PMA in a single training session. However, during a threat memory retrieval session the next day, adult mice showed robust avoidance memory, but P35 mice spent significantly less time on the platform and P23 mice displayed a rapid extinction response. To identify age-dependent differences in the underlying circuit function, we utilized optogenetic methods to manipulate mPFC-NAc and mPFC-BLA projections during the PMA retrieval session in mice trained at P23, P35, and P60+. In adults, excitation of mPFC-BLA neurons augmented avoidance while excitation of mPFC-NAc neurons reduced it. In contrast, at P23, excitation of mPFC-BLA neurons reduced avoidance. This suggests that mPFC-BLA neurons undergo circuit changes in early life that shape how they modulate avoidance behavior, potentially indicating a critical period in which disruption to circuit maturation may lead to maladaptive avoidance later on. At P35, inhibition of mPFC-NAc neurons increased avoidance, suggesting that mPFC-NAc neurons may be more active in adolescence, contributing to the lower levels of avoidance displayed by P35 mice. Together these findings indicate that the maturation of mPFC-BLA and mPFC-NAc circuits contribute to developmental changes in learned threat avoidance. Understanding how mPFC circuits develop in the typical brain will build an important foundation to understand how perturbed development may contribute to disease states.

Disclosures: C.B. Klune: None. **C.M. Goodpaster:** None. **R. Chen:** None. **N.S. Jones:** None. **L.A. DeNardo:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.10

Topic: G.01. Fear and Aversive Learning and Memory

Title: Prefrontal cortical circuits coordinate observational fear learning

Authors: *S. E. SILVERSTEIN, O. BUKALO, J. SCHAFFER, R. O'SULLIVAN, A. HOLMES;
NIAAA, NIH, Bethesda, MD

Abstract: Learning about threats from observing others has significant adaptive value. Specifically, observational fear learning (OFL) illustrates how social organisms learn from environmental interactions to promote safety and avoid harm. However, excessive responding to observed threats can interfere with essential functions, potentially leading to trauma-related disorders. Canonical 'reactive' defensive circuits, which generate rapid responses to imminent danger, are complemented by higher-order cognitive systems recruited when threats are more distal or ambiguous, yet the neural circuits underlying OFL are inadequately defined. To address this, we vicariously conditioned mice to an auditory cue by watching a demonstrator undergo cue-shock pairings. Using a combination of anatomical tracing, immunohistochemistry, and *in vivo* optogenetics and calcium imaging, we found that dorsomedial prefrontal cortex (dmPFC) inhibition disrupted OFL formation ($p<0.05$). Given the complexity of OFL, which requires simultaneous integration of social, contextual, and cued information, we tested how dmPFC might modulate OFL by coordinating diverse upstream activity. We found that input to the dmPFC from basolateral amygdala (BLA) and ventral hippocampus (vHPC), respectively, promote and constrain observer freezing ($p<0.05$). Cellular-resolution neuronal recordings demonstrated dmPFC neurons separately code observed and directly experienced threat, as well as opposing (active/passive) defensive behavior states. The capacity of dmPFC neurons to code for multiple aspects of OFL could partly reflect differential functional contributions of dmPFC projection populations. To address this question, we examined the role of dmPFC projections to PAG. We found that dmPFC projections to midbrain periaqueductal gray (PAG) signal the dynamic transfer of threat information from demonstrator-to-observer to limit observer freezing. Together, our findings reveal how dmPFC coordinates long-range inputs and outputs to calibrate behavioral responses to observed threat. Moreover, this suggests that maladaptive responses to socially learned threat could arise from deficits in dmPFC and its interacting circuits and explain why neuropsychiatric disorders as diverse as psychopathy and PTSD are linked to dmPFC damage.

Disclosures: **S.E. Silverstein:** None. **O. Bukalo:** None. **J. Schaffer:** None. **R. O'Sullivan:** None. **A. Holmes:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.11

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIMH K01MH116264
NSF GRFP

Title: Early life adversity disrupts medial prefrontal cortex dependent approach-avoidance behavior in adolescents

Authors: *C. M. GOODPASTER, N. S. JONES, L. A. DENARDO;
Physiol., UCLA, Los Angeles, CA

Abstract: The medial prefrontal cortex (mPFC) plays a key role in evaluating and executing adaptive responses to threats. In order to establish mature cognitive abilities, the mPFC undergoes prolonged maturation. Adolescence marks the opening of a sensitive period that, paired with increased exploration, may facilitate the development of nuanced responses to perceived threats. Unfortunately, this also opens a long window during which insults can perturb healthy mPFC development. Early life adversity (ELA), such as neglect or maltreatment in childhood, is known to cause lasting structural and functional alterations in mPFC and its target regions, including the basolateral amygdala (BLA). These changes impact emotional and cognitive processing which can lead to psychiatric illnesses, such as anxiety and depression. Exaggerated, maladaptive responses to perceived threat, usually at the expense of rewarding experiences, are hallmarks of these conditions. Symptoms often manifest during adolescence, an important period in which rewarding exploratory interactions with the environment have significant influence over brain development and the refinement of adaptive behavioral strategies. Yet, it is not known how ELA alters the neural circuitry underlying threat response during development. To fill this knowledge gap, I employ a limited nesting and bedding (LBN) model of ELA in mice that simulates aspects of resource scarcity and altered parental care that contribute to ELA in humans. To assess differences in threat encoding and retrieval I utilize a platform-mediated avoidance (PMA) task, in which a fear conditioned tone prompts mice to navigate to a safety platform while competing with an innate motivation to explore a novel object. LBN accelerates learning of the PMA task and leads to the excessive avoidance during retrieval in comparison to standard reared mice, measured by increased time spent on the platform. Using fiber photometry, I will assess how mPFC to BLA connections drive adolescent behavior in PMA and how it becomes disrupted following ELA. Thus far, most ELA research has largely focused on behaviors and cellular-level outcomes in adults; here we are uncovering new insights into how early experiences impact developmental circuit dynamics that are necessary for refined behaviors.

Disclosures: C.M. Goodpaster: None. **N.S. Jones:** None. **L.A. DeNardo:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.12

Title: WITHDRAWN

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.13

Topic: G.01. Fear and Aversive Learning and Memory

Support: National Institutes of Health (MH106435)
National Institutes of Health (MH045573)

Title: Caudal area 47/12: a cross-species link of persistent avoidance circuits

Authors: *L. R. TRAMBAIOLLI[1], F. J. MARTÍNEZ-RIVERA[2], G. J. QUIRK[3], S. N. HABER[4];
[1]McLean Hosp. - Harvard Med. Sch., Somerville, MA; [2]Icahn Sch. of Med. at Mount Sinai, New York, NY; [3]Univ. of the Philippines, Manila, Philippines; [4]Pharmacol. & Physiol., Univ. of Rochester, Rochester, NY

Abstract: The caudal area 47/12 of the primate brain has been recently described as a node of the salience network in humans and nonhuman primates (NHP) (Trambaiolli et al. 2022). This region supports stimulus-outcome predictions associated with salient stimuli and prepares appropriate responses later selected by the anterior cingulate cortex (ACC). Abnormal computations in this region could lead to pathological behaviors, such as the persistent avoidance observed in patients with obsessive-compulsive disorder (OCD).
In rodents, a circuit including the agranular insular/lateral orbital (AI/LO) cortex, rostral prelimbic (rPL) cortex, ventral striatum (VS), and basolateral amygdala (BLA) is suggested to control persistent avoidance (Martínez-Rivera et al. 2022). The AI/LO has excitatory inputs to rPL neurons that project to the VS to control avoidance expression. However, hypoactivity in AI/LO decreases excitatory drive from rPL to the VS, which makes VS more susceptible to BLA excitatory inputs which drive avoidance expression.
Herein, we hypothesize that the rodent AI/LO is homologous to the caudal 47/12 and that the same persistence avoidance circuitry can be tracked in NHP. To evaluate this, we used seven anterograde injections in healthy macaque monkeys: two in caudal 47/12, three in area 24, and two in the BLA. Under dark-field microscopy, we identified that caudal 47/12 projected densely to the rostral area 24 (ACC), which bear circuit-level homology with rostral PL in rats (Sharma et al. 2020). The three injections in rostral 24 were placed at different rostrocaudal levels (pregenueal to postgenual), respectively. While pregenual 24 projected densely to the VS and the BLA, weaker projections were observed after postgenual injections. Dense axonal labeling from the BLA and pregenual 24 overlap in the VS. BLA projections to caudal 47/12 were also observed.
Altogether, these findings suggest that the persistent avoidance circuitry identified in rodents can be translated to NHP. Importantly, the NHP caudal 47/12 may correspond to this circuit's AI/LO node, receiving dense connections from BLA. These bottom-up projections may cause biased attention to negative stimuli and impaired response preparation. This connection could also be associated with the hypoactivity in AI/LO observed in avoidant rodents and caudal 47/12 in OCD patients. In fact, recent studies report that OCD patients have amygdala hyperactivation during symptom provocation. Deficits in the devaluation of negative stimuli and hypoactivation of vlPFC (including area 47/12) are also observed in patients undergoing an avoidance devaluation task (Chase et al. 2020).

Disclosures: **L.R. Trambaiolli:** None. **F.J. Martínez-Rivera:** None. **G.J. Quirk:** None. **S.N. Haber:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.14

Topic: G.01. Fear and Aversive Learning and Memory

Support: JPMJSP2119

Title: Inhibition of neurons by the hM4Di DREADD in the bed nucleus of the stria terminalis increases disgust and anxiety in the retrieval of conditioned taste aversion.

Authors: *E. KIKUCHI[1,2], T. INUI[1], S. SU[1], M. FUNAHASHI[1];
[1]Oral Physiol., Grad Sch. Dent. Med., [2]Orthodont., Grad. Sch. Dent. Med., Hokkaido Univ., Sapporo, Japan

Abstract: Conditioned taste aversion (CTA) is produced by pairing a taste as a conditioned stimulus (CS) with visceral malaise as an unconditioned stimulus (US). Conditioned animals show disgust and anxiety to the CS in the retrieval of CTA. The bed nucleus of the stria terminalis (BNST) plays a critical role in anxiety and fear states. However, no reports showed the involvement of the BNST in CTA. This study examined the effects of chemogenetic inhibition of the BNST neurons on CS consumption behaviors following the establishment of CTA. Male C57/BL6 mice (n = 19) were injected with AAV8-hSyn-hM4Di-mCherry (0.5 µl/side) into the BNST. They were conditioned by pairing 0.2% saccharin solution (CS) with malaise-inducing lithium chloride in a behavioral analysis apparatus. The mice were again exposed to the CS in Tests 1-3. Two groups were formed from mice with similar saccharin intakes in Conditioning and Test 1. In Test 2, half of the mice (n = 10) were given deschloroclozapine (DCZ; 0.05 mg/kg, i.p., a hM4Di ligand) and were exposed to the CS 30 min after injection. Another half of the mice (n = 9) received a vehicle (1% DMSO in saline, i.p.). The mice were exposed to the CS 48 hours after DCZ or vehicle injection in Test 3. CS consumption was drastically reduced in the DCZ-injection group (0.09 ± 0.03 ml) compared to the vehicle-injection group (0.27 ± 0.04 ml) (unpaired t-test; $p < 0.01$). The analysis of lick microstructure showed that the size of burst in the DCZ group (5.35 ± 1.78 licks) was significantly smaller than that in the vehicle group (11.75 ± 2.1 licks) (unpaired t-test; $p < 0.01$). Approach behaviors were categorized into Entry-Lick (mice approached the spout containing the CS and licked it) and Entry-Stop (mice approached the spout but not licked it). The probability of Entry-Stop (the number of Entry-Stop / the number of all approaches) was calculated. The probability of Entry-Stop in the DCZ group (61.0 % ± 8.2) was significantly higher than that in the vehicle group (34.2 % ± 7.2) (unpaired t-test; $p < 0.05$). The burst size and probability of Entry-Stop are indices for taste palatability and anxiety, respectively. Therefore, the smaller burst size and more frequent Entry-Stop in the DCZ group indicate the enhancement of the

disgust and anxiety for the CS. We concluded that the BNST neurons play an important role in the retrieval of CTA.

Disclosures: E. Kikuchi: None. **T. Inui:** None. **S. Su:** None. **M. Funahashi:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.15

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIMH Grant #1R01MH117421-01A1
Whitehall Foundation
Charles Hood Foundation
Tommy Fuss Center for Neuropsychiatric Disease Research
Sleep Research Society grant NS103161

Title: Lateral septum modulates cortical state to tune responsivity to threat stimuli

Authors: S. BRITO[1], M. HASHIMOTO[1], A. VENNER[2], A. PASQUALINI[1], T. YANG[1], P. M. FULLER[2], ***T. ANTHONY**[3];
[1]Boston Children's Hosp., Boston, MA; [2]Beth Israel Deaconness Med. Ctr., Boston, MA; [3]Boston Children's Hospital, Harvard Med. Sch., Boston, MA

Abstract: Sudden unexpected environmental changes capture attention and when perceived as potentially dangerous, evoke defensive behavioral states. Perturbations of the lateral septum (LS) can produce extreme hyperdefensiveness even to innocuous stimuli, but how this structure influences stimulus-evoked defensive responses and threat perception has not been defined. Here, we show that *Crhr2*-expressing neurons in mouse LS are phasically activated upon detection of threatening but not rewarding stimuli. Threat stimulus-driven activity predicts the probability but not vigor or type of defensive behavior evoked. Although necessary for and sufficient to potentiate stimulus-triggered defensive responses, LS^{Crhr2} neurons do not promote specific behaviors. Rather, their stimulation elicits negative valence and physiological arousal. Moreover, LS^{Crhr2} activity tracks brain state fluctuations, and drives cortical activation and rapid awakening in the absence of threat. Together, our findings suggest that LS directs bottom-up modulation of cortical function to evoke preparatory defensive internal states and selectively enhance responsivity to threat-related stimuli.

Disclosures: S. Brito: None. **M. Hashimoto:** None. **A. Venner:** None. **A. Pasqualini:** None. **T. Yang:** None. **P.M. Fuller:** None. **T. Anthony:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.16

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH Grant R00DA044331

Title: The role of central amygdala D2-expressing neurons in punishment learning

Authors: *J. R. WATSON[1], P. VENTO[2];
[1]Psychology, Univ. of South California, Columbia, SC; [2]Psychology, Univ. of South Carolina, Columbia, SC

Abstract: The central nucleus of the amygdala (CeA) plays a critical role in regulating autonomic and behavioral responses to aversive stimuli and has been implicated in psychiatric disorders such as addiction, post-traumatic stress disorder (PTSD), and anxiety disorders. However, little is known about how this region encodes punishment and what mechanisms within this region drive punishment learning. Dopamine D2 receptor-expressing neurons play a key role in encoding aversive experiences and a subpopulation of CeA neurons express D2 receptors. Here, we investigate the effect of bilateral lesions of D2 neurons in the CeA on punished responding for food in a novel two-lever discrimination task where rats choose between a large (3 pellet) punished food reward vs a small (1 pellet) safer reward option that was never accompanied by punishment (footshock). Specifically, transgenic rats expressing cre-recombinase under control of the promotor for the dopamine D2 receptor received bilateral stereotaxic injections of a cre-dependent inducer of apoptosis, taCasp3 (pAAV5-flex-taCasp3-TEVp), or control virus (pAAV2-hSyn-DIO-mCherry). After recovery from surgery, rats were restricted to 85% of their free-fed bodyweight and trained to discriminate between two levers that yielded either the small or large food reward in response to a single lever press (FR1). Test sessions consisted of 60 trials presented over two distinct phases in which rats were randomly presented one lever (either small or large reward) over 30 “forced” trials (Phase 1). This was followed by an additional 30 “choice” trials (Phase 2) in which both the large and small reward lever options were presented simultaneously; however, pressing for the large reward was immediately followed by a brief mild footshock. Responses on the small reward lever were never associated with punishment, thereby making this reward choice the “safer” of the two options. Shock intensity increased by 30% each test session until rats’ preference shifted to the small reward option on ≥85% of trials, referred to as “shock switchpoint”. Rats with lesions of CeA D2 neurons displayed significantly lower shock switchpoints and earlier transitioning to the safe, small reward lever compared to controls. Notably, CeA D2-lesioned rats displayed similar preference for the large reward prior to shock testing, indicating the effect on shock switchpoint was unlikely the result of a preexisting bias or non-specific learning impairment. Together, these results suggest that CeA D2 neurons serve an essential role in learning to suppress reward-seeking in the presence of punishment.

Disclosures: J.R. Watson: None. **P. Vento:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.17

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH R01MH112355
BRAIN Initiative 1U01 MH10913301
NIH P30DA048736
R01 DA033396-04S1
Inscopix DECODE Award

Title: Decoding Noradrenergic Circuitry in Aversive Contextual Processing

Authors: *E. T. ZHANG[1], G. S. SAGLIMBENI[1], S. C. PIANTADOSI[1], J. FENG[2], Y. LI[2], M. R. BRUCHAS[1];
[1]Univ. of Washington, Seattle, WA; [2]Sch. of Life Sci., Peking Univ., Beijing, China

Abstract: The dentate gyrus (DG), a hippocampal subregion, has been heavily implicated in contextual memory and pattern separation. The DG plays a critical role in combining existing episodic information with new contextual information, and deficits in this process are thought to underlie multiple disease states including post-traumatic stress disorder (PTSD). The DG receives dense projections from the locus coeruleus (LC), a region in the brainstem that provides the primary source of norepinephrine (NE) in the brain and plays a key role in the regulation of anxiety and arousal states. In response to stressful situations, LC-NE activity increases, leading to modulation of many different structures across the brain, including the DG. Our prior studies have implicated LC-DG noradrenergic projections in modulating aversive contextual processing; however, the exact mechanisms by which this occurs remain largely unknown. ***Hence, we hypothesize that increased NE release from LC to DG α- and β-adrenergic receptors (ARs) due to aversive stimuli causes contextual generalization via increased neuron excitability and regulation of neural ensembles in the DG.*** Here, using a novel NE sensor ($GRAB_{NE}$) we show spatiotemporal dynamics of endogenous NE release in DG during aversive stimuli, finding that there were distinct differences in NE release dynamics compared to neutral stimuli. Further, we compare and characterize differences between evoked NE release through optogenetic stimulation at various frequencies to endogenous release dynamics. Lastly, using *in vivo* Ca^{2+} imaging, we implicate α- and β-ARs in the DG in mediating pattern separation and aversive contextual processing, and elucidate neural ensemble activity in DG in response to α_1- and β-AR selective pharmacological agents, finding that α_1-AR selective antagonist prazosin and β-AR selective antagonist propranolol result in increased activity in DG ensemble firing, especially in conjunction with increased LC-NE activity. ***Collectively, our data suggest that LC-DG noradrenergic projections facilitate aversive contextual processing via increased NE release targeting α_1- and β-ARs in the DG.*** Current work is focused on further elucidating spatiotemporal dynamics of NE release, both endogenous and evoked, and to determine whether evoked release of NE is sufficient to create an aversive behavioral response in place of an aversive stimulus or during aversive contextual processing. Additionally, the use of *in vitro* and

in vivo Ca^{2+} imaging will allow for further understanding of the downstream receptors which respond to NE release during aversive contextual processing.

Disclosures: E.T. Zhang: None. **G.S. Saglimbeni:** None. **S.C. Piantadosi:** None. **J. Feng:** None. **Y. Li:** None. **M.R. Bruchas:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.18

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01MH111918

Title: Probing Biobehavioral Endophenotypes of Resilience and Suceptibility using Learned Helplessness and Acute Social Defeat Stress in Male and Female Mice

Authors: *D. FORTUNA[1], H. MCBRIDE[1], L. BRYAN[2], K. R. ANDERSON[1], A. LIPSHUTZ[2], A. MANGANARO[1], D. DUMITRIU[1];
[1]Pediatrics, Columbia Univ. Med. Ctr., New York, NY; [2]Barnard College, Columbia Univ., New York, NY

Abstract: Chronic mental disorders play a significant role in disability across the world. Stressful life events can induce psychopathology and lead to increased rates of chronic mental health disorders. An individual's response to these stressful events varies greatly and can impact behavioral and neurobiological outcomes. Subjects exposed to stress may cope differently depending on various intrinsic and extrinsic factors resulting in active, passive, or maladaptive coping styles. Rodent models such as Learned Helplessness (LH) and Acute Social Defeat Stress (ASDS) are used to uncover a stressor's impact on behavior and the brain's structure and function. LH and ASDS are two well-validated models of divergent stress responses within homogenous (inbred) rodent populations. Both models allow categorization into "resilient" and "susceptible" subpopulations. LH exposes mice to inescapable shocks that cause inappropriate passive behavior or "helplessness" in a subset of mice (LH susceptible), while other mice continue to exhibit normal escape behavior (LH resilient). ASDS exposes mice to repeated bouts of social aggression by an aggressive mouse, which leads to social avoidance in a subset of mice (ASDS susceptible), while other mice continue to show normal social approach (ASDS resilient). Here, we use LH and ASDS (tested 10 days apart on the same cohort of mice) to ascertain if susceptibility and resilience in various settings tap into biobehavioral endophenotypes. Specifically, we hypothesize an LH susceptible mouse is more likely to be ASDS susceptible. Similarly, we hypothesize an LH resilient mouse is more likely to be ASDS resilient. Preliminary data (n=23 mice), shows susceptibility on LH to have a positive predictive value (PPV) of 50% for ASDS susceptibility, and resilience on LH to have a PPV of 73% for ASDS resilience. Additional cohorts are needed to confirm these results, but early data suggest resiliency but not

susceptibility might tap into a biobehavioral endophenotype. In order to test for the neural substrates of these endophenotypes, we are using the Targeted Recombination in Active Populations (TRAP) x Ai mouse model. These mice are genetically modified to allow indelible trapping of activated neurons in the presence of the drug tamoxifen, followed by immunohistochemistry against cFos to stain a second set of activated neurons, thus allowing us to track the neuronal activity at two different time points to study the overlapping and divergent neuronal ensembles activated by LH and ASDS, respectively.

Disclosures: D. Fortuna: None. **H. McBride:** None. **L. Bryan:** None. **K.R. Anderson:** None. **A. Lipshutz:** None. **A. Manganaro:** None. **D. Dumitriu:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.19

Topic: G.01. Fear and Aversive Learning and Memory

Support: K01 MH123783

Title: Dynamic neural coding in the basolateral amygdala links two defensive responses

Authors: *P.-K. O'NEILL[1], J. MESZAROS[1], P. WARREN[1], D. SALZMAN[1,2];
[1]Columbia Univ., New York, NY; [2]Zuckerman Inst., New York, NY

Abstract: When under threat, animals exhibit flexible defensive responses that can shift between freezing, flight, and attack. The basolateral amygdala (BLA) is widely recognized for its role in mediating freezing responses arising from the presentation of an aversively conditioned stimulus (CS) in rodents. However, conditioned stimuli often elicit multiple adaptive behavioral responses. It is unclear how BLA activity observed during one type of defensive response elicited by a CS relates to other subsequent responses. Here we utilize a behavioral assay - the virtual burrow - in which head-fixed mice display two types of defensive behaviors that can occur in sequence: freezing-like behavior (tremble) and flight-like behavior (ingress into the burrow). Both ingress and tremble arise naturally - without explicit training - following Pavlovian aversive conditioning. Two-photon imaging of BLA reveals that activity in neural ensembles during time intervals that precede an ingress can predict subsequent ingress. This predictive signaling occurs specifically during periods of tremble that can occur several seconds prior to the ingress. These results suggest that BLA ensemble activity during tremble can represent an emotional state predictive of subsequent ingress, and not necessarily the motor action of ingress itself. By representing emotional states that can produce multiple defensive behaviors, the BLA may thereby provide an underlying neural substrate needed for valence-congruent shifts between different defensive behaviors.

Disclosures: P. O'Neill: None. **J. Meszaros:** None. **P. Warren:** None. **D. Salzman:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.20

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01MH112355
F31DA056148

Title: Norepinephrine and dopamine release from locus coeruleus projections during stress exposure and reward seeking

Authors: *A. K. MATARASSO[1], E. SEAHOLM[1], I. RODRIGUEZ REYES[2], J. FENG[4,5], Y. LI[4,5], M. R. BRUCHAS[3];
[1]Dept. of Bioengineering, [3]Dept. of Anesthesiol. and Pain Med., [2]Univ. of Washington, Seattle, WA; [4]State Key Lab. of Membrane Biology, Peking Univ., Beijing, China; [5]PKU-IDG/McGovern Inst. for Brain Res., Beijing, China

Abstract: The locus coeruleus (LC) is a small neuromodulator source with broad projections across the central nervous system that predominantly influence arousal, anxiety, and learning. For example, LC excitation leads to release of NE in the BLA, producing anxiety-like behavior in mice. In dorsal CA1, NE release and minor DA release from optogenetically activated LC terminals improved novelty-associated spatial learning. However, the conditions and behavioral events under which DA release occurs, and the relative magnitude of release remain unclear. Here, we used biosensors to isolate monoamine release following selective photo-stimulation of LC terminals in these regions. We also explore the endogenous dynamics of NE and DA release in BLA and hippocampus in response to appetitive and aversive stimuli. To target NE neurons, we used mice expressing Cre-recombinase under the dopamine beta hydroxylase (*Dbh)* promotor. *Dbh-cre* mice were injected with the Cre-dependent red-shifted Channelrhodopsin, ChrimsonR in the LC, and with genetically-encoded neuromodulator sensors GRAB-NE and dLight (to sense NE and DA, respectively) in BLA and CA1. Optic fibers were implanted to detect bulk fluorescence from each sensor. To characterize the properties of monoamine release following diverse physiological activity, optogenetic stimulation (for 3s at 1, 3, 5, 10, 15, and 20hz) of ChrimsonR-infected LC terminals revealed frequency response profiles of release in CA1, and slightly diminished release of DA from LC terminals at higher frequencies. Longer photo-stimulation (30s) led to more varied results, where the BLA had greater NE than DA release. LC-CA1 terminals had NE and DA release profiles that increased linearly until saturation at higher frequencies. *Ex vivo* 2-photon imaging of sensors revealed the fluorescent response to ligand. This modality also elucidated optogenetically elicited release of monoamines under pharmacological manipulation. In experiments measuring endogenous release, we found increased NE release in response to salient anxiogenic stimuli, such as repeated shock and looming stimuli, in the BLA and CA1. DA had a larger and quicker release in the BLA than CA1 during looming. During operant reward conditioning, NE signal decreased in CA1 after mice

nose poked for reward and increased after reward was delivered. In the BLA, NE and DA are released in response to both completing actions for reward and receiving the reward. These data reveal release of DA and NE across LC projections to BLA and CA1. (Funded by R01MH112355, F31DA056148)

Disclosures: **A.K. Matarasso:** None. **E. Seaholm:** None. **I. Rodriguez Reyes:** None. **J. Feng:** None. **Y. Li:** None. **M.R. Bruchas:** None.

Poster

556. Neurobiology of Fear and Aversion

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 556.21

Topic: G.01. Fear and Aversive Learning and Memory

Support: JSPS KAKENHI 20K23041

Title: Activation of the neurons in the central amygdala attenuates disgust responses and reduces fearful approaches in the retrieval of conditioned taste aversion

Authors: T. INUI[1], E. KIKUCHI[1,2], M. FUNAHASHI[1];
[1]Oral Physiol., Grad Sch. Dent. Med., [2]Orthodont., Grad. Sch. Dent. Med., Hokkaido Univ., Sapporo, Japan

Abstract: The central amygdala (CeA) receives gustatory and visceral inputs from the brainstem. The CeA is suggested to be involved in conditioned taste aversion (CTA), which is established by pairing a taste as a conditioned stimulus (CS) with a visceral malaise. CTA decreases the taste palatability of a CS and increases fearful approaches to it. Since the role of the CeA in these behavioral responses remains unclear, we used Gq-DREADD (designer receptors exclusively activated by designer drugs) to assess how the excitation of the neurons in the CeA alters the taste palatability and fearful approaches to a CS during the retrieval of CTA. Male C57/BL mice received injections of AAV8-hSyn-hM3Dq-mCherry (0.3 µl/side) into the bilateral CeA. After the surgery, they were trained to drink deionized water for 15 min in a behavioral analysis apparatus. Then the mice were conditioned by pairing 0.2% saccharin solution as a CS with 0.3 M lithium chloride (2% B.W., i.p.). In Test 1, the conditioned mice were presented with the CS in the apparatus to examine the establishment and robustness of the CTA. They were divided into the experimental and control groups whose average CS consumptions in Test 1 were as close as possible. Tests 2 and 3 examined the effect of excitation of the CeA neurons by administering deschloroclozapine (DCZ, 50 µg/kg, 0.5% B.W.), a ligand to hM3Dq, in the experimental group. The control group was administered with a vehicle (1% DMSO in saline, 0.5% B.W.) instead of DCZ. Shortly (90 min; Test 2) or two days (Test 3) after the administration, the mice were presented with the CS. The experimental group consumed a significantly larger amount of CS than the control group in Test 2 (p < 0.05). The microstructural lick analysis revealed that the experimental group showed augmentation of the

lick number during the first minute of the session and the mean size of burst licking (p < 0.05, respectively). Since the burst size is an index of taste palatability, the DCZ injection was likely to attenuate disgust for the CS, resulting in larger consumption immediately after the start of the session. The analysis of approaching behavior revealed that the frequency of the access to the syringe containing the CS in the experimental group was significantly larger than that in the control group (p < 0.01), indicating that the DCZ injection reduced fearful approaching the CS. It appears that the DCZ injection temporarily altered these behavioral responses because the group differences were significant in Test 2 but not in Test 3. These results suggest that the neuronal activities of the CeA play a role in decreasing taste palatability and increasing fearful approaches during the retrieval of CTA.

Disclosures: **T. Inui:** None. **E. Kikuchi:** None. **M. Funahashi:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.01

Topic: G.02. Reward and Appetitive Learning and Memory

Support: The Ray and Dagmar Dolby Family Fund
BBRF NARSAD Young Investigator Grant
CIHR Postdoctoral Fellowship
NIH R01 MH108623

Title: Decoding stress susceptibility from activity in an extended ventral hippocampal network

Authors: *F. XIA[1], N. VISHWAKARMA[1], V. FASCIANELLI[2], F. G. GHINGER[1], S. FUSI[2], M. KHEIRBEK[1];
[1]Psychiatry and Behavioral Sci., Univ. of California San Francisco, San Francisco, CA;
[2]Zuckerman Institute, Theoretical Neurosci., Columbia Univ., New York, NY

Abstract: Major depressive disorder is characterized by network-level functional alterations, with the vHPC and its connected regions being central components. However, how changes in emotional state are represented in the activity patterns of single neurons and populations in the extended vHPC network remains unclear. Here, using high-density Neuropixels probes, we recorded single unit activity across the extended vHPC network, including vHPC subregions, amygdala (AMY), medial prefrontal cortex (mPFC), and thalamus. This allowed us to investigate how stress modulates representations of emotional information in the vHPC circuit and the effects on reward-seeking behavior. Mice were first subjected to chronic social defeat stress, a well-validated rodent model for induction of depression-related behavior. After defeat, we recorded single unit activity across a vHPC-AMY-mPFC-thalamic network as mice performed a head-fixed sucrose preference test where they freely choose between consuming water vs. sucrose rewards, a classic test of anhedonia-related behavior. Our results show that

sucrose preference after defeat was correlated with social interaction ratio, a standard measure of stress susceptibility. At the single neuron level, we found that spiking activity was greatly elevated during reward consumption in control (non-stressed) mice across the vHPC-AMY-mPFC-thalamic network, but stressed mice showed region-specific changes in firing patterns, including decreased activity in AMY and vHPC neurons, and elevated firing rates in mPFC neurons. Interestingly, despite reduced sucrose preference behaviorally in the stressed mice, neurons in vHPC and AMY showed increased reward selectivity. To probe this phenomenon further at the population level, we used support vector machine classifiers to decode reward representations. We found that in stressed mice, future reward choice (sucrose or water) could be decoded seconds before the choice was made in the AMY, mPFC, and vHPC neurons, suggesting a rigidity in future reward representations. In addition, neurons in these regions showed smaller change in decoding accuracy from pre- to post-reward, suggesting reduced reward sensitivity in comparison to controls. Furthermore, coordinated activity between vHPC and AMY neurons is correlated with individual animal's sucrose preference, suggesting that reduced vHPC-AMY interaction may underlie anhedonia. Together, our results show that chronic stress alters single neuron firing patterns and population representations of reward in the vHPC network, and these changes may be responsible for ultimately driving anhedonic behavior following stress.

Disclosures: **F. Xia:** None. **N. Vishwakarma:** None. **V. Fascianelli:** None. **F.G. Ghinger:** None. **S. Fusi:** None. **M. Kheirbek:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.02

Topic: G.02. Reward and Appetitive Learning and Memory

Support: F31DA053724 (J.E.E)
R01DA044315 (L.S.Z)
K99DA054265 (B.J.)

Title: Dissociable activity dynamics in mesolimbic dopamine subpopulations during reinforcement learning and motivation

Authors: *J. ELUM[1], B. JUAREZ[2], G. LOGINOV[3], S. NG-EVANS[2], L. ZWEIFEL[4];
[1]Grad. Program in Neurosci., [2]Pharmacol., [3]Neurosci., [4]Pharmacol. & Psychiatry, Univ. of Washington, Seattle, WA

Abstract: The mesolimbic dopamine system regulates multiple behavioral dimensions that contribute to learning, motivation, and performance. A central question is how the mesolimbic dopamine system influences these distinct functions. In reinforcement learning and motivation, a classic two-component system has been proposed in which dopamine release in the nucleus

accumbens (Nac) core provides prediction error signals to regulate reinforcement learning and simultaneous dopamine release in the NAc shell signals incentive salience and promotes motivated responses. To determine if ventral tegmental area (VTA) dopamine subpopulations display differential calcium dynamics at the cell body level during reinforcement learning and motivation, we recorded bulk calcium dynamics in two distinct cell populations that project to either the core or shell subregion of the NAc. We find that NAc core- and shell-projecting dopamine populations are activated by actions, cues, and rewards. However, the calcium dynamics during these events differ between the two populations. NAc shell-projecting VTA dopamine neurons showed larger responses during action initiation (lever press) and displayed a sustained increase in activity during the cue and reward outcome periods. In contrast, in the NAc core-projecting population these signals return to near baseline levels between discrete events. During unexpected reward omission, the NAc core-projecting population displayed temporally discrete decreases in calcium signals consistent with a prediction error-encoding model that were not observed in the shell-projecting dopamine cells. By optogenetically manipulating both the NAc shell-projecting and core-projecting VTA dopamine populations we find that these populations differentially regulate cued reinstatement behavior. These findings support the distinct functionality of these neuronal subtypes in learning and motivation through differential regulation of neural activity at the level of the cell body.

Disclosures: **J. Elum:** None. **B. Juarez:** None. **G. Loginov:** None. **S. Ng-Evans:** None. **L. Zweifel:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.03

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH Grant 1F32MH127801-01A1
NIH Grant R01DA04431

Title: Dynamic encoding of valence and salience in central amygdala neurons during appetitive and aversive learning

Authors: *M.-S. KONG[1], B. SURFACE[3,4], L. S. ZWEIFEL[1,2];
[1]Dept. of Psychiatry and Behavioral Sci., [2]Dept. of Pharmacol., Univ. of Washington, Seattle, WA; [3]Dept. of Biol., Tacoma Community Col., Tacoma, WA; [4]NIH Blueprint for Neurosci. Res., NIH, Bethesda, MD

Abstract: Valence is the subjective value allocated to stimuli determining affective state and behavioral response. Positive valence is assigned to reinforcing stimuli, which leads to approach behaviors. In contrast, negative valence associated with aversive stimuli promotes behavioral avoidance. Salience reflects the strength of the stimulus by endowing it with the ability to attract

attention and prompt preparatory responses. The central nucleus of the amygdala (CeA) has emerged as a brain structure that detects valence and salience at the early integration point to engage behavioral outcomes. It remains unclear the extent to which the CeA encodes valence or salience at the single-cell level. To address this, we classified valence vs. salience encoding CeA neurons by monitoring calcium dynamics during appetitive and aversive learning. Following injection of a conditional GCaMP6 expressing virus into the CeA of Vgat-Cre mice, a gradient-index lens was implanted to record fluorescent calcium signals. After postoperative recovery, mice were maintained at ~85% of their preoperative body weight to motivate food-seeking behavior during the experiments. Mice underwent Pavlovian appetitive and Pavlovian threat conditioning in a counterbalanced fashion. For the appetitive conditioning, a tone was paired with a food reward for 20 trials on 10 consecutive days. For the aversive conditioning, a different frequency tone was paired with a foot shock 10 times in a single conditioning session Fluorescent calcium transients were collected during the appetitive and aversive learning using the one-photon *in vivo* calcium imaging technique (Inscopix nVoke system). We found that 53% of CeA neurons were responsive during the last day of appetitive learning, where 31% of neurons were excited and 22% of neurons were inhibited by the food reinforcer. Interestingly, this proportion was conserved during the aversive learning; 32% excited and 21% inhibited by the foot shock. Valence and salience encoding neurons were designated based on their response during the two conditioning sessions. Among neurons that were responsive in both paradigms, we found that 60% of neurons encoded either positive or negative valence, and 15% of neurons encoded salience (i.e., encoding the extent of attentional value). These results suggest the functional heterogeneity of the CeA in encoding two opposing valences and provide further evidence that the CeA is an important valence/salience detection center of the brain to promote appropriate approach and avoidance behavioral responses.

Disclosures: **M. Kong:** None. **B. Surface:** None. **L.S. Zweifel:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.04

Topic: G.02. Reward and Appetitive Learning and Memory

Support: PHS NRSA T32GM007270 from NIGMS

Title: TRPC6 channels signal reward uncertainty in VTA-Tacr3 neurons during instrumental learning

Authors: *M. X. BERNSTEIN[1], L. S. ZWEIFEL[2];
[1]Grad. Program In Neurosci., [2]Psychiatry & Behavioral Sciences, Pharmacol., Univ. of Washington, Seattle, WA

Abstract: Midbrain dopamine (DA) neurons play a critical role in modulating reward-seeking behavior. Dysregulation of DA signaling has been linked to a variety of neurological and psychiatric disorders, such as schizophrenia, substance use disorders, and depressive disorders. Hyperforin, the main active ingredient in the herbal supplement St. John's wort, has modest antidepressant action through its effect on serotonin reuptake. Hyperforin has also been shown to modulate calcium ion flux through transient receptor potential canonical type 6 (TRPC6) channels. TRPC6 channels are enriched in DA neurons and calcium signaling is known to play an important role in regulating DA neuron function, but the functional significance of TRPC6 enrichment in DA neurons has not yet been established. Our lab has recently discovered that tachykinin receptor 3 (Tacr3)-expressing neurons are a minimally sufficient population of midbrain DA neurons that promote reward reinforcement behavior. Additionally, Tacr3 activation is directly linked to the TRPC channel activity. To determine whether TRPC6 channels are a key regulator of calcium dynamics in VTA-Tacr3 neurons during instrumental learning, we used CRISPR/Cas9 technology to selectively mutate the Trpc6 gene to generate loss of function in a cell-type specific manner within the VTA of adult mice. We recorded *in vivo* calcium dynamics in Tacr3 neurons with and without TRPC6 channels during instrumental learning. We find evidence that TRPC6 channels contribute to reward uncertainty during instrumental learning. Additionally, TRPC6 channels regulate the scalability of the response of Tacr3 neurons when a reward is unexpected. Together, these findings advance our understanding of the molecular mechanisms underlying motivated behavior, which is especially critical in the context of dopamine-mediated disorders.

Disclosures: M.X. Bernstein: None. **L.S. Zweifel:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.05

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NARSAD Young Investigator Grant
NSF GR Fellowship
NIH R01 MH111754
One Mind/IMHRO
NIH R01 MH108623
NIH R56 MH117961

Title: Neural dynamics of associative learning across the dorsoventral hippocampus

Authors: *J. BIANE[1], M. A. LADOW[2], F. STEFANINI[3], S. BODDU[4], A. FAN[5], S. HASSAN[4], N. DUNDAR[4], D. APODACA-MONTANO[4], V. FAYNER[4], N. I. WOODS[4], M. KHEIRBEK[6];
[1]Univ. of California San Francisco, Univ. of California, San Fransisco, San Francisco, CA; [2]Univ. of California San Francisco,, San Francisco, CA; [3]Columbia Univ., New York, NY;

[4]Univ. of California San Francisco, San Francisco, CA; [5]Univ. of California, Berkeley, Univ. of California, Berkeley, Berkeley, CA; [6]Univ. of California, San Francisco, Univ. of California San Francisco Neurosci. Grad. Program, San Francisco, CA

Abstract: The hippocampus creates a cognitive map of the world to optimize predictions and guide behavior, with the dorsal and ventral hippocampus believed to play unique roles in this process. To understand how such representations evolve with learning and are updated with new experience, we used 2-photon calcium imaging to track the same dCA1 or vCA1 neurons across days as mice learned to associate odor stimuli with different outcomes. During reward (sucrose) associative learning, we find that, initially, odors elicited robust responses in dCA1, whereas in vCA1 encoding of odor identity was weak, but was invigorated following odor-outcome associative learning. Neural representations in both regions rapidly reorganized with learning, then stabilized into ensembles that stored odor representations for days, even after extinction or pairing with a different outcome. Additionally, both dCA1 and vCA1 displayed reward anticipation signals that generalized across predictive odors and were stable across days. In contrast to this strong encoding of reward anticipation, outcome anticipation signals were only weakly encoded when anticipating inescapable shocks, and were further reduced with extended task exposure. These data suggest that hippocampal engagement during outcome expectation may be modulated by the behavioral relevance of the outcome (ie, behaviorally relevant (lick-dependent sucrose) reward vs irrelevant (inescapable) shock). Collectively, these results identify how the hippocampus encodes, stores, and updates learned associations, and illuminates the unique contributions of dorsal and ventral hippocampus.

Disclosures: J. Biane: None. **M.A. Ladow:** None. **F. Stefanini:** None. **S. Boddu:** None. **A. Fan:** None. **S. Hassan:** None. **N. Dundar:** None. **D. Apodaca-Montano:** None. **V. Fayner:** None. **N.I. Woods:** None. **M. Kheirbek:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.06

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NARSAD YI Award
R01MH108623
NSF GRFP

Title: Ventral CA1 ensembles strongly encode identity, but not valence, of salient stimuli

Authors: *M. LADOW[1], J. BIANE[1], R. O'SULLIVAN[2], M. M. GERGUES[3], A. FAN[4], D. APODACA-MONTANO[2], M. KHEIRBEK[5];
[1]Univ. of California San Francisco, San Francisco, CA; [2]UCSF, San Francisco, CA; [3]Neurosci. Grad. Program, Univ. of California, San Fransisco, San Francisco, CA; [4]Univ. of California,

Berkeley, San Ramon, CA; [5]Psychiatry, Univ. of California San Francisco Neurosci. Grad. Program, San Francisco, CA

Abstract: In the last 20 years the ventral hippocampus has been increasingly implicated as an important component of multiple networks that control emotional and motivated behavior. It can modulate a diverse set of behaviors, including anxiety-related behavior, reward seeking, social memory and control of stress responses. Due to it's anatomical organization, the largely non-overlapping pyramidal neuron outputs to distinct downstream targets, a dominant hypothesis of it's function has emerged, one that posits that appetitive and aversive stimuli are encoded within distinct ensembles of neurons. Here, we directly tested this hypothesis using 2-photon calcium imaging of dorsal and ventral CA1 region of the hippocampus (dCA1, vCA1) during exposure to a battery of salient stimuli, including appetitive and aversive odorants, sucrose rewards and aversive foot shocks. Analysis of single neuron tuning responses found subpopulations of neurons that were tuned to distinct stimuli, with a subset of neurons having stable stimulus responses over multiple days. In addition, we found populations of neurons that responded to multiple stimuli, regardless of valence class. Using linear classifiers to determine how stimulus identity was represented within the population, we found that, in vCA1, stimulus identity could be discriminated from the population activity for all pairs of stimuli tested, regardless of valence class. In addition, across-session decoder that was trained on activity one day, and tested on another day performed significantly better than chance, indicating that ensembles encoding each salient stimulus was stable across days. These data highlight the need to revise the dominant hypothesis of vCA1 function, and indicates that instead of distinct vCA1 ensembles encoding appetitive and aversive stimuli, vCA1 ensembles represent the identity of salient stimuli irrespective of their valence class. Ongoing work will identify how these ensembles represent spatially anxiogenic stimuli, and how these representations map onto the well-described anatomy of vCA1.

Disclosures: **M. Ladow:** None. **J. Biane:** None. **R. O'Sullivan:** None. **M.M. Gergues:** None. **A. Fan:** None. **D. Apodaca-Montano:** None. **M. Kheirbek:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.07

Topic: G.02. Reward and Appetitive Learning and Memory

Support: R01MH108623

Title: Local control of ventral hippocampal output by classes of inhibitory interneurons controls approach and avoidance behaviors

Authors: *J. X. BRATSCH-PRINCE[1], M. KHEIRBEK[2];
[1]Univ. of California, San Francisco, San Francisco, CA; [2]Psychiatry, Univ. of California San Francisco Neurosci. Grad. Program, San Francisco, CA

Abstract: The ventral hippocampus (vHPC) plays a critical role in mood and anxiety-related behaviors. Excitatory pyramidal neurons (PNs) in the CA1 subregion of the vHPC send projections to several cortical and subcortical targets. Activity in projection-defined vCA1 PNs has been shown to influence distinct approach and avoidance behaviors depending on the target structure. This suggest that vCA1 must transform incoming information regarding the salient nature of an environment to an output signal to specific downstream targets to support appropriate behavioral selection. At the local circuit level, activity in hippocampal PNs is regulated by a diverse pool of local inhibitory interneurons (INs), including the largely nonoverlapping populations of those expressing parvalbumin (PV), somatostatin (SST), or vasoactive intestinal polypeptide (VIP), which differ in their anatomical and electrophysiological profile. This suggests, in vCA1, the balance of activity in these classes of INs can differentially tune circuit function to drive distinct PN output patterns and behavior. However, how this occurs during behavior remains unknown. Here, using in vivo calcium imaging and optogenetics in mice, we show that in vCA1 projection-defined PNs, PV, SST, and VIP INs are differentially modulated during multiple tests of anxiety-related behavior. High-resolution analysis of mouse behavior found that during exploratory-approach behaviors, PV and VIP IN neurons were recruited, while SST IN activity decreased. Conversely, during avoidance trajectories the opposite occurred, SST INs were recruited, while PV and VIP INs decreased firing. This coordinated activity in vCA1 INs during approach/avoidance behaviors was associated with recruitment of distinct projection-defined vCA1 output PNs. This reveals the competitive balance between vCA1 IN populations mediates vCA1 output by transforming inputs into a vCA1 output signal that drives appropriate behavioral selection during anxiety-related behaviors.

Disclosures: J.X. Bratsch-Prince: None. **M. Kheirbek:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.08

Topic: G.02. Reward and Appetitive Learning and Memory

Support: 1R01DC019813

Title: Neuromodulatory control of dentate gyrus granule cells during discrimination learning

Authors: *H. CHOI, F. XIA, J. BIANE, G. TELIAN, M. KHEIRBEK;
Univ. of California, San Fransisco, San Francisco, CA

Abstract: The brain transforms experiences into patterns of activity that control emotions, decisions and behaviors. Discriminating these patterns of activity allows these experiences to be

stored into distinct entities, separating important stimuli from unimportant ones, which is a crucial feature of episodic memory. One way that neural circuits may discriminate experiences is by, at the population level, separating the neural representations of these experiences through learning, allowing a readout area to better decode the stimulus from the patterns of activity. We have recently found that the dentate gyrus subregion of the hippocampus classifies cortical representations of olfactory stimuli, increasing the distance between odor representations with learning. These studies indicate that, through associative learning, the CS+ odor becomes overrepresented at the expense of the CS- odor and odor representations reduce overlap, i.e., become more orthogonal. Here, we examined the neuromodulatory mechanisms underlying this process, and show that dopamine (DA) release in the DG may underlie some of the learning-induced enhancements of pattern separation in the dentate gyrus. To understand the dynamics of DA signaling in the DG, we performed an odor-reward conditioning experiment while performing 2-photon imaging of DA dynamics in DG GCs using the DA sensor GRAB-DA. To examine DA dynamics throughout odor-reward learning in the DG, we injected WT mice with AAV9-hSyn-DA4.4, and performed high resolution 2-photon cellular imaging of GRAB-DA expressing GCs in the DG. Before learning, we observed little DA activity in response to the odor delivery but observed a significant DA response to reward delivery/consumption. With learning, however, we found a temporal shift in the DA response, as the odor delivery and trace period elicited a robust DA response in DG GCs. Next, we traced the inputs to the DG to determine the relative reward source of dopamine to the dentate gyrus, and found TH expressing inputs arising from both ventral tegmental area and locus coeruleus. Ongoing work is examining the functional source of DA input to the DG, as well as the consequence of manipulating levels of DA in the DG on pattern separation in DG GCs. These studies highlight the importance of neuromodulatory input on discrimination processes in the DG.

Disclosures: **H. Choi:** None. **F. Xia:** None. **J. Biane:** None. **G. Telian:** None. **M. Kheirbek:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.09

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH 1R15MH122729-01

Title: Investigating the effect of differential striatal dopamine projections on reversal learning and immediate early gene (IEG) expression

Authors: *M. GHANEM, R. K. VAN DER MERWE, C. MOREHOUSE, C. MADDOX, R. ALBERT-LYONS, V. GROCE, C. D. HOWARD;
Neurosci., Oberlin Col., Oberlin, OH

Abstract: Different striatal compartments have differential contributions to behavioral flexibility. For example, the dorsomedial (DMS) and dorsolateral (DLS) striatum have been shown to facilitate goal-directed and habitual behaviors, respectively. However, how dopamine within these subregions contributes to reversal learning, a widely used metric of behavioral flexibility, remains unclear. To explore this, we used an instrumental reversal-learning task where mice pressed one of two levers to earn sucrose pellets before the active lever was reversed. We simultaneously used optogenetic stimulation in DATcre x Ai32 mice to activate dopamine release in either DMS or DLS during the learning phase with stimulation occurring at either lever presses or rewarded head entries. All mice could reverse by the end of a 10-day reversal phase, with DMS groups showing modest enhancement of flexibility. Next, in a separate group of animals, we assessed the expression of immediate-early genes associated with memory formation and synaptic plasticity (c-Fos and Arc) in WT mice during a similar reversal task. Throughout training and reversal, the DLS maintained a higher level of gene expression relative to DMS, while DMS had gradually decreasing levels as reversal progressed. This result supports a model in which the DMS and the DLS are active throughout learning with declining influence of DMS as learning progresses. We next aim to explore how stimulating dopamine during reversal, rather than during training, influences the rate of learning. These results provide insights into striatal subcircuits that guide reinforcement and reversal learning.

Disclosures: M. Ghanem: None. **R.K. Van Der Merwe:** None. **C. Morehouse:** None. **C. Maddox:** None. **R. Albert-Lyons:** None. **V. Groce:** None. **C.D. Howard:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.10

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH Grant R15MH122729-01

Title: Characterization of striatal dopamine projections across striatal subregions in reversal learning

Authors: *R. VAN DER MERWE[1], J. A. NADEL[3], D. COPES-FINKE[1], S. S. PAWELKO[1], J. S. SCOTT[1], M. GHANEM[1], M. FOX[1], C. MOREHOUSE[4], R. MCLAUGHLIN[1], C. MADDOX[1], R. ALBERT-LYONS[1], G. MALAKI[1], A. TUROCY[1], V. GROCE[1], X. JIN[5], C. D. HOWARD[2];
[2]Neurosci., [1]Oberlin Col., Oberlin, OH; [3]Dept. of Neurosci., Northwestern Univ., Chicago, IL; [4]Neurosci., Oberlin Col. Neurosci. Dept., Oberlin, OH; [5]East China Normal Univ., Shanghai, China

Abstract: Behavioral flexibility is key to survival in a dynamic environment. While flexible, goal-directed behaviors are thought to initially be dependent on dorsomedial striatum, they

become dependent on lateral striatum as behaviors become inflexible. Similarly, lesions of dopamine terminals in lateral striatum disrupt the development of inflexible habits. This work suggests that dopamine release in lateral striatum may drive inflexible behaviors, though few studies have investigated a causative role of subpopulations of striatal dopamine terminals in reversal learning, a measure of flexibility. Here, we performed two optogenetic experiments to activate dopamine terminals in dorsal medial (DMS), dorsal lateral (DLS), or ventral (NAc) striatum in DATcre mice that expressed channelrhodopsin-2 via viral injection (Experiment I, n=24) or through transgenic breeding with an Ai32 reporter line (Experiment II, n=23) to determine how specific dopamine subpopulations impact reversal learning. Mice performed a reversal task in which they self-stimulated DMS, DLS, or NAc dopamine terminals by pressing one of two levers before action-outcome lever contingencies were reversed. Consistent with presumed ventromedial/lateral striatal function, we found that mice self-stimulating ventromedial dopamine terminals largely reversed lever preference following contingency reversal, while mice self-stimulating dopamine terminals in DLS showed impaired reversal. Impairments in DLS mice were characterized by more regressive errors (pressing the now-inactive lever following first sampling of the now-active lever each day) and reliance on lose-stay (selection of inactive lever following selection of inactive lever) strategies following reversal, as well as reduced within-session learning, suggesting reward insensitivity and overreliance on previously learned actions. In contrast, relatively fewer control DATcre^{-} Ai32$^{+/-}$ mice (n=12) met inclusion criteria compared to experimental groups, and those that met criteria did not significantly update lever preference following lever reversal suggesting these effects are not due to off-target effects of laser stimulation. This work supports a model of striatal function in which dorsomedial and ventral dopamine facilitate goal-directed responding, and dorsolateral dopamine supports more inflexible responding.

Disclosures: **R. Van Der Merwe:** None. **J.A. Nadel:** None. **D. Copes-Finke:** None. **S.S. Pawelko:** None. **J.S. Scott:** None. **M. Ghanem:** None. **M. Fox:** None. **C. Morehouse:** None. **R. McLaughlin:** None. **C. Maddox:** None. **R. Albert-Lyons:** None. **G. Malaki:** None. **A. Turocy:** None. **V. Groce:** None. **X. Jin:** None. **C.D. Howard:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.11

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIDDK gran R01DK085721

Title: Recruitment of cortical and thalamic projections to the central amygdala in the control of feeding behavior under novelty

Authors: *E. GREINER, G. D. PETROVICH;
Boston Col., Boston Col., Chestnut Hill, MA

Abstract: Adaptive responses to novel foods and environments are essential for survival. Consumption of a new food could lead to illness or even death. Similarly, consumption can be greatly affected when it occurs in a novel context. Previously, we showed, that when rats were tested in a novel context, males habituated to novel taste and consumed large amounts of novel food, much faster than females, who showed suppressed consumption throughout testing. We have also found that food consumption in a novel context induced Fos in the central nucleus of the amygdala (CEA). However, the inputs that drive CEA activity are not known. The CEA receives direct inputs from the paraventricular nucleus of the thalamus (PVT), the infralimbic region of the medial prefrontal cortex (ILA), and the agranular insula (AI), which could control CEA behavioral output and specifically mediate appetitive behavior. Despite these regions being strong candidates for mediating the effects of novelty on food consumption, their connections with the CEA are yet to be examined and it is completely unknown if these inputs control food consumption under novelty differently in males and females. To establish the underlying neural circuitry that mediates the inhibition of food intake under novelty, we used a combination of retrograde tract tracing and Fos induction to determine whether PVT, ILA, and AI neurons that send direct projections to the CEA are specifically recruited during the consumption test under novelty and whether that activation was sex specific. Male and female Long Evans rats received unilateral injections of Fluorogold to retrogradely label cells in the CEA. After recovery and following 20hr acute food deprivation, rats were tested for consumption in either familiar or novel condition (n=8 per group for each sex). In the familiar condition, rats were fed in their home cages and were given a familiar food (rat chow) and in the novel condition, rats were fed in a novel context and were given a novel (Test Diet pellets) food. Consistent with our previous findings, male and female rats in a novel condition consumed less than those in a familiar condition ($F(1,26)=25.64$, $p<0.01$). Our preliminary neural analysis found Fos induction in retrogradely labelled neurons within each of the three regions of interest, indicating that the ILA, AI, and PVT inputs to the CEA were activated during the tests. Full analysis will test the prediction that the PVT, ILA, and AI pathways to the CEA will be differently recruited in rats that inhibit feeding under novelty compared to familiar condition and that there will be sex differences in activation patterns that are predictive of behavioral sex differences in habituation.

Disclosures: E. Greiner: None. **G.D. Petrovich:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.12

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIDDK grant R01DK085721

Title: Impact of hunger state on palatable food-cue associative learning and consumption in males and females

Authors: *R. SHTEYN, D. S. LAFFERTY, G. D. PETROVICH;
Boston Col., Boston Col., Boston, MA

Abstract: Physiological hunger state and food palatability could impact food consumption either jointly or independently through homeostatic and hedonic mechanisms. Both also influence food-seeking and learning about cues for food. Here we examined how males and females learn about and consume palatable food (PF; Test Diet pellets) under sated and hungry conditions. Male and female adult Sprague Dawley rats were either food restricted (85% *ad libitum* body weight) or had *ad libitum* access to regular chow (n=8 per group). Rats learned cue-food associations across 8 Pavlovian conditioning sessions, followed by cue-only presentations for 4 extinction sessions in a different context. They were then tested for renewal of conditioned responding (time spent at the food cup) to the food cue in the conditioning compared to extinction context, counterbalanced. Before learning and after renewal testing, rats were tested for consumption of PF and chow in their home cage (1hr test/day per food; counterbalanced). Hungry rats ate more PF and chow than sated rats regardless of sex ($p<.001$). Sated females ate more palatable food than sated males during each test ($p=.003$, $p=.006$). Hungry rats exhibited higher conditioned responding during early conditioning (sessions 1-3; $p<.005$) and had a higher learning rate than sated rats (higher rate of change from the first half of session 1 compared to second half of session 2; $p=.001$). The rate of learning during early conditioning was also marginally higher for females ($p=.053$). For the remaining 5 training sessions, all groups responded similarly. As expected, during extinction, all groups decreased their conditioned responding. All groups had significantly higher conditioned responding to the food cue in the acquisition compared to extinction contexts, confirming the renewal effect. Importantly, the amount of palatable food eaten during the initial consumption test was positively correlated with subsequent cue-food learning during early conditioning, in terms of both conditioned responding and learning rate ($p<.05$, $p<.001$). Overall, these results found that hungry rats of both sexes were more motivated to consume food and acquired cue-food responding faster initially. However, both sexes were able to learn, extinguish and renew responding without hunger. Sated females were more motivated to consume palatable food than sated males, and regardless of hunger state, females exhibited higher initial rates of learning. These findings suggest several sex-specific differences concerning food motivation. Future research of the neural substrates in both sexes is needed in order to identify the drivers of hedonic food motivation without hunger.

Disclosures: R. Shteyn: None. **D.S. Lafferty:** None. **G.D. Petrovich:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.13

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH NIDDK Grant R01DK085721

Title: Sex specific recruitment of the basolateral amygdala complex after habituation to novel food and feeding context

Authors: *Z. R. IRVING, E. M. GREINER, G. D. PETROVICH;
Boston Col., Boston, MA

Abstract: Novel foods and novel environments both impact consumption, but their interaction is poorly understood, especially how this interaction varies across habituation and by sex. Our prior work showed that rats consume less of a novel, palatable food when fed in a novel environment, and female rats habituated slower than males to eating in a novel context. We also established that amygdala nuclei were distinctly recruited upon exposure to novelty, with Fos induction in the anterior basomedial nucleus (BMAa) remaining high after habitation. Additional analyses showed that the anterior basolateral (BLAa) and lateral (LA) nuclei were recruited in a sex-specific manner after habituation to novelty. Here, we examined the posterior basomedial (BMAp) and the posterior basolateral (BLAp) nuclei, and evaluated correlations across amygdala nuclei. Male and female Long Evans rats (n=8/group) were food deprived for 20h prior to consumption of novel, palatable Test Diet pellets in either their home cage or a novel context for 4 habituation sessions and 1 final test. Rats were perfused 90 minutes after the start of the test, and brain tissue was processed for analysis of Fos induction. During the final test, rats tested in the novel context consumed significantly less than those tested in the home cage ($F(1,28)=16.75$, $p=0.02$), and females ate less than males ($F(1,28)=8.68$, $p=0.08$). Fos induction in the BMAp differed by context, with more induction in the novel context than the home cage ($F(1,22)=34.21$, $p=0.001$), but did not differ by sex. In the BLAp, Fos induction differed by both sex ($F(1,22)=12.38$, $p=0.06$) and context ($F(1,22)=13.17$, $p=0.055$), with more induction in females and rats tested in the novel context. Males tested in the novel context showed strong correlations in amygdala Fos induction. BMAp Fos induction was correlated with the BMAa ($p=0.058$) and the BLAa ($p=0.008$), but not with the BLAp, while the BLAp was correlated with the LA ($p=0.06$). Fos induction in home cage males was not consistently correlated across amygdala nuclei, except for a moderate correlation ($p=0.07$) between the BMAp and BLAp. For females tested in the novel context, Fos induction in the BMAp and BLAp were highly correlated ($p=0.009$), as well as the BMAp with the BMAa ($p=0.063$), and the BLAp with the BLAa ($p=0.04$), LA ($p=0.05$), and the BMAa ($p=0.053$). For females tested in their home cage, Fos induction in the BMAp and BLAp were not correlated, but BLAp Fos induction was correlated with the BLAa ($p=0.01$) and the BMAa ($p=0.02$). Collectively, these results suggest that the BLAp and the BMAp are part of a larger sex and context specific network of amygdala nuclei which control consumption after habituation to novelty.

Disclosures: Z.R. Irving: None. **E.M. Greiner:** None. **G.D. Petrovich:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.14

Topic: G.02. Reward and Appetitive Learning and Memory

Title: A novel neural pathway for neurotensin mediated regulation of hedonic feeding behavior and obesity

Authors: *A. J. TOSE[1], N. GAZIT SHIMONI[1], C. SENG[2], Y. JIN[3], T. LUKACSOVICH[2], H. YANG[5], J. P. H. VERHAREN[1], C. LIU[1], E. HU[1], L. W. TANG[1], J. READ[1], M. ZHANG[1], B. LIM[6], L. TIAN[4], C. FOLDY[2], S. LAMMEL[1];
[1]Mol. and Cell Biol., Univ. of California, Berkeley, Berkeley, CA; [2]Brain Res. Inst., Univ. of Zurich, Zurich, Switzerland; [3]Ctr. for Neurosci., [4]Biochem. and Mol. Med., Univ. of California, Davis, Davis, CA; [5]Zhejiang Univ., Hangzhou, China; [6]Biol. Sci., UCSD, La Jolla, CA

Abstract: The neuropeptide neurotensin (NTS) and its receptors are widely expressed in the mammalian brain. NTS modulates a wide range of physiological functions including sleep, feeding, blood pressure, body temperature and locomotion. Using patch-seq experiments, we found that NTS is highly expressed in ventral tegmental area (VTA)-projecting nucleus accumbens lateral shell (NAcLat→VTA) neurons. We then leveraged a novel neurotensin sensor (ntsLight1.1) to confirm that NTS is released in the NAcLat →VTA pathway. While we previously demonstrated that the NAcLat→VTA pathway represents a key circuit node of the brain's reward system (Yang et al., 2018; *Neuron*), the function of NTS in this pathway is entirely unknown.To further study the role of the NAcLat→VTA pathway underlying behavior, we performed *in vivo* electrophysiological recordings of opto-tagged NAcLat→VTA units to find that neural activity of NAcLat→VTA units is directly correlated with consumption of palatable food, but not regular chow. Consistent with this, we found that optogenetic stimulation of the NAcLat→VTA pathway strongly increased consumption of different types of palatable foods, but not standard chow. Conversely, local infusion of an NTS receptor antagonist into the VTA of freely behaving mice prevented hedonic feeding behavior induced by optogenetic stimulation of the NAcLat→VTA pathway. Intriguingly, in the high fat diet (HFD) obesity mouse model, we found that NTS neurotransmission was reduced in the NAcLat→VTA pathway after at least four weeks of HFD. *In vivo* electrophysiological recording verified in these mice that, unlike during regular diet, NAcLat→VTA unit activity no longer correlated with palatable food consumption. Optogenetic stimulation of the NAcLat→VTA pathway in HFD mice also failed to induce hedonic feeding behavior, however, this effect reappeared when mice returned to a regular diet. Lastly, when we selectively overexpressed NTS in the NAcLat→VTA pathway of mice exposed to HFD, we observed a significant reduction in weight gain compared to control mice. Together, these data reveal an unexpected role for NTS in the NAcLat→VTA pathway for promoting hedonic feeding behavior, which is dependent on food environment. These findings contrast the well-known anorexic effects of NTS in the lateral hypothalamus suggesting that circuit-specific manipulations of NTS neurons are critical in order to harness the translational potential of NTS in the treatment of obesity.

Disclosures: A.J. Tose: None. **N. Gazit Shimoni:** None. **C. Seng:** None. **Y. Jin:** None. **T. Lukacsovich:** None. **H. Yang:** None. **J.P.H. Verharen:** None. **C. Liu:** None. **E. Hu:** None. **L.W. Tang:** None. **J. Read:** None. **M. Zhang:** None. **B. Lim:** None. **L. Tian:** None. **C. Foldy:** None. **S. Lammel:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.15

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH R01-DA042889
NIH R01-MH123246
NIH R01-GM127468
One Mind Foundation 047483
ZonMW Rubicon Fellowship

Title: From Laboratory to Wild Mice: A Conserved Mechanism for Adaptive Behaviors in the Dopamine System

Authors: *J. P. H. VERHAREN, K. SAMEL, J. W. DE JONG, A. J. TOSE, E. TAN, M. W. NACHMAN, S. LAMMEL;
Univ. of California Berkeley, Berkeley, CA

Abstract: Adaptive behavior is key for survival in the adverse conditions of the real world. Yet, neurobiological studies are typically conducted in laboratory animals that are born and raised in artificial environments deprived of natural dangers. Here, we compared five different strains of laboratory mice (C57Bl/6J, BALB/cJ) and wild mice (captured in Brazil, Canada, USA and subsequently bred in our laboratories) in identical behavioral assays to find that they exhibit a diverse range of variability in both exploration and escape behaviors. This raises two key questions: (i) are the same neural mechanisms shared between animals raised in drastically different environments and if so, (ii) what neural modulation can account for the observed behavioral variability? We find that exploration behavior across laboratory and wild mice can be predicted based on dopamine receptor expression levels in the medial nucleus accumbens (NAcMed). Using dLight recordings in wild and laboratory mice, we demonstrate that re-exposure to an adverse environment suppresses dopamine release in the NAcMed. Using chemogenetics, we further show that behavioral variability across mouse strains results from divergent activation levels of dopamine D1- and D2 receptor-expressing NAcMed neurons. Finally, we propose a mechanistic model in which behavioral variability can be conceptualized based on the predation risk in the animals' recent environment. Together, these findings reveal a unifying framework of dopaminergic modulation that regulates exploration behavior under varying environmental conditions across laboratory and wild mice.

Disclosures: J.P.H. Verharen: None. **K. Samel:** None. **J.W. de Jong:** None. **A.J. Tose:** None. **E. Tan:** None. **M.W. Nachman:** None. **S. Lammel:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.16

Topic: G.02. Reward and Appetitive Learning and Memory

Title: Value and temporal derivative encoding in midbrain dopamine neurons

Authors: Y. LIANG, *J. W. DE JONG, J. P. H. VERHAREN, S. LAMMEL;
Dept. of Mol. and Cell Biol. and Helen Wills Neurosci. Inst., Univ. of California Berkeley, Berkeley, CA

Abstract: Ventral tegmental area (VTA) dopamine (DA) neurons are often thought to uniformly encode temporal-difference (TD) reward prediction errors (RPEs), which is a derivative-like learning signal that tracks the rate-of-change of an underlying value function. Conversely, DA release in the nucleus accumbens (NAc), the most prominent projection target of these neurons, has been implicated in a broad range of functions, including value-like signals such as "motivation", "wanting" and "value of work". The contrasting nature of value encoding in DA release versus derivative encoding in DA cell body activity and the fact that DA projections to distinct NAc subregions mediate diverse behavioral functions (de Jong et al., 2019; *Neuron*), raises the question of how DA cell body activity and DA release in different mesoaccumbal pathways contribute to reward learning and motivated behavior. To address this question, we developed a novel approach for large-scale recordings of genetically identified DA neurons integrating Neuropixels electrode arrays with optogenetics. We then used this approach to study neural activity patterns of DA neurons in distinct VTA subregions in mice performing a reward-seeking task. Using the same task, we conducted fiber photometry-based recordings of DA neuron activity and DA release that take the precise topographical organization of the mesoaccumbal DA system into consideration. Our results show that DA neurons in the medial VTA encode a value-like signal, while DA neurons in the lateral VTA encode TD RPEs. Furthermore, we demonstrate that within anatomically defined mesoaccumbal pathways, DA activity translates into DA release during behavior, and we provide evidence that VTA DA cell body activity is sufficient to explain DA release in different NAc subregions. Lastly, we developed a simple computational model to demonstrate that different patterns of VTA DA cell body activity and DA release can easily be conceptualized in the context of value and derivative encoding. Together, these findings reveal a framework of DA function underlying reward learning and motivated behavior that recognizes the circuit-specific specialization of VTA DA neurons.

Disclosures: Y. Liang: None. **J.W. De Jong:** None. **J.P.H. Verharen:** None. **S. Lammel:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.17

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH 1R01DA042889
TRDRP 26IP-0035
One Mind Foundation 047483
NARSAD Young Investigator 23543
Brain Research Foundation BRFSG-2015-7

Title: An inhibitory brainstem input to dopamine neurons encodes nicotine aversion

Authors: *C. LIU[1], A. J. TOSE[1], J. P. H. VERHAREN[1], Y. ZHU[2], J. W. DE JONG[2], J. X. DU[4], K. T. BEIER[5], S. LAMMEL[3];
[1]UC Berkeley, Berkeley, CA; [2]Univ. of California Berkeley, [3]Univ. of California Berkeley, Berkeley, CA; [4]Univ. of California San Diego, San Diego, CA; [5]Univ. of California Irvine, Irvine, CA

Abstract: Nicotine stimulates the dopamine (DA) system, which is essential for its rewarding effect. Nicotine is also highly aversive at high doses, yet our knowledge about nicotine's dose-dependent effects on DA circuits remains limited. Here, guided by computational modeling, we performed *in vivo* fiber photometry and pharmacological manipulations in mice to demonstrate that high doses of nicotine, which induce aversion-related behavior, cause biphasic excitatory and inhibitory responses in ventral tegmental area (VTA) DA neurons that can be dissociated by distinct projections to medial and lateral nucleus accumbens (NAc) subregions, respectively. Mechanistically, the inhibitory effects of aversive nicotine involve desensitization of α4β2 and activation of α7 nicotinic acetylcholine receptors. Additionally, we identify α7-dependent activation of upstream GABA neurons in the brainstem laterodorsal tegmentum as a key regulator of heterogeneous DA release in the NAc following aversive nicotine. Together, our findings provide a mechanistic circuit-level understanding of nicotine's dose-dependent effects on reward and aversion.

Disclosures: C. Liu: None. **A.J. Tose:** None. **J.P.H. Verharen:** None. **Y. Zhu:** None. **J.W. de Jong:** None. **J.X. Du:** None. **K.T. Beier:** None. **S. Lammel:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.18

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH Grant R01-DA045783

Title: Anticipation of reward selectively enhances accumbens D1+ neuron firing to promote work

Authors: *T. W. FAUST[1], V. L. COLLINS[3], A. MOHEBI[2], M. DUHNE[4], J. D. BERKE[2]; [1]UCSF, [2]Neurol., UCSF, San Francisco, CA; [3]Neurosci., UCLA, Minneapolis, MN; [4]Neurol., Univ. of California San Francisco, San Francisco, CA

Abstract: Dopamine in the nucleus accumbens (NAc) Core is critical for decisions to work: to engage in prolonged and/or effortful behavior to obtain rewards. NAc projection neurons express either dopamine D1 receptors or D2 (and adenosine A2a) receptors, and these "direct" and "indirect" subpopulations are broadly thought to facilitate and discourage motivated behavior respectively. However, recording studies to date have provided only limited and equivocal support for this hypothesis. We examined the spiking of optogenetically-identified, individual D1+ neurons (n=346) and A2a+ neurons (n=212) in in the NAc Core of freely-moving, transgenic D1-Cre and A2a-Cre rats (n= 4, 5; Pettibone et al. 2019). Rats performed an operant "bandit" task (Hamid et al. 2016), in which variability in reward rate influenced their motivation to initiate a trial by approaching and entering a nose poke port. During approach, spiking increased in a substantial fraction (27.9, 34.2%) of both D1+ and A2a+ neurons. However, in D1+ neurons, but not A2a+, this spiking was greater when more recent trials had been rewarded. This altered spiking coincided with a fast ramp in NAc Core dopamine activity, measured with dLight1.3b (n=10 rats; Mohebi et al. 2019), which also scaled with reward rate. Both dopamine and D1+ modulation by reward history did not depend on the specific choices made during each trial, and therefore appear closer to encoding "state value" rather than "action" or "chosen" values. Local NAc infusions of either the non-selective dopamine antagonist flupentixol (10 µg/0.5 µl) or the selective D1 antagonist SCH-23390 (1 µg/0.5 µl) caused fewer trials to be initiated, with longer approach latencies (n=6 rats). These results provide novel lines of evidence in favor of a vital, specific role for dopamine acting at NAc D1+ neurons in the motivation to initiate work.

Disclosures: T.W. Faust: None. **V.L. Collins:** None. **A. Mohebi:** None. **M. Duhne:** None. **J.D. Berke:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.19

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH Grant MH10697
CHDI

Title: Altered basal ganglia output during self-restraint

Authors: B.-M. GU[1], *J. BERKE[2];
[1]Dept. of Neurol. and Neurolog. Sci., Stanford, Stanford, CA; [2]Neurol., UCSF, San Francisco, CA

Abstract: Our capacity for behavioral inhibition is considered central to cognitive control and is compromised in a range of neurological and psychiatric disorders. Behavioral inhibition can be 'reactive' - e.g. aborting imminent actions in response to a Stop signal - or 'proactive' - e.g. restraining actions in anticipation of a possible Stop signal. Reactive inhibition has been shown to involve fast cue responses in frontal cortex and basal ganglia pathways, including from the subthalamic nucleus (STN) to the basal ganglia output nucleus substantia nigra pars reticulata (SNr) (Schmidt et al. 2013). The underlying mechanisms of proactive inhibition are less well understood, but have been proposed (Aron 2011) to involve the pathway from striatum via globus pallidus pars externa (GPe) "indirectly" to SNr. To examine how changes in basal ganglia output contribute to self-restraint, we recorded SNr neurons (n=619, from 10 adult male Long-Evans rats) during a proactive behavioral inhibition task (Gu et al., 2020). Rats responded to Go! cues with rapid leftward or rightward movements, but also prepared to cancel one of these movement directions, for which a Stop! cue might occur. This action restraint - visible as direction-selective slowing of reaction times - altered both rates and patterns of SNr spiking. Overall firing rate was elevated before the Go! cue, and this effect was driven by a subpopulation of direction-selective SNr neurons. In neural state space, this corresponded to a shift away from the restrained movement. Furthermore, SNr activity showed an increase in spike variability during proactive inhibition. Increased spike variability corresponded to variable state-space trajectories, slowing reaction times via reduced preparation to move. These findings open new perspectives on basal ganglia mechanisms for movement preparation and cognitive control, at a single cell level and also in population dynamics.

Disclosures: B. Gu: None. **J. Berke:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.20

Topic: G.02. Reward and Appetitive Learning and Memory

Title: Acute changes in the spiking of identified dopamine neurons produced by fentanyl in unrestrained rats.

Authors: *L. T. VINSON[1], A. MOHEBI[2], J. D. BERKE[2];
[1]Univ. of California, San Francisco, Oakland, CA; [2]Neurol., UCSF, San Francisco, CA

Abstract: The ability of abused drugs to enhance dopamine (DA) release in the ventral striatum is critical for their reinforcing properties. For opioids such as fentanyl, this enhanced DA release is thought to arise from actions at mu-opioid receptors on GABAergic neurons that provide

inhibition to DA neurons in the ventral tegmental area (VTA). Drug-suppressed firing of GABAergic neurons then produces increased firing of DA neurons through disinhibition. However, this model is based primarily on studies conducted in brain slices and under anesthesia—there have been no electrophysiological recordings of identified VTA DA neurons in awake behaving animals. Furthermore, it is unknown whether enhanced DA release reflects drug-induced changes in DA cell tonic firing rates, increased bursting, or both. We used optrodes (tetrodes surrounding an optic fiber) to record *in vivo* extracellular activity of VTA neurons in freely-moving male and female TH-Cre rats before and after subcutaneous fentanyl administration (20, 40µg/kg or saline). DA neurons were identified using viral-mediated, Cre-dependent expression of the opsin Chrimson, with 10ms red light pulses to evoke firing in Chrimson-expressing neurons. In initial results, the lower fentanyl dose increased locomotor activity, and the higher dose suppressed locomotor activity. Still, both doses consistently increased the firing rate of opto-tagged DA neurons (n=6), as expected. This increase appeared to reflect a broad shift in the inter-spike-interval histogram, rather than a specific increase in burst firing. Interestingly, other cells (n=14) presumed to be non-dopaminergic (based on higher firing rates and lack of light response) instead decreased their firing rate, often dramatically, following fentanyl administration. These preliminary results provide the first awake, *in vivo* support for a long-standing model of opioid influence over DA neuron firing.

Disclosures: L.T. Vinson: None. **A. Mohebi:** None. **J.D. Berke:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.21

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH Grant R01DA045783
NIH Grant R01NS123516
NIH Grant R01NS116626
NIH Grant R21AA027157
NIH Grant K01MH126223

Title: A Spectrum of Time Horizons for Dopamine Signals

Authors: *A. MOHEBI, W. WEI, J. D. BERKE;
UCSF, UCSF, San Francisco, CA

Abstract: Dopamine (DA) input to the striatum can encode reward prediction error (RPE), a critical signal for updating predictions of future rewards. However, it is unclear how this mechanism handles the need to make predictions, and provide feedback, over multiple time horizons: from seconds or less (if singing a song) to potentially hours or more (if hunting for food). We monitored dopamine dynamics in three distinct striatal subregions (dorsolateral-DLS,

dorsomedial-DMS, ventral-VS) using fiber photometry of fluorescent sensor dLight1.3b in n=20 adult rats. Spontaneous DA fluctuations monitored simultaneously across subregions showed very distinct dynamics. DLS DA changed rapidly and constantly while VS DA evolved more slowly, with rare large transient increases. The evoked DA response to an unpredicted reward cue was briefest in DLS and lasted longer in VS. We then studied the DA response to this reward cue when it was more- or less-expected, within an instrumental bandit task. In all subregions, the DA pulse was greater when fewer recent trials had been rewarded, consistent with positive RPE coding. However, the underlying reward prediction differed between subregions. We used a leaky integrator model to estimate the reward rate, and varied the decay parameter tau to assess the time scale over which reward history is integrated to predict future rewards. We found the strongest correlation to RPE using the longest integration times for VS DA (tau > 800s) and the briefest for DLS DA (tau < 200s). We then examined DA responses within a Pavlovian approach task with three distinct auditory cues (75,25,0% predictive of reward ~3s later) and found remarkably distinct DA responses in each subregion. DMS DA signals showed strong RPE coding, with a large pulse to 75%, small to 25%, and negative to 0% cues. VS DA discriminated between cues more slowly and weakly, with the response to all cues remaining positive. DLS showed weak responses to all cues. Using recurrent neural network models, we found we could reproduce these response patterns simply by discounting future rewards at different rates in different subregions. The resulting spectrum of time horizons for value computations can help achieve efficient learning and adaptive motivation for a wide range of behaviors.

Disclosures: **A. Mohebi:** None. **W. Wei:** None. **J.D. Berke:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.22

Topic: G.02. Reward and Appetitive Learning and Memory

Title: Dopamine value signals propagate through space during foraging

Authors: *T. A. KRAUSZ[1], A. E. COMRIE[1], A. E. KAHN[3], L. M. FRANK[5], N. D. DAW[4], J. D. BERKE[2];
[2]Neurol., [1]UCSF, San Francisco, CA; [4]Princeton Neurosci. Inst., [3]Princeton Univ., Princeton, NJ; [5]Dept. of Physiol., UC San Francisco, San Francisco, CA

Abstract: How do our brains use past experiences to predict future rewards, despite long gaps in space and time? Candidate algorithms are provided by Reinforcement Learning (RL) theory, but how closely these correspond to brain processes is not well understood, especially in deep, multistep decision problems such as spatial navigation. RL variables can be encoded by ventral striatum dopamine (DA) signals, including pulses that scale with reward prediction error (RPE), and ramps during reward approach that scale with value (reward prediction). Yet few studies have examined how these signals evolve within naturalistic environments, and how this sheds

light upon underlying algorithms. We examined DA dynamics using dLight photometry as rats (n=8) foraged for reward in a novel spatial task, the Hex Maze. This task varies both the probabilities of receiving reward, and the available paths to the reward locations. As expected, we observed clear positive RPE coding when rewards were obtained; unexpectedly, we also observed large positive transients when rats detected that previously blocked paths were now available. As rats ran, DA also ramped up in proportion to the spatially discounted value of their destination, and we compared the trial-to-trial evolution of this signal to RL models. Initial analyses indicate that ramping DA value signals are updated by at least two distinct processes: 1) During spatial navigation, value propagates progressively and locally along a rat's taken path, chaining backwards across successive trials as if using a temporal-difference algorithm; 2) Between trials, rewards or omissions update values throughout the maze - both on the taken path and elsewhere - consistent with model-based learning. Our results provide evidence for a dopaminergic value signal updated through two computationally-distinct mechanisms.

Disclosures: **T.A. Krausz:** None. **A.E. Comrie:** None. **A.E. Kahn:** None. **L.M. Frank:** None. **N.D. Daw:** None. **J.D. Berke:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.23

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH Grant F32DA052283
NIH Grant R01NS116626

Title: Homeostatic state-dependent regulation of dopamine neuron activity

Authors: *A. A. MAMALIGAS[1,6], Z. A. KNIGHT[1,2,7], J. D. BERKE[3,4,2,5], A. C. KREITZER[6,8]; [1]Physiol., [2]Kavli Inst. for Fundamental Neurosci., [3]Neurol., [4]Psychiatry and Behavioral Sci., [5]Weill Inst. for Neurosciences, Univ. of California, San Francisco, San Francisco, CA; [6]J. David Gladstone Inst., San Francisco, CA; [7]Howard Hughes Med. Inst., Chevy Chase, MD; [8]MapLight Therapeutics, Inc, Palo Alto, CA

Abstract: Animals seek rewards based on their needs. Imbalances in internal homeostatic state that occur over prolonged time courses, causing the sensations of hunger or thirst, drive reward-seeking behavior. Interoceptive neurons governing hunger and thirst have been recently discovered in the arcuate nucleus of the hypothalamus (ARC) and the subfornical organ, and stimulation of these cells drives animals to eat or drink, respectively. The mesolimbic dopamine (DA) system, projecting from the ventral tegmental area (VTA) to the nucleus accumbens, also regulates reward-seeking behavior. However, the circuit mechanisms linking activity in interoceptive cells to DA neuron activity remain unclear. To determine how DA neurons integrate information about homeostatic state to drive specific activity patterns, we use a

combination of optical calcium sensors, bulk and single neuron resolution imaging tools, and chemogenetic manipulation to measure DA activity in the ventral midbrain in hungry or thirsty mice. To test whether increased homeostatic need selectively boosts DA neuron activity based on the available reward, we used a freely-moving cued approach tone discrimination task in which food- and water-predictive cues were randomly delivered. We performed either fiber photometry or single-neuron resolution imaging of DA neuron calcium activity using a head-mounted mini-microscope (nVista, Inscopix). We found that DA neuron activity was selectively boosted for the deprived cue (e.g. the food-predictive cue produced a larger amplitude calcium transient in food deprived mice than did the water-predictive cue). This pattern was observed in both the bulk DA neuron signal as well as at the single neuron level, suggesting convergent signals from upstream interoceptive neurons in driving DA activity rather than separate circuits. Surprisingly, chemogenetic activation of hunger neurons in the ARC, creating a false perception of caloric imbalance, equally boosted DA neuron responses to both food- and water-predictive cues. This suggests that, while activation of ARC hunger neurons drives food seeking, in natural hunger, additional circuit mechanisms may be required to achieve selectivity of DA neuron responses to food cues.

Disclosures: **A.A. Mamaligas:** None. **Z.A. Knight:** None. **J.D. Berke:** None. **A.C. Kreitzer:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.24

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH Grant 5R01DA045783-05
Mariana Duhne Aguayo, Ph.D. is a is a Latin American Fellow in the Biomedical Sciences, supported by The Pew Charitable Trusts.
SECTEI/154/2021

Title: Relationships between cholinergic interneuron spiking and dopamine release in freely behaving rats

Authors: *M. DUHNE[1], J. R. PETTIBONE[2], A. MOHEBI[3], T. W. FAUST[3], J. D. BERKE[3]; [1]Univ. of California San Francisco, San Francisco, CA; [2]Neurol., Univ. of Mchigan, San Francisco, CA; [3]Neurol., UCSF, San Francisco, CA

Abstract: The striatum is an essential structure for adapting behavior to changing reward conditions. Key modulators of striatal function include dopamine (DA), provided by projections from midbrain neurons, and acetylcholine (ACh), predominantly provided by local cholinergic (ChAT+) interneurons. These two modulators influence each other's release, and their dynamic interplay is thought to critically control striatal plasticity mechanisms. Salient cues have been

reported to evoke a pause in the activity of striatal "tonically-active neurons" (TANs) - presumed to be ChAT+ - that coincides with a burst of midbrain DA cell firing signaling reward prediction errors. Yet no studies have reported the firing patterns of identified ChAT+ or their relationships to DA release. We have been optogenetically-tagging and comparing several classes of striatal interneurons: ChAT+ (n=45), parvalbumin-positive (PV+, n=5), and somatostatin-positive (SST+, n=4), in unrestrained rats performing instrumental and Pavlovian tasks. We also recorded local dopamine release during the same tasks in the same striatal subregions with the fluorescent sensor dLight1.3b. All three cell types were consistently tonically-active, confirming that tonic activity alone is a poor guide to cell type, and spike waveform was also insufficient for identification. In dorsal-lateral striatum ChAT+ neurons reliably showed classic TAN patterns, including a brief pause to a Go! cue that simultaneously evoked a local DA pulse. However, this relationship varied by subregion and behavioral event. In particular, in ventral striatum (nucleus accumbens) we found that a reward cue evoked simultaneous increases in both ChAT+ spiking and DA release. These results demonstrate that 1) unequivocal cell type identification is important for deciphering striatal microcircuit functions, and 2) striatal models should not simply assume that ChAT+ neurons pause throughout striatum in conjunction with phasic DA release.

Disclosures: M. Duhne: None. **J.R. Pettibone:** None. **A. Mohebi:** None. **T.W. Faust:** None. **J.D. Berke:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.25

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH Grant R01MH128669

Title: Ambiguity attitudes are dependent on reward magnitude

Authors: *K. M. ROTHENHOEFER[1], A. ALIKAYA[1], C. MASSOT[3], W. R. STAUFFER[2]; [2]Neurobio., [1]Univ. of Pittsburgh, Pittsburgh, PA; [3]CNBC, Carnegie Mellon Univ., Pittsburgh, PA

Abstract: Decision theory recognizes two forms of uncertainty: risk - in which the underlying probability distributions are known, and ambiguity - in which the underlying probability distributions are not known. Many decision-making theories based on risk, such as expected utility theory, fail to adequately describe real-world choice behavior. This failure partly reflects the fact that pure risk is rare: it is only encountered in casinos, coin-flips, and decision-making experiments. In most real-world cases, incomplete information, sparse data, and cognitive limitations create various states of ambiguity. Therefore, ambiguity, rather than risk, better describes the conditions of uncertainty under which most real-world decision-making occurs. Prior work has demonstrated that nonhuman primates (NHPs) demonstrate ambiguity aversion

during choices between risky and ambiguous stimuli. Here, we investigate how decision makers assign value to ambiguous options. We developed a novel NHP economic decision-making task with independent control over the potential outcome numbers, magnitudes, and probabilities. As with prior NHP studies of risky decision making, we observed risk-seeking tendencies for low magnitude outcomes, and decreased risk seeking tendencies with increased reward magnitudes. On a subset of trials, the outcome probability information was partially or fully obscured to create ambiguity in the outcome probability distribution. During ambiguity trials with small reward sizes, we observed ambiguity-seeking tendencies: the animals selected ambiguous options over safe rewards with similar values. With larger rewards, the trend was reversed: the animals selected safe outcomes over ambiguous options. Thus, NHP uncertainty attitudes under conditions of ambiguity were dependent on the potential reward magnitudes. In addition to the choice data, we analyzed pupil diameter and found that pupil dilation was less sensitive to the reward magnitudes following ambiguous cues, even at larger reward sizes. Pupil diameter has been previously shown to reflect learning rate and prediction error. Thus, the decreased sensitivity to the outcomes of ambiguous predictions indicates that those predictions might not be quickly updated. Together, these observations suggest that uncertainty itself plays a significant role in the valuation of risky and ambiguous options, but also that fundamental distinctions exist between these behavioral contexts. These results provide the basis for further behavioral and neuronal characterizations of decision making under ambiguity.

Disclosures: K.M. Rothenhoefer: None. **A. Alikaya:** None. **C. Massot:** None. **W.R. Stauffer:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.26

Topic: G.02. Reward and Appetitive Learning and Memory

Support: University of Pittsburgh Brain Institute
NIH DP2MH113095

Title: Contextual influences on risky decision making

Authors: *W. KERKHOFF, W. STAUFFER;
Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Effective economic decision making maximizes rewards and minimizes costs. However, utility models and other normative frameworks that follow utility maximization fail to capture or predict the full range of choice variabilities and decision makers' idiosyncrasies. Emotions have long been recognized as significant factors driving decision makers to depart from pure economic rationality; John Maynard Keynes recognized these "animal spirits" as influencing individual behavior and even postulated them as a source of broad economic

instability. Despite this, we do not know how emotional states and economic variables are integrated in the brain. To this end, we have constructed a contextual economic decision-making task intended to examine how emotional contexts influence risky choices in nonhuman primates. One NHP (rhesus macaque) was trained to predict rewards from informative visual stimuli that indicated both the magnitude and probability of all reward potential outcomes. Behavioral choice testing demonstrated that the decisions between two options were consistent with first order stochastic dominance. Thus, the animal properly inferred the values from the stimuli. Moreover, the animal's choices reflected similar risk attitudes as previously described. Simultaneously, we have established a custom face expression recording system to map rhesus macaque face expressions in different emotional/reward contexts. Preliminary data suggest that facial action units are sensitive to reward parameters and contexts. These results set the stage for investigating how well-defined emotional states influence the evaluation of risk, and ultimately, how emotional and economic variables are combined to generate behavior.

Disclosures: W. Kerkhoff: None. **W. Stauffer:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.27

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH DP2MH113095
NIH/NIMH R01MH128669

Title: Combinatorial algorithms guide economic algorithms

Authors: *T. HONG[1], W. R. STAUFFER[2];
[1]Carnegie Mellon Univ., Pittsburgh, PA; [2]Neurobio., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Economic decisions rarely have 'correct' responses. Instead, decision makers must deliberate and determine the most valuable option prior to making a choice. This process of deliberate consideration, or deliberation, can be computationally demanding values are often dependent on combinations of factors including context, alternatives, and internal states. To investigate the psychological and neural processes for managing computationally complex deliberations, we developed the 'knapsack task' based on the eponymous problem from computer science. The objective of each knapsack trial was to select subsets of items presented on a touchscreen in order to maximize juice reward, without exceeding a fixed limit of 0.8 ml. To understand how the animals optimized, we categorized their solutions according to the solutions' proximity to established computer algorithms. The animals exhibited a variety of strategies, including those that require combinatorial reasoning. Remarkably, we could predict what algorithm an animal would use, based on the other animal's behavior. The high correlations between the algorithms the two monkeys used for a given instance suggest that the application of

combinatorial reasoning was not random and likely determined by the instances. A key difference between the algorithms is the number of operations performed before the first selection. For example, when there are 5 items, greedy search only needs to identify the item with highest value and hence requires 5 operations, whereas Sahni-*k* algorithm considers all possible *k*-item combinations and hence requires at least 5 choose *k* operations. In both animals, the deliberation times preceding the first selection, and subsequent inter-selection intervals, reflected the number of operations prescribed by the corresponding algorithms at each step. These results provide strong evidence that the animals adapted algorithmic strategies and employed combinatorial reasoning to manage complex deliberations. Recurrent neural networks (RNNs) allowed us to probe the neural representations during the deliberation period. Decoding analysis suggested that RNNs mimicking the combinatorial algorithms serially represented multiple combinations during deliberation, whereas the greedy RNNs lacked such dynamic representations. Accordingly, causal manipulations revealed that RNNs performing combinatorial comparisons required longer deliberation to achieve target performance. These results establish a new behavioral paradigm for investigating the psychological and neural basis for combinatorial optimization and economic deliberation.

Disclosures: **T. Hong:** None. **W.R. Stauffer:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.28

Topic: G.02. Reward and Appetitive Learning and Memory

Title: Cholinergic modulation of context-gated reward prediction

Authors: *J. CHAVIRA[1], S. PETERSON[1], A. MAHERAS[2], R. KEIFLIN[1];
[1]Psych. & Brain Sci., [2]Molecular, Cell. & Developmental Biol., UC Santa Barbara, Santa Barbara, CA

Abstract: Hierarchical contextual control of associative memories, or context gating, is critical for adaptive behavior; it allows animals to derive context-dependent predictions from certain stimuli and engage in context-appropriate behavior. Acetylcholine (ACh) neurotransmission is known to be involved in several cognitive functions, however its role in the context gating of associative memories remains unexplored. Here we investigated the effects of systemic manipulations of ACh transmission on context-gated reward prediction. Three groups of rats were trained in a Pavlovian approach procedure, in which two brief auditory cues (X and Y) were presented in two different visual context (A or noA) and possibly followed by reward. The reward contingencies differed between groups. For a first group the reward-predictive status of each cue was informed by the background context, so that cue X was rewarded only in the context A while cue Y was rewarded only in the context noA; all rewarded trials resulted in the same outcome. Discriminated responding in this task critically relies on contextual gating. A

second group of rats was trained with similar contingencies with the exception that the two rewarded trial types resulted in two different food outcomes. In this group, context-dependent behavior could be facilitated by direct associations between contexts and their corresponding outcomes. Finally, for a third group, cue X was always rewarded and cue Y was never rewarded, regardless of context. In this group, reward prediction is largely independent of the background context. Following acquisition of discriminated performance, we assessed the effect of an acute treatment with the cholinergic drugs oxotremorine (muscarinic agonist) and nicotine, and the anticholinergic drugs scopolamine (muscarinic antagonist) and mecamylamine (nicotinic antagonist; saline + 4 doses of each drug). Low doses of scopolamine profoundly disrupted context-dependent discrimination performance, particularly in rats trained with a single outcome —the only form of discrimination that unambiguously relies on context-gated predictions. In contrast, scopolamine had little effect on simple discrimination performance. Oxotremorine, nicotine, and mecamylamine did not affect discrimination performance in any group. These results suggest that context-gated and context-independent reward predictions rely on partially dissociable neural substrates and that cholinergic transmission at muscarinic receptors is particularly critical for the contextual control of associative reward predictions.

Disclosures: **J. Chavira:** None. **S. Peterson:** None. **A. Maheras:** None. **R. Keiflin:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.29

Topic: G.02. Reward and Appetitive Learning and Memory

Support: UCSB Academic Senate Grant

Title: Dissociable roles of Orbitofrontal cortex and dorsal Hippocampus in context-gated reward prediction

Authors: S. PETERSON[1], J. CHAVIRA[1], A. MAHERAS[2], A. GARCIA ARANGO[1], E. SEAMANS[1], ***R. KEIFLIN**[1];
[1]Psychological & Brain Sci., [2]Molecular, Cell. & Developmental Biol., UC Santa Barbara, Santa Barbara, CA

Abstract: Contextual control over cue-evoked appetitive behavior is essential for adaptive behavior. The orbitofrontal cortex (OFC) and the dorsal hippocampus (DH) have been shown to encode appetitive cues in a context-dependent manner. However, the specific contribution of these regions in the contextual regulation of cue-evoked reward seeking remains uncertain. Here we used chemogenetic silencing to interrogate the necessity of the OFC and the DH in the expression and acquisition/generalization of contextual rules for cue-evoked reward seeking. Rats were assigned to 2 behavioral training conditions: context-gated, or simple Pavlovian discrimination. In the context-gated discrimination task the validity of two brief auditory cues (X

and Y) to signal reward depended on the contextual visual background (Cx.A or Cx. noA), so that cue X was rewarded only in the context A but not in the context noA, while the opposite was true for cue Y (A:X+ / X- / A:Y- / A:Y+). In the simple discrimination task, only one cue was rewarded regardless of context (A:X+ / X+ / A:Y- / A:Y-). For each training conditions, rats were made to express the inhibitory DREADD hM4di, or the control transgene mCherry, in the lateral OFC or the DH. OFC silencing (achieved by systemic injection of the DREADD ligand J60) disrupted context-gated but not simple discrimination performance. DH silencing had no effect in either task. To investigate OFC and DH contribution to the acquisition of contextual rules, we took advantage of the learning transfer effects predicted to occur in these tasks. The same rats, previously trained in context-gated or simple discrimination task, were introduced with a new cue (Z). Critically this cue was presented in a single context and always rewarded (A:Z+). Finally, responding to that cue was tested in both contexts. Consistent with the predictions of computational models, rats previously trained in the context-gated task showed preferential responding to that new cue when presented in its training context. In other words, these rats generalized an existing contextual rule to incorporate a new association. Silencing DH during this new learning episode had no effect on the development of conditioned responding but prevented the assimilation of this new association into existing contextual rule, as revealed by similar levels of responding to that new cue in both contexts during the final probe test. These results indicate specialized roles of OFC and DH in contextual control. Specifically, OFC critically contributes to the behavioral expression of contextual rules for cue-evoked predictions and the DH contributes the integration of new associations into existing contextual rules.

Disclosures: S. Peterson: None. **J. Chavira:** None. **A. Maheras:** None. **A. Garcia Arango:** None. **E. Seamans:** None. **R. Keiflin:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.30

Topic: G.02. Reward and Appetitive Learning and Memory

Title: Sex differences in context-gated reward predictions

Authors: *A. L. MAHERAS[1], S. PETERSON[2], J. I. CHAVIRA[2], R. KEIFLIN[2];
[1]Molecular, Cell. & Developmental Biol., [2]Psychological & Brain Sci., Univ. of California, Santa Barbara, Santa Barbara, CA

Abstract: When faced with ambiguous stimuli, organisms often rely on the background circumstances —the context— to interpret these stimuli and generate situation-appropriate predictions. Impaired context processing is a core deficit in several psychiatric disorders (schizophrenia, autism, PTSD). Importantly, sex differences in the prevalence and symptomatology of these disorders are well-established, suggesting potential sex differences in contextual processing. Here, we compared male and female rats as they learned a contextual rule

for reward prediction (the background context informing the validity of different reward cues).Rats were trained in a context-gated Pavlovian discrimination task in which the validity of two brief auditory cues (X and Y) to signal reward depends on the contextual visual background (Cx. A or Cx. noA). Specifically, cue X was rewarded only in the context A but not in the context noA, while the opposite was true for cue Y (A:X+ / X- / A:Y- / A:Y+). To control for nonspecific effects, we trained two additional groups in tasks that do not require contextual processing: simple discrimination (only one cue rewarded regardless of context: A:X+ / X+ / A:Y- / A:Y-) and no discrimination (both cues rewarded probabilistically regardless of context). After initial learning, the effect of an acute restrain test on performance was assessed for all rats. Critically, we observed significant sex differences only in the context-dependent discrimination task. While a majority (72%) of males successfully learned the task, only a minority (43%) of females learned the task after 70 daily sessions. Acute stress disrupted performance only in the context-dependent discrimination task and this effect was more pronounced in males. Therefore, it appears that female rats are slower to learn a context-dependent discrimination task, but once acquired, contextual processing is more robust in females and less sensitive to stress. These results suggest a trade-off between speed and robustness in the acquisition of context-gated predictions, with different distributions of sexes along this Fast - Steady continuum. These sex differences in contextual processing could contribute to the sex biases in certain psychiatric disorders.

Disclosures: **A.L. Maheras:** None. **S. Peterson:** None. **J.I. Chavira:** None. **R. Keiflin:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.31

Topic: G.02. Reward and Appetitive Learning and Memory

Title: Context-gated reward predictions: a c-Fos activation map

Authors: *S. PETERSON[1], J. I. CHAVIRA[1], A. L. MAHERAS[2], B. WU[1], D. E. SEAMANS[1], R. KEIFLIN[1];
[1]Psychological & Brain Sci., [2]Mol. Cell. & Developmental Biol., Univ. of California, Santa Barbara, Santa Barbara, CA

Abstract: Associative learning is generally conceived as the formation of a binary association between two events, so that event #1 automatically evokes the anticipation of event #2. However, animals can also engage in more complex forms of associative learning in which certain (contextual) stimuli acquire hierarchical control over associative memories. This allows for the context-informed interpretation of ambiguous stimuli (e.g., the word ‘apple’ might evoke different representations at the farmers market or the electronic store). Compared to the extensive literature devoted to understanding the neural mechanisms of “simple” Pavlovian predictions, fewer studies have investigated the neural substrates of the hierarchical contextual control of

associative predictions —or context gating. Here we aimed to generate and compare brain-wide activation maps corresponding to these two forms of reward prediction: context-independent and context-gated. Three groups of rats were trained in three different forms of Pavlovian appetitive discriminations. A first group of rats was trained in a context-dependent discrimination task, in which the validity of two brief auditory tones (X and Y) to signal reward was informed by the contextual visual background (A or noA). Specifically, cue X was rewarded only in the context A but not in the context noA, while the opposite was true for cue Y (A:X+ / X- / A:Y- / Y+). A second group was trained in a simple Pavlovian discrimination task in which one cue reliably signaled reward while the other cue was never paired with reward, regardless of the context (A:X+ / X+ / A:Y- / Y-). Finally, for a third group, both auditory cues were ambiguous predictors of reward regardless of the context (A:X+- / X+- / A:Y+- / Y+-). Following acquisition of stable performance in these tasks, we collected the brains from all rats, 120 min after the start of their final behavioral session. We then performed immunofluorescence labelling of NeuN (a general neuronal marker) and cFos (a molecular marker of activity) in selected structures. We observed that these different tasks resulted in partially dissociable neural activation maps, presumably reflecting the different associative or cognitive processes engaged in these tasks. Of critical importance was the claustrum, which showed a diminished activation in context-depended discrimination. This suggests that the claustrum could constitute a critical hub in the context informed processing of incoming stimuli and the contextual hierarchical control of associative reward predictions.

Disclosures: S. Peterson: None. **D.E. Seamans:** None.

Poster

557. Neural Basis of Reward II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 557.32

Topic: G.02. Reward and Appetitive Learning and Memory

Support: 1 P50 MH119467-01A1 (to DAP)

Title: Electrophysiological signatures of reward learning and its modulation by the nociceptin receptor antagonist J-113397

Authors: A. ITURRA-MENA[1,2], B. D. KANGAS[2], O. LUC[2], D. D. POTTER[3], D. A. PIZZAGALLI[2];
[1]Psychiatry Dept., Columbia Univ., New York, NY; [2]McLean Hosp., Harvard Med. Sch., Belmont, MA; [3]Harvard Med. Sch., Boston, MA

Abstract: Background: Major Depressive Disorder (MDD) is associated with impaired reward learning (i.e., the ability to modulate adaptive behaviors as a function of reinforcement history) and preclinical models relevant to depression have implicated increased levels of nociceptin receptors (NOP) in the corticostriatal-midbrain circuitry, opening a new promising pathway for

the treatment of MDD. **Aims:** The aims of this study were 1) to identify the electrophysiological signatures of reward learning using both event related potentials and spectral analyses while rats performed the Probabilistic Reward Task (PRT), and 2) to investigate whether blockade of nociceptin receptors - using the NOP antagonist J-113397 - would modulate behavioral and neurophysiological markers of reward learning. **Methods:** 11 rats (5 females and 6 males) were trained on a rodent touchscreen-based version of the PRT. Once the rats completed the PRT training phase, they were surgically implanted with deep electrodes in the anterior cingulate cortex (ACC) and nucleus accumbens (NAc) for local field potentials (LFP) recordings during the PRT. In each testing/recording session, the rats were injected either with vehicle or the NOP antagonist J-113397 (10 mg/kg), 15 minutes prior to the PRT testing and concurrent recording. **Results:** We identified a negative amplitude deflection in the ACC and NAc electrodes after reward feedback, as well as power increase in feedback-locked 1-5 Hz (delta) at 200-600 ms and 9-17 Hz band at 100-200 ms in both electrodes during rewarded trials. J-113397 had a main effect on feedback-locked delta band power in the NAc and increased the 9-17 Hz band power in ACC for lean rewarded trials, while it did not have an evident effect on the behavioral parameters. **Conclusion:** Using electrophysiology and touchscreen-based technology, we were able to show that the neural mechanisms that regulate reward learning in rats are similar to those in humans. Our study also provides initial insights about the effects of NOP receptor antagonism on reward learning.

Disclosures: A. Iturra-Mena: None. **B.D. Kangas:** None. **O. Luc:** None. **D.D. Potter:** None. **D.A. Pizzagalli:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Brain and Behavior Research Foundation, Dana Foundation, Millennium Pharmaceuticals, NIMH. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Compass Pathways, Engrail Therapeutics, Neumora Therapeutics, Neuroscience Software. F. Consulting Fees (e.g., advisory boards); Albright Stonebridge Group, Boehringer Ingelheim, Compass Pathways, Engrail Therapeutics, Neumora Therapeutics (formerly BlackThorn Therapeutics), Neurocrine Biosciences, Neuroscience Software, Otsuka, Sunovion, Takeda, Psychonomic Society (for editorial work) and from Alkermes.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.01

Topic: G.04. Emotion

Title: Top down emotion-regulation strategies' temporal effects on affect, attentional deployment, and working memory

Authors: *E. BAYKAL, J. M. JAMES, N. KELLING, G. L. MORENO;
Univ. of Houston - Clear Lake, Houston, TX

Abstract: Prior research has elucidated the effectiveness of top-down emotion regulation strategies of cognitive reappraisal (CR) and guided attention (GA) at minimizing negative feelings while also being cognitively demanding. However, the mechanisms underlying these processes are not well understood. The current study uses eye-tracking to explore the temporal effects (4s; 8s) of two top-down emotion regulation strategies (CR; GA) on attentional deployment, working memory load, and emotion regulation effectiveness. 54 participants (M_{age}=25.42±5.01yrs) completed an emotion regulation task while Pupil Labs eye-tracking hardware and software where utilized to observe pupillometry and gaze fixations. During the task, participants implemented CR or GA strategies while viewing negative images then rated their feelings. A 2x2 within-subjects MANOVA was used to examine temporal effects (4s; 8s) of the top-down emotion regulation strategies (GA; CR) on emotion regulation effectiveness (self-reported affect), working memory load (inter-trial change in pupil diameter), and attentional deployment (% of total fixations to negative stimuli). Pillai's trace showed a significant effect of strategy, time, and their interactions on outcomes (V = 0.85, F(6,1912) = 234.69, p < .0001) with a large effect size (η_p^2 = 0.42). Separate univariate ANOVAs revealed that longer duration trials (8s) yielded greater fixations to negative stimuli, while emotion regulation effectiveness was not significantly changed. CR resulted in substantially higher fixations to the negative-emotion stimuli than GA (F(1,956) = 3532.14, p < .001), yet was more effective at regulating emotion (F(1,956) = 7.39, p < .01). On average, participants experienced significantly greater pupil dilation across all conditions (M = 31.64, SE = 0.19) compared to baseline (M = 28.48, SE = 0.17), suggesting top-down emotion regulation's potential impact on working memory load (t(956) = 39.032, p < .0001, r = .78). Longer duration (8s) top-down emotion regulation also increased pupil dilation (F(1,956) = 58.76, p < .001). In conclusion, this work suggests that implementing top-down emotion regulation may increase working memory load, but also sustain emotion regulation effectiveness. A better understanding of the interplay between emotion regulation, attentional deployment, and working memory could reveal individual differences in interpreting and behaviorally responding to emotional stimuli.

Disclosures: E. Baykal: None. **J.M. James:** None. **N. Kelling:** None. **G.L. Moreno:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.02

Topic: G.04. Emotion

Support: the Korea Health Technology R&D Project through the Korea Health Industry Development Institute (KHIDI), funded by the Ministry of Health & Welfare, Republic of Korea (grant number : HI19C1234 to H.J.K.)
Institute for Basic Science grant (no. IBS-R015-D1 to C.-W.W.)

the National Research Foundation of Korea (grant nos. 2019R1C1C1004512, 2021M3E5D2A01022515 and 2021M3A9E4080780 to C.-W.W.)
the KIST Institutional Program (grant no. 2E30410-20-085 to C.-W.W.)

Title: Introspection makes your brain more like you: Periodic thought sampling enhances accuracy of functional connectome fingerprinting and behavior prediction at rest

Authors: *H.-J. KIM[1,2], C.-W. WOO[1,2], E. S. FINN[3];
[1]Inst. for Basic Sci. (IBS), Ctr. For Neurosci. Imaging Res. (CNIR), Suwon-si, Gyeonggi-do, Korea, Republic of; [2]Biomed. Engin., SungKyunKwan Univ. (SKKU), Suwon-si, Gyeonggi-do, Korea, Republic of; [3]Psychological and Brain Sci., Dartmouth Col., Hanover, NH

Abstract: Heterogeneity of brain structure and function has long been recognized, and the ability to predict individual differences in trait variables from brain measures is one of the most critical steps toward individualized neuroscience. Functional connectome fingerprinting is one such approach, which demonstrates that functional connectivity (FC) can serve as a 'fingerprint' to identify each individual from a large group of people. Researchers have shown that FC data collected with naturalistic stimuli (e.g., movie-watching) outperforms resting-state FC in both fingerprinting and behavioral prediction. However, there have been few studies on how introspective thinking shapes FC and its fingerprinting performance though it has been proposed that introspection could effectively induce idiosyncratic brain states. We collected fMRI data from 61 participants while they underwent two types of rest scans: one in which they were asked to report what they were thinking in a few words every 45 seconds ("thought-sampling"), and one pure rest scan without any thought probes. We applied the fingerprinting approach in which the accuracy was defined as the number of within-subject correlations being greater than any other between-subject correlations between two separate runs (thus the chance level was approximately 1/61). The fingerprinting result showed that the thought-sampling scan provided higher fingerprinting accuracies (95.1-100%, permutation $p < 0.001$ against chance) compared to the rest-only scan (88.5-93.4%). This result was consistent in the retest data from a subset of participants ($n = 29$, test-retest interval of 3 months on average). In both the thought-sampling and rest-only runs, functional connections among the medial frontal, frontoparietal, and default mode networks played an important role in distinguishing each individual from others. Lastly, data collected during the thought-sampling scan accurately predicted individual differences in traits measured with multiple behavioral surveys, whereas predictions based on rest-only data were not as accurate. These results suggest that even without external stimuli, a mere change of cognitive states that enhance introspection can induce individually unique FC patterns. Overall, we show that adding intermittent thought probes to rest scans to enhance introspection produces more FC-based accurate predictions of self-identity and trait-like features compared to pure rest, providing a possibility of using thought sampling not only in healthy populations but also in clinical populations given that it is a simple but potent method for capturing individually distinct FC features.

Disclosures: H. Kim: None. **C. Woo:** None. **E.S. Finn:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.03

Topic: G.04. Emotion

Title: Acute effects of slow breathing on emotion regulation

Authors: *L. KURDZIEL, L. MCDEVITT;
Merrimack Col., Merrimack Col., North Andover, MA

Abstract: Being able to regulate emotions is critical to coping with stress (Wang & Saudino, 2011). Stress has also been shown to reduce emotion regulation ability. Stress-based HPA axis activation reduces top-down control of emotion regulation (Arnsten, 2009; Hermans et al., 2014). Acutely reducing HPA axis activation could therefore allow a person to better utilize emotion regulation strategies. Slow-paced breathing can activate the parasympathetic nervous system through increasing vagal tone (Sinha et al., 2013; You, Laborde, Zammit, Iskra, Borges, & Dosseville, 2021; You, Laborde, Zammit, Iskra, Borges, Dosseville, et al., 2021; Zaccaro et al., 2018). In this study, we examined whether a short bout of slow breathing could acutely improve emotion regulation in young adults. Participants (n=13) were asked to regulate their emotional reactions to negative images by either increasing, decreasing, or maintaining the emotional intensity. In the experimental condition, participants completed a 3 minute voluntary breath control exercise, known as box breathing, prior to the experimental trials. For negative images, there was a significant main effect of condition ($F(1,24)=11.898$, $p=0.005$) on valence, such that the slow breathing condition resulted in more positive valence ratings. In addition, there was a significant main effect of emotion regulation instructions ($F(1,24)=17.243$, $p < 0.001$) on valence such that more negative valence ratings were reported for the enhance instruction than the maintain and the suppress instructions. Similarly there were significant main effects of condition ($F(1,24)=18.86$, $p=0.001$) and emotion regulation instructions ($F(1,24)=14.09$, $p < 0.001$) on arousal with the control condition associated with increased arousal compared to the breathing condition, and arousal levels following the pattern of the instructions. There was also a significant condition by emotion regulation instruction interaction ($F(1,24)=5.259$, $p=0.013$) for self-reported success at following the regulation instruction, such that in the slow breathing condition, participants rated themselves as equally successful at all instructions (all p's >0.100), whereas in the control condition, participants were significantly worse at suppressing their emotional states to negative images compared to both enhancing ($t(12)=3.934$, $p=0.002$) and maintaining ($t(12)=2.799$, $p = 0.016$) them. Results indicate that slow breathing improved emotional valence, reduced emotional arousal, and increased success at using emotion regulation strategies. This supports that slow breathing can be an effective tool at helping to improve cognitive regulation of emotions.

Disclosures: L. Kurdziel: None. **L. McDevitt:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.04

Topic: G.04. Emotion

Support: •Program of Excellence funds from the University of Nebraska
•University of Nebraska Collaboration Initiative
•BIG Idea Pilot grant from the University of Nebraska at Omaha
•Research Development Grant through the University of Nebraska at Omaha
The National Institute of General Medical Sciences, 1U54GM115458-01

Title: Cortisol responses to social videos and depressive symptoms in caregivers to older adults with chronic conditions

Authors: *N. MILLER[1], J. BEADLE[2];
[1]Grad. Program in Neurosci. and Behavior, [2]Gerontology, Univ. of Nebraska, Omaha, Omaha, NE

Abstract: Title: Cortisol responses to social videos and depressive symptoms in caregivers to older adults with chronic conditions
Keywords: caregiving, aging, cortisol, depression**Abstract**Caregivers to older adults with chronic conditions often experience significant, chronic stress due to the challenging emotional and physical demands of their role. Furthermore, many caregivers experience depressive symptoms due to the stressful nature of their role. Yet, little is known about the degree to which caregivers differ from non-caregivers in their stress-related hormonal response to social contexts and how this relates to depressive symptoms. The pilot sample included 12 caregivers to older adults with chronic conditions (Mage=48.4) and 13 healthy non-caregiver comparison participants (Mage=52.5) who were not significantly different in age (p=.6). Participants were excluded if they had a history of psychiatric or neurological disease. The study included viewing a series of two social videos - (1) a non-emotional video, and (2) an emotional video depicting others' who are suffering. Depressive symptoms were measured by the Beck Depression Inventory. Salivary cortisol was measured prior to and after the videos. For the post-video cortisol assessments, we found a significant group x video type interaction $p<.05$. Follow-up planned comparisons showed a marginally significant effect (p=.08), such that caregivers had lower cortisol in response to the non-emotional video than healthy comparison participants. In the caregiver group, there was a negative correlation between depressive symptoms and cortisol response to the non-emotional video (r= -.66, p=.01). Specifically, caregivers with higher depressive symptoms tended to have lower cortisol levels in response to the non-emotional, social video than caregivers with lower depressive symptoms. There was no significant relationship found in the non-caregiver comparison group. These results have implications for caregiver stress and depressive symptoms in the context of social settings. Future research is needed to replicate these findings in a larger sample and to examine relationships with perceived chronic stress and caregiver burden.

Disclosures: **N. Miller:** None. **J. Beadle:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you

are a PI for a drug study, report that research relationship even if those funds come to an institution.; •Program of Excellence funds from the University of Nebraska, •University of Nebraska Collaboration Initiative Planning and Seed, •BIG Idea Pilot grant from the University of Nebraska at Omaha, •Research Development Grant through the University of Nebraska at Omaha. F. Consulting Fees (e.g., advisory boards); Section Editor for Journal of Current Behavioral Neuroscience Reports.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.05

Topic: G.04. Emotion

Support: АП 08856595, Ministry of Education and Science of Kazakhstan, "EEG/MRI study of brain development, emotional-cognitive functions, and genetic markers in different age groups", PI Kustubayeva A.M.

Title: Erp responses to emotional conflict in youth: age and gender differences

Authors: A. KUSTUBAYEVA[1], A. KAMZANOVA[2], ***M. ZHOLDASSOVA**[3], G. DATHABAYEVA[2], A. BEKTURSYNOVA[2], G. MATTHEWS[4];
[1]Dept. of Biophysics, Biomedicine, and Neurosci., [2]Depatment of Biophysics, Biomedicine, and Neurosci., [3]Ctr. for Cognitive Neurosci., al-Farabi Kazakh Natl. Univ., Almaty, Kazakhstan; [4]Depatment of Psychology, George Mason Univ., Fairfax, VA, VA

Abstract: The maturation of brain systems for affective control underpins changes in emotion-regulation during adolescence that impact social functioning and mental health. Boys and girls differ in the expression and regulation of emotion, but the role of gender in the development of affective control is not fully understood. The present research aimed to investigate effects of age and gender on brain activity during performance of an emotion conflict task in healthy adolescents, additionally its relation to emotional intelligence. The study was approved by the local Ethics Committee. Electroencephalogram (EEG) was recorded from 50 participants (mean age 15.88 SD=2.62) during task performance based on emotional pictures from Databrary database (Benda&Scherf, 2020) by using ANT Neuro with 64 electrodes. Participants had to differentiate congruency of auditory and visual stimuli with emotion expressed in four types of basic emotional faces (fear, sad, anger, happy). Conflict was manipulated by overlaying emotion names that were congruent or incongruent with the face. The EEG/ERPlab toolbox (Lopez-Calderon& Luck, 2014) was used for preprocessing and measurements of N170 amplitude for P7, P8 and average P300 amplitude for the networks (central, anterior, posterior, left and right hemisphere) in sex and age groups (12-15 and 16-20 y.o.). Behavioral data analysis revealed significant within subject main effects of condition (slower to incongruent, F=17.973, p=0.00) and emotion (slower to sad faces, F=11.066, p=0.02), an emotion*condition interaction (F=10.305, p=0.02), a and a gender effect (slower in boys, F=5.025, p=0.030). N170 amplitude

analysis for P8 electrode showed a significant emotion effect (more negative amplitude for angry face, F=2.812, p=0.042). There were also significant emotion*condition (F=2.818, =0.041) and emotion*condition*age (F=2.716, p=0.047) interactions. P300 amplitude analyses showed a significant decrease with age in central network (F=4.647, p=0.036) for incongruent face. Increased P300 amplitudes in central (F=4.144, p=0.047) and left hemisphere networks (F=3.434, p=0.007) were observed for happy face in females in comparison to males. A subsidiary aim was to test for individual differences in brain activation. We report correlations between N170 and P300 amplitude with indexes emotion regulation. In conclusion, the study shows gender and age differences in neural response to emotion conflict that varied with emotion and brain network. These findings contribute to understanding gender differences in regulating emotion as the brain matures during adolescence.

Disclosures: **A. Kustubayeva:** None. **A. Kamzanova:** None. **M. Zholdassova:** None. **G. Dathabayeva:** None. **A. Bektursynova:** None. **G. Matthews:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.06

Topic: G.04. Emotion

Title: The influence of trait depression and self-relevance on corrugator activity during the imagination and recall of personal episodic events

Authors: *L. FAUL, J. M. ROTHROCK, K. S. LABAR;
Ctr. for Cognitive Neurosci., Duke Univ., Durham, NC

Abstract: Electromyographic (EMG) activity of the corrugator supercilii increases during unpleasant emotional states and decreases during pleasant states. Corrugator response is commonly assessed during the brief presentation of visual stimuli, but comparatively less work has examined sustained activity during the mental simulation of personal events. Importantly, examining how emotional response to such events is shaped by trait depression may help elucidate biases in emotion expression and episodic memory that contribute to mood disorders. Accordingly, we analyzed EMG data from a total of 80 participants (M age 21.3 yrs; 47 F) with varying levels of self-reported trait depression. Participants first imagined hypothetical scenarios depicting emotional events while providing ratings of valence, arousal, and likelihood of the depicted event occurring, which reflects self-relevance. Trait depression attenuated the relationship between self-reported arousal and valence, such that neutral scenarios elicited comparable arousal as happy and sad scenarios. Sad scenarios were also deemed equally likely to occur as neutral and happy scenarios only among highly depressed individuals. Corrugator response was most sensitive to the valence of each scenario for individuals with low to average depression, whereby activity increased for sad scenarios and remained at baseline or decreased for happy scenarios. By contrast, highly depressed individuals exhibited a similar pattern of

activity only when scenarios were perceived as very likely to occur, but otherwise corrugator activity was unassociated with scenario valence. After the imagination task, participants were randomly assigned to repeatedly recall either happy or sad autobiographical events. Recalling happy memories significantly improved self-reported mood whereas recalling sad memories significantly deteriorated mood, and this latter effect was amplified among individuals high in trait depression. Corrugator activity increased during sad memory recall and decreased during happy memory recall, and the extent of this effect predicted overall change in mood after the task. Increased trait depression was associated with stronger corrugator activity during sad memory recall. Our findings suggest that emotional response in depression is shaped by self-relevance of episodic events, whereby less self-relevant imagined scenarios are associated with a pattern of emotional context insensitivity, while recalling sad personal memories elicits an amplified psychophysiological response.

Disclosures: **L. Faul:** None. **J.M. Rothrock:** None. **K.S. LaBar:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.07

Topic: G.04. Emotion

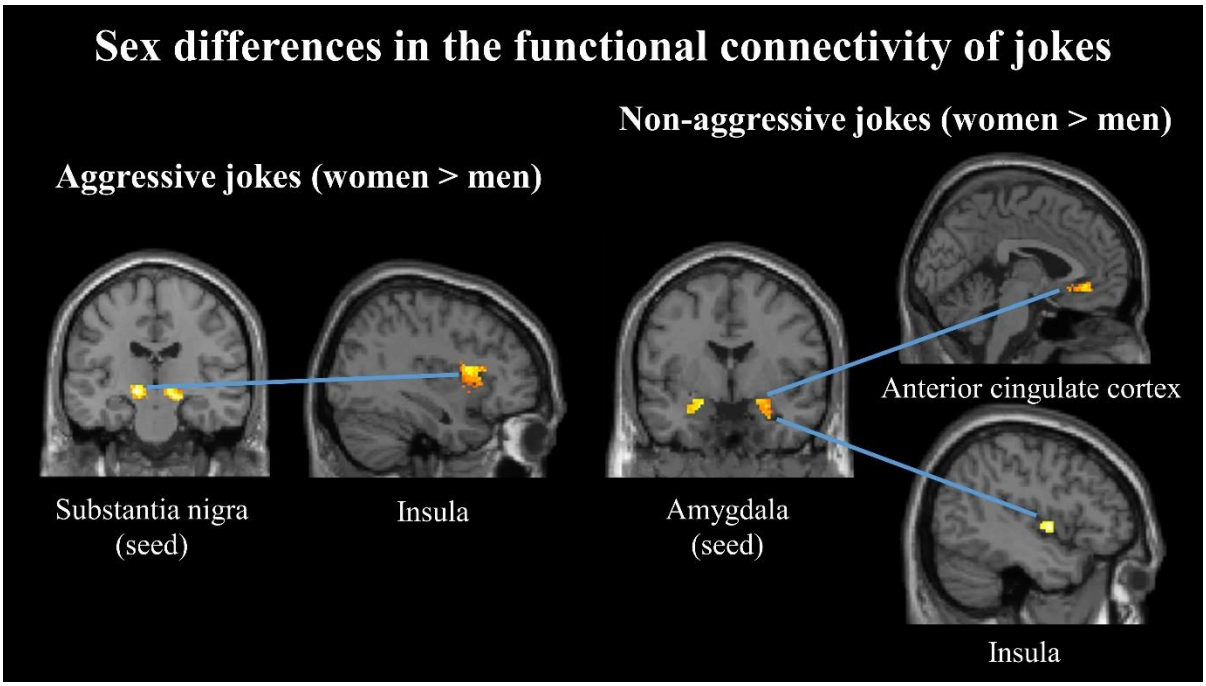
Support: MOST 109-2410-H-007-023-MY3
NTHU-YS-B-2020003

Title: Sex Differences in the Functional Connectivity of Aggressive and Non-aggressive Jokes

Authors: C.-H. YEH[1], *Y.-C. CHAN[2];
[2]Natl. Tsing Hua Univ., [1]Natl. Tsing Hua Univ., Hsinchu, Taiwan

Abstract: Sex differences in the appreciation of jokes have become increasing interest. The findings are complex based on behavioral studies, suggesting distinctions between different types involving aggression or not, and underlying neural mechanisms have yet to be fully understood. Participants included 16 men and 16 women, all of whom were right-handed with no history of neurological or psychiatric problems. The present study used a total of 64 aggressive joke and nonjoke stimulus pairs (32 aggressive stimulus pairs for each) and 64 non-aggressive joke and nonjoke stimulus pairs (32 non-aggressive stimulus pairs for each). The nonjokes were corresponding baseline stimuli which were constructed by replacing the punch lines. The present study used a 2 × 2 mixed factorial design, with group (men versus women) and type (aggressive versus non-aggressive) as factors, in an event-related fMRI paradigm. Imaging was performed using a 3 T Siemens Magnetom Prisma scanner. The functional images were pre-processed and analyzed using SPM12 implemented in Matlab. Psychophysiological interaction (PPI) analysis demonstrated that women showed functional connectivity between substantia nigra (SN) of the midbrain (seed) and insula coupling for aggressive jokes (AJ) versus non-aggressive jokes (NJ).

Also, women showed functional connectivity between SN (seed) and insula and middle temporal gyrus coupling for AJ versus NJ. Conversely, women exhibited functional connectivity between SN (seed) and ventral anterior cingulate cortex (vACC), insula, and temporoparietal junction (TPJ) for NJ versus AJ. Also, women displayed functional connectivity between amygdala (seed) and vACC and insula for NJ versus AJ. The findings may show that women displayed greater functional connectivity than men did in the mesolimbic reward system, which suggestions a more robust bottom-up emotional response during humor appreciation in both aggressive jokes and non-aggressive jokes.



Disclosures: **C. Yeh:** None. **Y. Chan:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.08

Topic: G.04. Emotion

Support: Kavli Institute for Neuroscience at Yale
Yale Center for Clinical Investigation
NeuroNext Fellowship
VA National Center for PTSD

Title: Human single neurons display mixed selectivity for emotional valence

Authors: *A. ALJISHI[1], M. LIANG[1], A. P. KAYE[2], E. C. DAMISAH[1];
[1]Neurosurg., [2]Psychiatry, Yale Univ., New Haven, CT

Abstract: With overwhelming sensory information that we constantly receive, survival require attending to motivationally salient information. One suggested mechanism that the brain utilizes to distinguish important information is by processing the valence of stimuli. Hence, studying how the brain processes emotional valence can further our understanding of how emotions modulate human attention and cognition. Most human studies have investigated the neural basis of emotional valence using fMRI or EEG. Here, we utilize microwire-recordings in multiple brain regions to identify the substrates of emotional valence in humans on a single neuron level. We recorded the activity of 99 neurons across the amygdala (AMY), hippocampus (HPC), anterior insula (aINS), and anterior cingulate (ACC) of three pharmaco-resistant epileptic patients while performing an affective visual task. Subjects were presented with pictures (International Affective Picture System) that have different emotional arousal and valence belonging to four categories (animals, humans, scenes, and objects). They were asked to rate the unpleasantness of each picture (subjective emotional valence). We analyzed the changes in neurons' firing patterns associated with the presentation of stimuli, the subjective ratings, and stimuli categories. Valence-signaling neurons were defined based on the correlation between their firing rate and the subjective rating of unpleasantness. We found that 31% of recorded neurons were responsive to the task. Category-selective neurons were ubiquitous, suggesting that category selectivity as a higher order feature may not be region-specific. However, a group of neurons in AMY, HPC, and aINS displayed mixed selectivity for category and emotional valence. For example, scene-selective neurons in aINS had firing rate changes that correlated with the positive valence of stimuli within the scene category, but not other categories. Our findings suggest the existence of category-selective neurons that signal emotional valence within AMY, HPC, and aINS. This mixed selectivity (category and valence) highlights the complex role of these neurons in valence processing. The existence of such complex selectivity could be for organizing response to emotionally salient stimuli that require category-specific behavior. In an ongoing work, we seek to analyze the relationship between this neuronal population and attention-related neurons to enhance our understanding of how emotional valence regulates attention.

Disclosures: A. Aljishi: None. **M. Liang:** None. **A.P. Kaye:** None. **E.C. Damisah:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.09

Topic: G.04. Emotion

Title: Amygdala volume and emotion regulation in aging

Authors: *M. ZHANG, D. ZEITHAMOVA;
Psychology, Univ. of Oregon, Eugene, OR

Abstract: General cognitive ability declines in older adulthood; however, older adults show higher levels of emotional well-being. Emotional regulation is a determinant factor to understanding affective change through age. Here we asked how emotional regulation changes over adulthood and how such changes may be related to amygdala volume in older adults. Using data from participants ages 18-87 from the Cambridge Centre for Ageing and Neuroscience (Cam-CAN) repository that included measures of emotional reactivity, subjective feeling of emotion regulation and emotion regulation success in response to short film clips. We then correlated emotion behaviors with amygdala volume, known to decline in aging. No statistically significant results were found for emotion regulation success. Subjective emotion regulation declined with age, but this effect was not explained by decline in amygdala volume. Emotional reactivity also declined with age. Moreover, the relation between emotional reactivity and amygdala volume differed among age groups. In young adults, emotion reactivity decreased with increased amygdala volume. In contrast, in older adults, reactivity increased with increased amygdala volume, suggesting preserved function. These findings imply that emotional control is related to brain volume change.

Disclosures: M. Zhang: None. **D. Zeithamova:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.10

Topic: G.04. Emotion

Title: Mid-latency gamma-band responses to faces in the amygdala depend on the attended to expression. Insights from human intracranial recordings.

Authors: *E. WEIDNER[1], S. MORATTI[2,3], S. SCHINDLER[4], P. GREWE[5,6], C. BIEN[5], J. KISSLER[1];
[1]Psychology, Affective Neurosci., Univ. of Bielefeld, Bielefeld, Germany; [2]Exptl. Psychology, Complutense Univ. Madrid, Madrid, Spain; [3]Lab. for Clin. Neurosci., Ctr. for Biomed. Technology, Tech. Univ. of Madrid, Madrid, Spain; [4]4 Inst. of Med. Psychology and Systems Neurosci., Univ. Münster, Münster, Germany; [5]Epileptology (Krankenhaus Mara), Med. Sch. OWL, Bielefeld, Germany; [6]Clin. Neuropsychology and Epilepsy Res., Med. Sch. OWL, Bielefeld Univ., Bielefeld, Germany

Abstract: The amygdala is assumed to contribute to a bottom-up attentional bias for emotional faces. However, its interaction with top-down attentional processes awaits further clarification. Here, we studied the context-dependence of emotion-driven responses in the human amygdala during face processing. We conducted intracranial EEG depth recordings of the amygdala in 7

patients with epilepsy (3 right, 4 left, age M = 35.29, range = 19-63) and analyzed oscillatory activity in the gamma band (30-120 Hz) using non-parametric cluster-based permutation tests. Three randomized blocks consisting of angry, neutral, and happy facial expressions were presented, and one expression was denoted as the target category in each block. The target had to be detected via button-press. Angry and happy faces were detected faster than neutral faces. An early (about 200 ms) gamma-power increase during the neutral target condition was found for both angry and happy distractors compared to neutral targets. This might indicate an automatic attentional shift towards emotional distractors, even when they are not task-relevant. A mid-latency onset (about 300 ms) increase in amygdala gamma-activity was selective to attended angry faces. Attention to happy faces did not result in an electrophysiological differentiation of facial expressions in the gamma frequency range. Still, overall, gamma-band activity was most pronounced in the attend to happy run, however, without difference between targets and non-targets. The present data shed light on the attentional modulation of amygdala threat-sensitivity. Incremental target effects seem to be selective to angry faces as they were not present for neutral and happy faces. The enhanced processing of emotional distractors vs. neutral targets also corroborates assumptions about an automatic emotion-driven attentional capture when emotion is not in the focus of attention. These results uncover possible mechanisms of how different task-induced attentional biases can modulate oscillatory neural responses to emotional stimuli. Assuming that individuals differ in implicit attentional biases that the present explicit instructions might mimic, these data might help explaining individual differences in threat processing.

Disclosures: E. Weidner: None. **S. Moratti:** None. **S. Schindler:** None. **P. Grewe:** None. **C. Bien:** None. **J. Kissler:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.11

Topic: G.04. Emotion

Support: NIH Grant GM137863

Title: Negativity bias in empathy deficient individuals: an ERP study

Authors: *R. HURTADO, A. BARSEGYAN, J. SCHINDLER, A. LLANES-GAXIOLA, S.-M. KANG;
Psychology, California State Univ. Northridge, Northridge, CA

Abstract: The present study aimed to understand how individuals with low empathic (LE) traits would process emotional information differently than those with high empathic (HE) traits by using electroencephalography (EEG). Previous research has shown that negatively valenced emotional information attracts more attention than positively or neutrally valenced information.

This sensitivity to negative emotional information is referred to as the negativity bias (Ito, Larsen, Smith, & Cacioppo, 1998). By measuring event-related brain potentials, previous work demonstrated that the negativity bias emerged during the early information processing (Hajcak & Olvet, 2008). We hypothesized that people with a difficulty in empathizing with others would be less likely to exhibit the negativity bias compared to those normally empathic. To test this hypothesis, 45 college students were asked to passively view a total of 135 negative, positive, and neural stimuli taken from the International Affective Picture System in a random order, while their EEG was recorded. The empathic traits of the participants were assessed using self-reported measures including the Empathy Quotient (Baron-Cohen & Wheelwright, 2004) and the Interpersonal Reactivity Index (Davis, 1983). The participants were divided into two groups, High Empathic (HE) and Low Empathic (LE) groups, based on their affective empathy scores. The results of the current study revealed that there was a marginally significant interaction effect between valence conditions and affective empathy groups on the average Late Positive Potential amplitudes of the medial lateral region of the cortex, $F(2,86) = 2.979$, $p = .056$, $eta^2 = .065$. Against our hypothesis, this effect suggests that the LE individuals actively recruited more neural resources to process negative emotional information than the HE participants did. Implications of the current findings were discussed in terms of neural compensatory efforts to make up the empathic deficits among the people with low emotional empathy.

Disclosures: R. Hurtado: None. **A. Barsegyan:** None. **J. Schindler:** None. **A. Llanes-Gaxiola:** None. **S. Kang:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.12

Topic: G.04. Emotion

Title: Fake laughter increases beneficial brain EEG gamma frequency (25-40 Hz)

Authors: *L. S. BERK, S. SRIDHAR, K. PATEL, T. LOHMAN, G. BAINS, N. DAHER, E. LOHMAN, G. OLUSETIRE, M. CHOUHAN;
Loma Linda Univ., Loma Linda, CA

Abstract: *Problem Statement:* In recent years, there is interest in investigating brain health benefits of positive human emotions, specifically the psycho-physiological mechanisms associated with humor associated mirthful laughter (HAML) and its possible therapeutic efficacy. However, there are claims that Fake Laughter (FL) could be similar in response specifically with brain frequency modulation & benefits.Our previous research has shown HAML can provide both physical and brain health benefits (increase of gamma frequency). However, there is no known study that has explored brain electroencephalography (EEG) oscillations from FL, specifically beneficial gamma frequency. *Objective:* Our research aimed to determine if brain frequency modulation induced from FL increases Power Spectral Density

(μV^2) of beneficial gamma frequency (25-40 Hz). *Methods:* In a quasi-experimental design, 15 university graduate students, ages 22-35, were recruited. Subjects were: 1) first asked to produce FL for 5 minutes (no cognitive or emotional context provided); after a 5-minute washout period they 2) watched an 8-minute humor video (HV). EEG wave band activity (0.5-49 Hz) was acquired from 24 cerebral cortical scalp locations using the B-Alert 24X wireless telemetric system. *Results*: There were two outstanding oscillation outcomes: 1) Topographical maps of absolute Alpha Power (8-13 Hz) was averaged across all participants for each stage of the experimental protocol. All data were normalized for effect size (Hedges-G). Statistically significant differences in Alpha Power was observed between the two stages ($p<0.05$, paired t-test, not corrected for multiple comparison). Alpha Power decreased significantly Post FL to HV ($p=0.004$, $G= 0.87$) and increased back to Post HV ($p=0.003$, $G= 0.86$); 2) Topographical maps of Gamma Power (25-40 Hz) were averaged across the participants for each stage of the protocol. Statistically significant differences in Gamma Power were observed between stages of the protocol ($p<0.05$, paired t-test, not corrected for multiple comparison). Gamma Power increased significantly from baseline for the FL task ($p=0.001$, $G= 0.987$) and decreased back in the Post FL period ($p=0.004$, $G= 1.01$). *Conclusion:* A significant decrease in Alpha Power was observed during the Humor Video indicative of arousal to the external world. More profoundly was significant increase in Gamma Power μV^2 (25-40 Hz) during Fake Laughter. Thus, we postulate there may be similar potential benefits from Fake Laughter to psycho-physiological mechanisms as seen in HAML (enhanced cognitive processing, recall, memory & synaptic plasticity) as well as the enhancement of positive emotions.

Disclosures: L.S. Berk: None. **S. Sridhar:** None. **K. Patel:** None. **T. Lohman:** None. **G. Bains:** None. **N. Daher:** None. **E. Lohman:** None. **G. Olusetire:** None. **M. Chouhan:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.13

Topic: G.04. Emotion

Support: National Geographic

Title: Neuroforecasting nature imagery impact on internet donations

Authors: *T. SRIRANGARAJAN[1], N. SAWE[1], T. THYS[2], B. D. KNUTSON[3];
[1]Stanford Univ., Stanford, CA; [2]Sea Studios Foundation, Natl. Geographic, Monterey, CA; [3]Dept Psychology, Stanford Univ. Dept. of Psychology, Stanford, CA

Abstract: Donations to endangered species can help offset threats and internet campaigns facilitate these donations, but it is not always clear whether individuals efficiently donate to protect the most endangered species or use some other psychological proxy (e.g., salience or cuteness in the case of charismatic megafauna). Therefore, to better understand and forecast why

people donate to protect endangered species, we combined behavioral, neuroimaging, and representative survey experiments. We found that brain activity in brain regions associated with anticipatory affect predicted individual donations and also forecast donations on social media. Physical attributes that elicited excitement (e.g., cuteness) and anxiety (e.g., threat) tended to increase donations. These findings suggest that the efficiency of donations to offset extinction might be enhanced by boosting affective elements of the presentation of endangered species.

Disclosures: **T. Srirangarajan:** None. **N. Sawe:** None. **T. Thys:** None. **B.D. Knutson:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.14

Topic: G.04. Emotion

Support: NRF-2019M3C7A1032262
NRF-2021M3E5D9025026
S0254-22-1002

Title: Neural correlates of subjective well-being in healthy individuals: A multimodal neuroimaging study

Authors: *H.-Y. JUNG[1], C. PAE[1], Y.-G. HWANG[1], S. LEE[1], N. KANG[1], H.-J. KIM[1], I. AN[1], M.-K. KIM[2], M. BANG[1], S. CHO[3], S.-H. LEE[1];
[1]Dept. of Psychiatry, CHA Bundang medical center, Seongnam, Korea, Republic of; [2]Dept. of Psychiatry, CHA Ilsan medical center, Goyang, Korea, Republic of; [3]Dept. of Psychiatry, Kangbuk Samsung Hosp., Seoul, Korea, Republic of

Abstract: Background: Subjective well-being (SWB) or happiness reflects an individual's cognitive and affective assessment of life. Although relationship between psychological factors and SWB has drawn much attention from researchers, the precise neural structural correlates of SWB are broadly unknown. In the present study, we aimed to investigate the associations between gray matter (GM) volumes, cortical characteristic's changes such as gyrification, sulcal depth, white matter (WM) microstructures, and SWB in healthy individuals using multimodal T1-weighted images and diffusion tensor imaging. **Methods:** We enrolled 70 healthy individuals using magnetic resonance imaging. We measured their SWB using the Concise Measure of Subjective Well-Being. Voxel-wise statistical analysis of GM volumes was performed using voxel-based morphometry, while gyrification and sulcal depth were performed with surface-based analysis using Statistical Parametric Mapping 12 and the Computational Anatomy Toolbox 12. In addition, WM fractional anisotropy (FA) values were analyzed using tract-based spatial statistics. **Results:** In healthy individuals, higher levels of SWB were significantly correlated with increased GM volumes of the insula, and gyrification of the insula and the posterior cingulate cortex. Moreover, SWB were significantly and negatively correlated with the

sulcal depth of the medial frontal gyrus and FA values in clusters of the body of the corpus callosum, precuneus WM, and fornix cres/stria terminalis. Additionally, a correlational analysis revealed that GM volumes and FA values in these significant regions were significantly correlated with severity of psychological symptoms such as depression, anxiety, and quality of life. **Discussion:** The findings of the present study indicate that GM volumes, cortical characteristic's changes and WM microstructures in the regions related to default mode network, or adaptive integration of affective processing and emotional regulation may contribute to higher SWB and psychological symptom severity as well as quality of life in healthy individuals.
Keywords: Subjective well-being, Insula, Default mode network, Voxel-based morphometry

Disclosures: H. Jung: None. **C. Pae:** None. **Y. Hwang:** None. **S. Lee:** None. **N. Kang:** None. **H. Kim:** None. **I. An:** None. **M. Kim:** None. **M. Bang:** None. **S. Cho:** None. **S. Lee:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.15

Topic: G.04. Emotion

Title: Quantitative evaluation of food preference using physiological and psychophysical measurements

Authors: H. LI[1], S. LI[1], K. MATSUO[2], ***T. OKAMOTO**[1];
[1]Kyushu Univ., Fukuoka, Japan; [2]Nichirei Foods Inc., Tokyo, Japan

Abstract: Various indices have been developed to evaluate food preference. One of the promising candidates for its objective evaluation is brain activity. Food preference and brain activity are closely related, but it is difficult to record brain activity during eating. Brain activity during eating continuously elicits myoelectric activity and body movements. Another candidate is behavioral evaluation using cognitive tasks. Although objective evaluation was possible by devising cognitive tasks, the measurement of indirect responses poses unique challenges. In this study, we aimed to physiologically and psychophysically quantify food preference before and after eating by introducing Stroop cognitive tasks and recording electroencephalographs (EEGs) using mobile dry-EEG headset. To evaluate food preference, two types of fried rice were prepared in this study. One was considered tasty (Sample 1; Nichirei Foods Inc., Tokyo, Japan), and the other was considered comparatively tasteless (Sample 2; other food company). Twenty healthy students (19-26 years old) participated in this experiment. They were divided into two Groups: 10 students in Group 1 ate Sample 1 and 10 students in Group 2 ate Sample 2. After providing written informed consent, the participants were engaged in a baseline block of experiment including undergoing an EEG recording, performing a Stroop task, eating a given sample, undergoing an EEG recording, and undergoing subjective evaluations. Subsequently, they were engaged in three test blocks in an order similar to that of the baseline block. In the

Stroop task, the stimuli were presented randomly in four colors: "red," "blue," "yellow," and "green." They were instructed to indicate the meaning or the color of the word based on the cue by pressing 1, 2, 3, or 4 on the keyboard while looking at the monitor. After recording, we performed frequency analysis on EEGs and obtained amplitudes in delta, theta, alpha, beta, and gamma bands. The results showed that Group 1 showed higher theta amplitudes in the frontal midline region, and Group 2 showed higher alpha amplitudes in the occipital region. This indicates that high preference food would make participants pay more attention on the task and let participants be more aroused during the cognitive task. We can conclude that our methodology is useful for the physiological and psychophysical quantification of food preference.

Disclosures: H. Li: None. **S. Li:** None. **K. Matsuo:** None. **T. Okamoto:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.16

Topic: G.04. Emotion

Title: A quantitative approach to consumers' behavior accelerated by emotions

Authors: *Y. EDAGAWA[1], S. AOKI[2], J. YAMAKAGE[1], Y. HIRABAYASHI[2], Y. KOHATA[3], T. SHIMURA[2], T. OHNO[1];
[1]Waseda Univ., Tokyo, Japan; [2]Dentsu ScienceJam Inc., Tokyo, Japan; [3]Dentsu Inc., Tokyo, Japan

Abstract: Emotions play an important role in human behavior. And there should be a mechanism by which consumer purchasing behavior is driven by changes in consumer insight between the recognition of the desired product and the actual purchase. We are currently familiar with various consumer behavior models. However, none of these models so far have focused on changes in emotions and have shown changes at the purchasing site. Therefore, we attempted to model consumers' behavior driven by emotions. First, we extracted factors related to emotion through a questionnaire survey of 3,000 respondents regarding their purchasing experiences that resulted in a good mood. Comparing emotional and non-emotional factors, we found that the involvement of emotional factors was about twice as high in the good mood purchasing experience. Next, to measure emotional changes in the actual in-store purchasing experience, we performed EEG measurements and the subjects walked around among the in-store displayed products. We then successfully measured time-course changes in emotion by using a system that extracts emotion-related characteristic brain waves from the measured EEG. After measuring the EEG in the store, the subjects were given a questionnaire that included recall of the products they remembered, and the products in the subjects' first evoked set were detected. Then, by combining the results with those of the EEG measurements, we obtained the following results. When subjects were in front of their own memorable products, they showed increases in expectancy,

liking, interest, satisfaction, and positive affect values, and conversely, decreases in stress and sedation values. In addition, for products in the first evoked set, the subjects spent more time browsing for those products, suggesting that the increase in these emotional values may have contributed to an increase in the product browsing time. This indicates the importance of emotional factors in purchasing behavior. And it suggests that changes in brain activity involving multiple emotions actually occur when experiencing a product of interest and increasing purchase intention.

Disclosures: **Y. Edagawa:** None. **S. Aoki:** None. **J. Yamakage:** None. **Y. Hirabayashi:** None. **Y. Kohata:** None. **T. Shimura:** None. **T. Ohno:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.17

Topic: G.04. Emotion

Support: the Chinese Scholarship Council (201806040186)

Title: Emotional information facilitates and interferes with memory integration through distinct hippocampal processes

Authors: *Y. ZHU, W. LIU, N. KOHN, G. FERNÁNDEZ;
Donders Inst. for Brain, Cognition and Behaviour, Nijmegen, Netherlands

Abstract: Emotional information affects related experiences through integrated memories. However, the neurobiological mechanisms of how emotion modulates memory integration for related information with different valences (i.e., neutral or negative) remain unclear. In a between-subject functional magnetic resonance imaging (fMRI) study, we manipulated the valence of stimuli used in an associative memory paradigm to examine emotion-modulated memory integration. Specifically, participants were divided into three groups: integrating emotional events with related neutral events, integrating both related emotional events and integrating both related neutral events as control. Behaviorally, emotional information facilitated its integration with neutral events, but interfered with its integration with emotional events. Neurally, our trial-by-trial pattern similarity analysis revealed that the emotion-induced facilitation effect, occurring on memory integration of emotional and neutral events, was associated with increased hippocampal reactivation (i.e., higher pattern similarity) during both encoding and retrieval. Using the hippocampal region as a seed, our psycho-physiological interaction (PPI) analysis revealed that the facilitation effect was also supported by strengthened hippocampal connectivity with the amygdala and a set of neocortical areas, including the temporoparietal junction (TPJ), supplementary motor area (SMA), middle cingulate gyrus (MCC) and precuneus. In contrast, the emotion-induced interference effect, occurring on memory integration of both emotional events, was associated with impaired hippocampal

reactivation during retrieval, which appeared to offset increased reactivation during encoding. Besides, similar but relatively weak hippocampal connectivity was found underlying this interference integration. Taken together, emotional information facilitates memory integration of emotional and neutral events while it interferes with the integration of two emotional events, through distinct hippocampal reactivation and connectivity patterns. Our findings advance the understanding of neurobiological mechanisms by which emotion adaptively modulates memory integration, and provide novel insights into the maladaptive emotion modulation on integration.

Disclosures: **Y. Zhu:** None. **W. Liu:** None. **N. Kohn:** None. **G. Fernández:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.18

Topic: G.04. Emotion

Support: Japan Society for the Promotion of Science [KAKENHI Grant Number JP15K19828 (H.O.) and JP15K09365 (T.T)

Title: Heterogenous functional connectivity in normal volunteer during mild dyspnea

Authors: A. YORITA, T. KAWAYAMA, T. KINOSHITA, H. ODA, Y. TOKUNAGA, T. TATEISHI, T. HOSHINO, ***T. TANIWAKI**;
Dept Med, Kurume Univ., Kurume Univ., Fukuoka 830-0011, Japan

Abstract: Several studies have mapped brain regions associated with respiratory control and respiratory perception. However, its effect to resting state networks is unknown. Our objective was to determine the resting state networks during mild dyspnea. Resting-state functional magnetic resonance imaging data was collected for 35 healthy volunteer with or without mild dyspnea induced by resistive load. Functional connectivity (FC) was analyzed using Statistical Parametric Mapping 12, the CONN toolbox and whole brain ROI-to-ROI analysis with cluster level-comparison. Respiratory score showed that some participants felt dyspnea only with resistive load and others complained dyspnea at both conditions. The former represented increase of FCs between the motor cortex/salience network and visual cortex, while the latter indicated enhanced FCs between the orbito-frontal cortex and posterior temporal gyrus. These results suggest the heterogeneity of healthy volunteer against mild dyspnea.

Disclosures: **A. Yorita:** None. **T. Kawayama:** None. **T. Kinoshita:** None. **H. Oda:** None. **Y. Tokunaga:** None. **T. Tateishi:** None. **T. Hoshino:** None. **T. Taniwaki:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.19

Topic: G.04. Emotion

Title: Context-sensitive interaction dominant brain dynamics during emotional experiences in upper beta band

Authors: *S. MISHRA[1], N. SRINIVASAN[2], U. S. TIWARY[1];
[1]Indian Inst. of Information Technol. Allahabad, Prayagraj, India; [2]Dept. of Cognitive Sceicne, Indian Inst. of Technol. Kanpur, Kanpur, India

Abstract: The self-organized activity of the brain involves coordination among multiple interdependent components that interact across scales based on context, task and conceptual knowledge. The brain's activity once attributed to criticality is now shown to reflect multiplicative interaction across temporal scales. In our earlier work, we found a dynamic range and temporal variability in functional connectivity patterns for naturalistic emotional events in the upper beta band. Hence, in the presented work, we hypothesize that the temporal variability in functional connectivity could be related to the coordination of different mechanisms in various time scales. We performed a leader wavelet-based multifractal study to assess the multiplicative interaction between different time scales. Emotional videos were used to elicit emotional events, and the brain activity of 40 subjects was recorded using EEG. Subjects marked their emotional events using a mouse click while watching the stimulus and, after watching the stimulus, labeled these emotional events. Movie frames around the click duration were presented to help subjects recall the emotional events and label them. With this simple strategy, we were able to record emotional brain activity with reduced impact of the mind-wandering activity. Prototypical microstates and microstate sequences were calculated by segmenting and clustering the global field potential in the EEG signal. Then embedding sequences using cumulative random walk were generated. The embedding sequences were used to calculate the leader wavelet coefficients at multiple scales. Then, calculating heterogeneous variations across scales using a range of statistical moments gave a multifractal spectrum following the surrogate and statistical analysis of different spectrum parameters. The intermittent variability generated by the multiplicative interaction across temporal scales was observed in the upper beta band during emotional events. In addition, intermittent variability was also observed in the gamma band for emotional stimuli rated as less context familiar. We interpret that the multiplicative interaction between scale-dependent mechanisms might be facilitating context-sensitive emotional experiences in the upper beta band. In comparison, interaction activity in the gamma band for less context-familiar stimuli may facilitate neural adaptation to the context based on hierarchical bottom-up error feedback. Clinically, the lack of multiplicative interaction in the beta band was reported to be related to the reduction in flexible brain dynamics that could play a role in motor and cognitive disorders.

Disclosures: S. Mishra: None. **N. Srinivasan:** None. **U.S. Tiwary:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.20

Topic: G.04. Emotion

Support: NRF of Korea, Basic Science Research Program (2020R1A2C2007770)
NRF of Korea, Basic Science Research Program (2020R1A4A1018714)
NRF of Korea, Neurological Disorder Research Program (2020M3E5D9079913)

Title: Interpersonal Information Processing while Observing Others' Behavior in the Human Brain

Authors: *W. KANG[1], M. KWON[1], S.-H. LEE[1,2];
[1]Dept. of BIo and Brain Engineering, Col. of Engin., [2]Program of Brain and Cognitive Engineering, Col. of Engin., Korea Advanced Inst. in Sci. and Technol. (KAIST), Daejeon, Korea, Republic of

Abstract: Recognizing and understanding the social relationships of others is essential to our social interactions. Prior social neuroscience studies have reported the involvement of the parietal regions, including the superior temporal sulcus (STS) and inferior parietal lobule (IPL), and prefrontal cortex, while observing social interactions. However, how interpersonal information is processed in the brain during subjective recognition of social interactions remains still elusive. Here we performed an event-related functional magnetic resonance imaging (fMRI) experiment to investigate neural processing of interpersonal information. In the experiment, participants were asked to watch movie clips and rate social relationships based on the behavior of a subject and a target given in each movie clip. To reveal the dimensions of social relationship, we used principal component analysis (PCA) and found two components that can explain about 85% of the subjective rating of social relationship. To examine the neural response patterns associated with subjective ratings of interpersonal information, we used representational analysis (RSA) with Euclidean distances in regard to two principal components for each clip and found significant response patterns associated with dimensions of social relationships in the cortical regions, including the STS, IPL and dorsolateral prefrontal cortex (dlPFC). Furthermore, the identity information of subjects and targets in the movie clips could be decoded from the neural responses in the IPL and STS. These results suggest shared neural processing of interpersonal information across different individuals in the cortical network including the STS and IPL.

Disclosures: **W. Kang:** None. **M. Kwon:** None. **S. Lee:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.21

Topic: G.04. Emotion

Support: IBS-R015-D1
2019R1C1C1004512
2021M3E5D2A01022515
2021M3A9E4080780
2E30410-20-085

Title: Bodily maps of spontaneous thought

Authors: *H. SHIN[1,3], B. KIM[4], H. KIM[1,3], C.-W. WOO[1,2,3];
[1]Biomed. Engin., [2]Intelligent Precision Healthcare Convergence, Sungkyunkwan Univ., Suwon, Korea, Republic of; [3]Ctr. for Neurosci. Imaging Research, Inst. for Basic Sci. (IBS), Suwon, Korea, Republic of; [4]Psychological and Brain Sci., Dartmouth Col., Hanover, NH

Abstract: The body and the mind are intertwined. Researchers have recently begun to investigate how bodily variables, such as metabolic states and interoceptive signals, shape the spontaneous flow of thought, but the relationship between people's conception of bodily sensations and spontaneous thought is unknown. Here we aimed to identify the topography of bodily sensations that underlies content dimensions of spontaneous thought, such as valence and self-relevance, using the emBODY tool. A total of 62 participants underwent an fMRI experiment with a recently developed thought sampling task, Free Association Semantic Task (FAST). During the FAST, participants were asked to generate a chain of concepts that spontaneously came to their mind every 2.5 seconds, starting from a given seed word. Each participant generated 160 concepts across four runs. After the fMRI scan, the participants rated each concept on the content dimension scales of valence, self-relevance, time, vividness, and safety-threat. Lastly, participants colored the body areas felt activated with red and felt deactivated with blue for each self-generated concept. We developed body map-based predictive models of content dimensions with machine learning techniques using 29 participants' data. We then tested the models on one hold-out training dataset and two independent test datasets (total n = 78). The body map-based predictive models showed good performance in the training data (rs = .392 to .643 with leave-one-participant-out cross-validation) except for the time dimension. The well-performing models also showed good prediction performance across the test datasets (for valence, rs = .308 to .672; for self-relevance, rs = .355 to .679; for vividness, rs = .327 to .633; for safety-threat, rs = .202 to .578). In addition, when we tested our valence model on the body maps from Nummenmaa et al., 2014 and 2018, the model was able to predict the valence rating scores of multiple types of emotion and feeling words (rs = .624 and .402, respectively). Lastly, we correlated the trial-by-trial body maps with the fMRI data from the concept reflection period. The results showed the face vs. feet/hands somatotopy in the primary somatosensory cortex and the significant correlations between the heart area and multiple brain regions within the default mode network. These results suggest that the conceptual body map patterns were reflected in the brain activity in a neurobiologically plausible way. Overall, this study identified the topography of perceived bodily sensations that could explain the content dimensions of spontaneous thought, supporting that the body plays an important role in the spontaneous flow of our mind.

Disclosures: **H. Shin:** None. **B. Kim:** None. **H. Kim:** None. **C. Woo:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.22

Topic: G.04. Emotion

Support: NBRC Core Fund
Flagship Grant

Title: 1/f component of eeg signals as predictive marker of emotional arousal states

Authors: *R. M. BORAH, A. BANERJEE;
Computat., Natl. Brain Res. Ctr., Gurgaon, India

Abstract: Emotional arousal refers to a state of heightened physiological activity in response to evocative stimulus or situations. Previous evidence have primarily focused on the changes in canonical frequency bands (periodic activity) to explain the electrophysiological basis of different arousal states (e.g. calm and excitement). Recent developments in electrophysiological time series analysis postulates that the aperiodic background often ignored in the past as electrophysiological noise may be a dominant player in guiding functional brain states. Consequently, a pertinent question arise to what extent do the aperiodic activity, influence the emotional arousal states. Here, we examined whether the slope of the aperiodic component (1/f) of the power spectra, changes differentially along the arousal and valence axis, using the Electroencephalography (EEG) (N=29) data obtained from "Database for Emotion Analysis using Physiological Signals (DEAP)". EEG was recorded as participants watched 60s long 40 different music videos. After each video, participants rated their valence (unpleasant to pleasant), arousal (calm/bored to stressed/excited), dominance (submission to feeling in control), liking on 1-9 Likert scale and familiarity with the video on 1-4 Likert scale. We categorized the data based on the corresponding median split of the arousal and valence ratings of participants into high and low arousal trials, high and low valence trials, respectively. Subsequently, we estimated power spectrum in two separate time windows (0-30s and 30-60s) and decomposed the spectra into periodic and aperiodic components using fitting oscillations and one over f (FOOOF) algorithm over 4-40Hz range. Our results show that the 1/f slope for high arousal were steeper in than low arousal states (p &It 0.001, paired permutation test), whereas no significant difference was observed between high and low valence states. Furthermore, two-way ANOVA with factors of arousal states (2 levels: high and low) and time windows (2 levels: 0-30s, 30-60s) revealed significant effects of only arousal states [F=8.24, p=0.0041]. There was neither effect of time windows [F=0.35, p=0.552] neither any significant interaction between the two factors [F=0.05, p=0.8148].Overall our results demonstrate the significance of aperiodic activity in determining emotional arousal states. Moreover, these results also contribute to the growing body of research endorsing the physiological role of aperiodic activity.

Disclosures: R.M. Borah: None. **A. Banerjee:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.23

Topic: G.04. Emotion

Support: Nanyang Assistant Professorship (Award no. 021080-00001)

Title: Neural correlates of personality traits during emotion processing: A task-based fMRI study

Authors: *S. SIEW, J. YU;
Psychology, Nanyang Technological Univ., Singapore, Singapore

Abstract: Introduction According to the trait-congruency hypothesis, individual differences in stable personality traits influence emotion processing. All the big five personality traits have been linked to emotion recognition. However, recent findings are inconsistent. To establish more clarity, we sought to investigate the neural correlates of the different personality traits during emotion processing. **Methodology** The PIOP1 dataset from the Amsterdam Open MRI collection database was used. 187 participants completed the NEO-FFI personality questionnaire and had valid fMRI data. Subjects were aged 18 to 26 years old (M_{age} = 22.2 years) and 44% were females. Task-based fMRI data collected during an emotion-matching task was studied in relation to personality traits. In the emotion trials, subjects were presented with an emotional target face that was either stereotypical anger or fear and two emotional probes. They had to match the probes to the target. In the control trial, subjects matched the orientation of an oval. Both male and female, and White, Black, and Asian faces were used. The first-level analysis was modelled using an event-related design with motion parameters included as nuisance covariates. In the second-level analysis, a one-sample t-test was conducted to identify areas significantly activated in the emotion > control trials. Then, a multiple regression was conducted to look at the association between emotion > control contrasts and personality traits. Age and gender were included as control variables and all analyses were corrected using the threshold-free cluster enhancement. **Results** Significant clusters in the emotion > control trials were highly symmetrical and located mostly in the amygdala, visual and motor cortex regions. Activation of the left inferior and middle frontal gyrus was associated with agreeableness. Less openness was associated with stronger activation of clusters in the left middle and right superior frontal gyrus and primary motor cortex. No significant clusters of activation were associated with the other personality traits. **Discussion** During the processing of negative emotions in the task, people who were more agreeable had more activation in the left inferior frontal gyrus, which was commonly implicated in response inhibition tasks. This could point to a top-down emotion regulation approach. Those who were less open had greater activation in the middle and superior frontal gyrus and this could also reflect a top-down emotion regulation process via attention inhibition. In conclusion, the traits of agreeableness and openness do seem to influence emotion processing as shown by their different activation patterns.

Disclosures: **S. Siew:** None. **J. Yu:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.24

Topic: G.04. Emotion

Title: Emotional Response to Recycling Behaviours and its Neural Correlates

Authors: *Y. WANG, K. ITO, X. HONG;
Nanyang Technological Univ., Singapore, Singapore

Abstract: Abstract It is shown that people's emotional reaction towards environmental destruction shared the same underlying neural circuit of empathy to human or animal torturing (Geiger et al., 2017). However, most of the current studies on emotional reaction toward environment destruction relied on self-report measurements. Hence, in this study, we aim to explore the neural correlates of pro-environmental behavior with their domain-general cognitive system, namely, "analytic and holistic thinking" by functional near infrared spectroscopy (fNIRs). We tested 44 participants (21 males and 23 women) in this study to detect their hemodynamic changes on pre-frontal cortex (PFC) corresponding to recycling behaviour. All participants answered the questionnaire "analytic-holistic scale" (AHS), consisting of contradiction, perception, attention and causality that define their analytic-holistic thinking patterns before the experiment. During the experiment, all individuals were presented with two videos (the correct recycling behaviour, or the wrong recycling behaviour) in random order. We conducted a generalised linear mixed model (GLMM) to investigate the role of analytic and holistic thinking in determining hemodynamic changes. We find a significant main effect of AHS subscale "change" score, shown in the left Inferior frontal gyrus (IFG), $F(1,42) = 4.69$, $p < .05$,$\eta^2 = .10$. Furthermore, we find a significant interaction effect between recycling condition and AHS sub scale "contradiction". The contradictions score was significantly interacting with recycling condition, which was found in the anterior PFC [$F(1,42) = 3.99$, $p < .05$,$\eta^2 = .05$], right medial frontal gyrus (MFG) [$F(1,42) = 4.19$, $p < .05$,$\eta^2 = .09$], and right inferior frontal gyrus (IFG) [$F(1,42) = 4.90$, $p < .05$,$\eta^2 = .06$]. This indicated that under right recycling behaviour condition, as contradiction score increased, the haemoglobin manifested a consistent upward trend across all three channels, whereas the haemoglobin accordingly declined corresponding to the growth of contradiction score when wrong recycling behaviour was viewed. These findings suggested that people with holistic thinking might be tolerable to behaviours which are destructive the environment in neural responses.

Disclosures: **Y. Wang:** None. **K. Ito:** None. **X. Hong:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.25

Topic: G.04. Emotion

Support: UCSD Office of Research Affairs Center Launch Program

Title: Affect modeling of stereoencephalographic signals during naturalistic acoustic stimuli

Authors: *A. PATEL[1,2], J. HUANG[1], G. CHAU[1], S. BEN-HAIM[3], J. SHIH[4], T.-P. JUNG[2], V. GILJA[1];
[1]Electrical and Computer Engin., [2]Inst. for Neural Computation, [3]Neurolog. Surgery, [4]Neurosciences, UCSD, La Jolla, CA

Abstract: Affective response is a complex signal composed of changes in behavioral response, physiological response, and perceived experience. The mapping of these behavioral and physiological responses to reported experiences can differ greatly across individuals and be affected by many factors at both short and long timescales. Existing decoders using non-invasive electroencephalography (EEG) and heart-rate variability have demonstrated their ability to classify affect across a range of auditory and visual stimuli. These recordings capture heavily processed features and provide a glimpse into the underlying subcortical networks. Recent invasive stereoEEG studies have demonstrated discriminability among mood measures across long, unstructured recordings (Kirkby et. al., 2018; Sani et. al. 2018). Mood and affect are intertwined cognitive processes where mood is considered to be a state variation spanning long timescales, and affect spanning finer timescales. To better understand the underlying mechanisms driving affect, we explore the temporal sensitivity of affect modeling techniques in relevant subcortical structures in two primary axes of affect - valence and arousal - from the International Affective Digitized Sound database (Bradley & Lang, 2007). Four male subjects with intractable epilepsy were implanted with stereoEEG probes covering limbic structures. The task involved presenting a series of 6 second auditory stimuli and subsequent surveys. As an initial strategy, physiological power bands of the neural data including alpha and beta corresponding to active stimulus presentation periods are decomposed into independent components. These features are used to construct statistical models, which are evaluated against results from existing IADS-EEG studies (Bos, 2006) with comparable decoder performance (> 60%) in a subset of subjects. Incorporating dynamic models that continuously track neural states, Sani et al. demonstrated improvement in decoding mood states. To model finer timescale affect states, we extract latent temporal features using Gaussian-Process Factor Analysis. A set of spectro-spatial neural features demonstrated improved decoder performance (> 70%) in a set of subjects. To further understand the mapping between neural activity and perceived affect, the long-term variation must be separated from rapid variations related to stimulus triggered events; improved dynamic models are then needed to model the short-term variation. Such a direct measurement and understanding of the neural circuits that give rise to affect could enable objective evaluation of affect through machine learning based inference.

Disclosures: **A. Patel:** None. **J. Huang:** None. **G. Chau:** None. **S. Ben-Haim:** None. **J. Shih:** None. **T. Jung:** None. **V. Gilja:** E. Ownership Interest (stock, stock options, royalty, receipt of

intellectual property rights/patent holder, excluding diversified mutual funds); Neuralink, Corp., Paradromics, Inc.. F. Consulting Fees (e.g., advisory boards); Paradromics, Inc..

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.26

Topic: G.04. Emotion

Support: NRF-2021R1I1A1A01048880
NRF-2019M3C7A1032262
NRF-2021M3E5D9025026

Title: Superior longitudinal fasciculus in the mirroring network can be associated with self-compassion in healthy individuals

Authors: *Y.-G. HWANG[1,2], C. PAE[1], H.-Y. JUNG[1,2], N. KANG[1], C. PARK[1], M. BANG[1], M.-K. KIM[3], S.-H. LEE[1];
[1]Psychiatry, CHA Bundang Med. Ctr., Seongnam, Korea, Republic of; [2]Grad. Sch. of Clin. Counseling Psychology, CHA Univ., Seongnam, Korea, Republic of; [3]Psychiatry, CHA Ilsan Med. Ctr., Goyang, Korea, Republic of

Abstract: Motivation: Self-compassion involves taking an emotionally positive attitude towards oneself when suffering. Self-compassion has positive effects on psychological health as well as a protective role in preventing depression and anxiety in healthy individuals. However, little research has been conducted to show white matter (WM) connectivity in neuroimaging studies of self-compassion. Recent studies have demonstrated that the mirroring network can be associated with neurocognitive functions and emotion processing. We hypothesized that mirroring networks are related to levels of self-compassion in healthy individuals.**Methods**: Magnetic resonance imaging (MRI) data were obtained from 71 healthy participants, whose levels of self-compassion and its six components were measured with self-compassion scale (SCS). Voxel-wise correlation analysis was performed between total/six-subscale scores of SCS and fractional anisotropy (FA) values of mirroring network regions of interest (ROIs) using tract-based spatial statistics (TBSS). For controlling multiple correlation comparisons, Bonferroni correction was used. Exploratory correlation analysis was performed between the FA values of self-compassion-related WM regions and total scores of the Connor–Davidson resilience scale (CD-RISC)/brief resilience scale (BRS)/state-trait anxiety inventory (STAI).**Results**: We found that SCS total scores were negatively correlated with the FA values of the superior longitudinal fasciculus (SLF) [p <0.001 (FWE-corrected)]. Among six subscale scores of SCS, the mindfulness subscale scores were negatively correlated with the same regions [p =0.006 (FWE-corrected)]. FA values of SLF regions were found to be negatively correlated with CD-RISC and BRS total scores (r = -0.337, p =0.004; r = -0.336, p =0.004; respectively). The significantly positive correlation between FA values of SLF and STAI total scores did not survive the Bonferroni correction (r = -0.274, p

=0.038).**Conclusions**: Our findings suggest that nonjudgmentally mindful awareness of one's emotion may be negatively associated with the SLF regions involved in self-referential processing and
mind-wandering. These low WM microstructures can be associated with the prevention of negative emotions such as anxiety in healthy individuals and resources that help them resilient from stressful events.

Disclosures: Y. Hwang: None. **C. Pae:** None. **H. Jung:** None. **N. Kang:** None. **C. Park:** None. **M. Bang:** None. **M. Kim:** None. **S. Lee:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.27

Topic: H.04. Executive Functions

Support: CRC 1193

Title: Spatio-temporal dynamics of limited resource competition between emotion and cognition - evidence from an eeg/fem beamforming study

Authors: *A. DIETRICH[1], E. PINZUTI[2], Y. CABRAL-CALDERIN[3], F. MÜLLER-DAHLHAUS[4], M. WIBRAL[5], O. TÜSCHER[4,6];
[1]Goethe Univ. Frankfurt, MEG Unit, Brain Imaging Ctr. Frankfurt, Frankfurt am Main, Germany; [2]Systemic Mechanisms of Resilience, Leibniz Inst. for Resilience Res. (LIR), Mainz, Germany; [3]Neural and Envrn. Rhythms, Max Planck Inst. for Empirical Aesthetics, Frankfurt, Germany; [4]Dept. of Psychiatry and Psychotherapy, Univ. Med. Ctr. of the Johannes Gutenberg Univ. Mainz, Mainz, Germany; [5]Dept. of Data-driven Analysis of Biol. Networks, Univ. of Göttingen, Göttingen, Germany; [6]Systemic Mechanisms of Resilience, Leibniz Inst. for Resilience Res., Mainz, Germany

Abstract: As the brain's processing resources are finite, but multiple processes, such as emotion and cognition, might compete over such limited resources, an appropriate distribution of resources is required. In the past, emotion and cognition were considered separate systems, antagonistically competing over limited processing resources, with certain brain areas preferably processing either emotion or cognition. Recently this view is challenged by more integrative approaches, yet so far these hypotheses are lacking detailed neurophysiological information. In this study, we use emotion cognition interaction to investigate the neurophysiological and temporal dynamics of solving limited resource competition via an electroencephalography (EEG)/ Finite Element Modeling (FEM) beamforming approach in a large cohort (N=103) of healthy human subjects. Emotion-cognition interaction was measured via an emotional Eriksen Flanker task, where emotional task-irrelevant images preceded the flanker signal. Our source analysis reveals the right Inferior Frontal Gyrus (IFG) as a shared location for processing

emotional and cognitive information, with the strongest interaction effects in the beta frequency range in *pars triangularis*, thus requiring a distribution of limited resources. To investigate the temporal dynamics of such resource competition in right IFG, we used Support Vector Machine (SVM) analysis, revealing that at the transition time point between emotional and cognitive processing, the competition over processing resources is strongest, forcing the system to integrate both types of information. This occurs only for a brief point in time and is resolved afterwards, despite lingering emotional processing and even ramped up cognitive processing. Furthermore, the neurophysiological interaction at this transition time point is relevant for behavior as it correlates with reaction times and accuracy, with stronger interactions in the beta band having a negative impact on behavioral outcomes. Apparently, the brain tries to prevent this resource competition as the IFG establishes top-down influences on visual areas, as revealed by granger analysis. How well the brain can exert such top-down influences is fundamental for behavioral performance, as the connectivity measure correlates with the interference effect in the behavior. In summary, our findings reveal the neurophysiological and spatio-temporal dynamics of resource competition on source space with unprecedented detail, advancing the understanding of competition over limited resources in the human brain.

Disclosures: **A. Dietrich:** None. **E. Pinzuti:** None. **Y. Cabral-Calderin:** None. **F. Müller-Dahlhaus:** None. **M. Wibral:** None. **O. Tüscher:** None.

Poster

558. Emotion Processing in the Human Brain

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 558.28

Topic: G.01. Fear and Aversive Learning and Memory

Support: NWO VICI #453- 12-001
NWO Research Talent Grant #406-18-540

Title: Are there dissociable roles of the human BNST and amygdala in threat anticipation, confrontation and the switch to defensive action?

Authors: *F. KLUMPERS[1,2], A. M. HULSMAN[1,2], M. M. HASHEMI[2], R. KALDEWAIJ[2], W. ZHANG[2], A. Z. L. WESTER[2], V. A. VAN AST[2], S. KOCH[2], L. D. DE VOOGD[2], K. ROELOFS[1,2];
[1]Behavioural Sci. Institute, Radboud Univ., Nijmegen, Netherlands; [2]Donders Institute, Radboud Univ., Nijmegen, Netherlands

Abstract: Defensive responses in threatening situations are critical for survival. Aberrations in the mechanisms driving these responses may however lead to psychopathology, such as Post Traumatic Stress Disorder (PTSD). Previous work indicates a switch in neural activation may occur as a function of threat proximity: the Bed Nucleus Stria Terminalis (BNST) shows a relatively stronger response during threat anticipation but the amygdala shows a stronger reaction

for actual confrontation of the threat (Klumpers et al., 2017). However, there is ongoing debate about such a dissociation of the functional roles of the amygdala and BNST (Hur et al., 2020) and little is known about how these dissociations might support defensive actions. This study serves to provide more insight in the role of the BNST and amygdala, not only in passive fear but also defensive action. We used a large dataset (N=406; 110 females) in which police recruits and age- and gender-matched healthy controls were tested during a functional MRI emotional go/nogo task under threat of electric shock (Hashemi et al., 2019). This paradigm entails three consecutive phases: shock *anticipation*, the *switch to action* during which the participant needs to shoot or withhold from shooting and lastly *confrontation* when a shock is actually administered. Results support the hypothesized switch in neural activation throughout the different phases of the shooting task. BNST activity rises during anticipation and the switch to defensive action while neural activity assessed by BOLD switches sharply to the dorsal amygdala during confrontation. Additionally, a significant positive correlation between childhood maltreatment and amygdala activity was found during threat anticipation. These robust results replicate and extend earlier findings (Klumpers et al., 2017; Visser et al., 2021) to suggest a distinct functional role for the BNST and amygdala during defensive responding and that early life stress may change the balance of activity between these regions.

Disclosures: F. Klumpers: None. **A.M. Hulsman:** None. **M.M. Hashemi:** None. **R. Kaldewaij:** None. **W. Zhang:** None. **A.Z.L. Wester:** None. **V.A. Van Ast:** None. **S. Koch:** None. **L.D. De Voogd:** None. **K. Roelofs:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.01

Topic: G.07. Post-Traumatic Stress Disorder

Title: An educational review for healthcare providers on the psychiatric applications of ketamine and its metabolites

Authors: *T. CUTTING[1], R. E. HARTMAN[2];
[1]Loma Linda Univ., Loma Linda Univ., Loma Linda, CA; [2]Psychology, Loma Linda Univ., Temecula, CA

Abstract: We reviewed the published research on novel pharmacological interventions for the treatment of posttraumatic stress disorder (PTSD) and attenuation of stress-induced depressive-like behavior in humans and animal models. Multiple studies have demonstrated ketamine's efficacy in attenuating treatment resistant depression and PTSD. The stereoisomers of ketamine, *R*-ketamine and *S*-ketamine have also demonstrated efficacy in attenuation of stress and depressive symptoms, with *S*- ketamine gaining FDA approval for treatment resistant depression and suicidal ideation in 2019. Although *R*-ketamine has not yet seen FDA approval, its lack of psychotomimetic properties, as seen in ketamine and *S*-ketamine, may reduce its side effect

profile and abuse potential.We also reviewed more recent research revealing a prophylactic application for ketamine and *R*-ketamine in attenuating reactions to stress. Numerous studies indicate that prophylactic use of ketamine, along with *R*-ketamine and its hydroxynorketamine metabolites, can be effective in preventing depressive and fearful behavior induced by stress. According to current estimates, approximately 21 million adults experience at least one major depressive episode annually. PTSD is a potentially debilitating condition as well, affecting 6.8% of adults over the course of their lifetime. Additionally, suicide rates in the U.S. have been on the rise with a nearly 35% increase over the last 20 years. Traditional pharmacological treatments for depression and suicidality are limited in efficacy and have a delayed onset of therapeutic effects. Similarly, treatment options for those experiencing PTSD have historically been limited in scope and efficacy. Ketamine remains underutilized in its treatment potential for psychiatric disorders. In addition to attenuating current psychopathology, preventative treatments for PTSD remain unrealized in clinical application. Prophylactic treatments could potentially reduce rates of the disorder in first responders, military personnel, foreign aid workers, and other occupations where exposure to traumatic stress can be anticipated. We propose disseminating this review for the purpose of educating physicians and other healthcare providers on current uses and potential future applications of ketamine and its stereoisomers.

Disclosures: T. Cutting: None. **R.E. Hartman:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.02

Topic: G.07. Post-Traumatic Stress Disorder

Title: Stress-related gene sets identify potential pathophysiological changes in post-mortem PTSD frontal cortex

Authors: *K. A. YOUNG[1], M. J. GIRGENTI[2], D. A. CRUZ[3], B. R. HUBER[4], M. W. LOGUE[5], D. E. WILLIAMSON[6];
[1]Texas A&M Col. of Medicine/CTVHCS, Temple, TX; [2]Yale Univ. Sch. of Med., New Haven, CT; [3]Psychiatry & Behavioral Sci., Duke Sch. of Med., Durham, NC; [4]VA Boston Healthcare and Natl. PTSD Brain Bank, Boston, MA; [5]Natl. Ctr. for PTSD, VA Boston Healthcare Syst. and Boston Univ. Schools of Med. and Publ. Hlth., Boston, MA; [6]Psychiatry & Behavioral Sci., Duke Sch. of Med. and Durham VA HCS, Durham, NC

Abstract: Three well-provisioned post-mortem gene expression studies of PTSD have recently been performed, using different tissue preparations and data analytic pipelines (Girgenti 2021 PMID:33349712, Lougue 2021 PMID:34646915, Jaffe 2022 bioRxiv 2021.01.12.426438). We performed gene set enrichment analysis (GSEA) on each mixed-sex dataset using probability ranked DEGs. Analysis of Hallmark gene sets identified ANDROGEN_RESPONSE, HYPOXIA, ANGIOGENESIS, HEME_METABOLISM, GLYCOLYSIS,

PROTEIN_SECRETION and MYC_TARGETS_V1 as common up-regulated gene sets in PTSD. In addition, Reactome analysis indicated high RNA metabolism as an additional activated function in PTSD. ANDROGEN_RESPONSE was the most consistently up-regulated Hallmark gene set, with FDR q values lower than 0.006 across all three sets. Examination of jointly up-regulated genes within the leading edge of the ANDROGEN_RESPONSE sets found 11 common up-regulated genes in PTSD (AKAP12, ARID5B, ELL2, ALDH1A3, PA2G4, ABHD2, ZMIZ1, NKX3-1, INPP4B, XRCC5 and BMPR1B). Several of these genes are involved in cancer invasiveness and/or motility, including the most consistently up-regulated transcript in the androgen set in PTSD, AKAP12 (gravin). AKAP12 was significantly FDR up-regulated in PTSD in all four cortical regions in Girgenti, in the medial (but not basolateral) amygdala in Jaffe and nominally up-regulated in MDD cortex. AKAP12 is found in perivascular cells in the CNS and is involved in enhancing and repairing the blood brain barrier after ischemic damage, although exact mechanisms and cell-type expression after vascular stress in humans is not well described. In the mouse, this transcript is mainly found in endothelial cells, while it is not selectively expressed in the primary human brain cell types. Interestingly, AKAP12, a multi-functional kinase and calmodulin anchoring protein, has been identified as an important component of some synaptic structures in neurons, where its expression is associated with adrenergic signaling, synaptic plasticity and learning/memory. These findings highlight potentially interlinked cortical functions (hypoxia, androgen response, angiogenesis) that may be consistently activated by uncharacterized stressors in PTSD brain, and a potential new target affecting PTSD pathophysiology and neuronal plasticity, AKAP12. In addition to PTSD/MDD, we cannot rule out the possibility that the present findings may be related clinical and/or abuse factors that are poorly controlled in post-mortem psychiatric human cohorts, including psychiatric medications, sleep disturbances, obstructive sleep apnea, hypertension and drug/alcohol abuse.

Disclosures: K.A. Young: None. **M.J. Girgenti:** None. **D.A. Cruz:** None. **B.R. Huber:** None. **M.W. Logue:** None. **D.E. WIlliamson:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.03

Topic: G.07. Post-Traumatic Stress Disorder

Support: Centre Thématique de Recherche en Neurosciences : Neuro COVID-19

Title: The "echo" effect of COVID-19 on healthcare workers: a study to oversee the impact of the preventive measures and risk perception on mental health and inflammation.

Authors: *C. CANIVET[1], É. BOILARD[1,2], C. MERETTE[2,3], C. M. MORIN[3,2], J. DESLAURIERS[1,2];
[1]Ctr. de Recherche du CHU de Québec-Université Laval, Québec, QC, Canada; [2]Univ. Laval, Québec, QC, Canada; [3]Ctr. de recherche CERVO, Québec, QC, Canada

Abstract: Background: During the COVID-19 pandemic, the regional authorities provided hospitals with guidelines to prevent infection among the healthcare workers. How the perception of the implemented measures affect their psychological health and stress remains unknown. Moreover, numerous studies show a relationship between inflammatory status and work-related distress. Brain-derived exosomes (BDEs) are extracellular vesicles that may be produced by CNS cells, cross the BBB, then passively release cytokines in the blood. These BDEs may be potential targets to blood identify CNS-specific biomarkers. We hypothesized that specific measures implemented are critical to prevent distress in healthcare workers and that inflammation from BDEs are associated with risk to develop psychiatric symptoms.
Methods: Fifteen participants participated to this study. Inclusion criteria were: 18-59 years-old man or woman; and working full-time as a CHU de Québec healthcare provider (nurses, therapists, etc.) during the COVID-19 crisis. All participants completed four self-report surveys three times (1st visit, 3 and 6 months later): Kessler psychological distress (K10); perception of risk and preventive measures; impact of event scale-revised; and MBI-Emotional Exhaustion Scale. Blood sample was drawn for enrichment of BDEs. For each visit, we divided the participants based on their K10 score: no/low vs moderate/severe psychological distress. Welch-corrected t test was used for statistical analyses.
Results: Participants with moderate/severe psychological distress tend to experience intrusion symptoms ($p=0.07$) and were more at risk to develop exhaustion ($p\leq0.01$) than those with low distress. On the 2nd visit, the same group was still at elevated risk for exhaustion ($p\leq0.01$), presented higher hyperarousal score ($p\leq0.01$), and tends to have poorer perception of implemented measures (p=0.08) *vs* participants with low distress. On the 3rd visit, participants with moderate/severe distress displayed higher hyperarousal score ($p\leq0.05$), posttraumatic symptoms ($p\leq0.05$), job-related stress ($p\leq0.05$) and risk of exhaustion ($p\leq0.005$) *vs* those with low distress.
Conclusion: Altogether these findings show that healthcare workers with a moderate/severe psychological distress exhibit increased intrusion and hyperarousal symptoms, and are at high risk for emotional burnout. Moderate or severe psychological distress is associated with high job-related stress and negative perception of the implemented measures. Complementary analyses, insomnia data, and measurements of circulating levels of inflammatory markers from brain-derived exosomes will also be presented.

Disclosures: C. Canivet: None. **É. Boilard:** None. **C. Merette:** None. **C.M. Morin:** None. **J. Deslauriers:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.04

Topic: G.07. Post-Traumatic Stress Disorder

Support: Glenn Greenberg and Linda Vester Foundation

Title: Multi-factoral analysis of neural and behavioral correlations in post traumatic stress disorder (PTSD)

Authors: *R. WALTERS[1], B. SHEN[2], E. TELL[1], B. MOALLEM[3], L. DOAN[4], C. M. RAIO[6], K. LOUIE[7], M. R. MILAD[5], P. W. GLIMCHER[8];
[1]Neurosci., [2]Neurosci. Inst., [3]Psychiatry, [4]Anesthesiol., [5]Dept. of Psychiatry, NYU Grossman Sch. of Med., New York, NY; [6]Psychiatry, NYU Sch. of Med., New York, NY; [7]Ctr. for Neural Sci., [8]Neural Sci., New York Univ., New York, NY

Abstract: Post-Traumatic Stress Disorder (PTSD) is a debilitating anxiety disorder that impacts 6-9% of the US population, with no effective cure. Exposure to traumatic events is unique to the human condition and is an etiologic factor in this disorder. The multifactorial nature of PTSD makes it difficult to understand the mechanistic neural changes involved in the disorder. Few studies have combined learning and reward-related neural analyses with deeply-profiled behavioral data. Promising meta-analyses in PTSD neuroimaging have shown hyperactivated network activity in the amygdala, the dorsal anterior cingulate cortex (dACC), and hypoactivity in the ventral medial prefrontal cortex (vmPFC), and inferior frontal gyrus, amongst others. It is known that this altered activity is observed when subjects are performing fear-related tasks and engaging in reward-processing systems. Our long-term goal is to understand how non-pharmacological interventions like stellate ganglion block impede sympathetic outflow to the brain. To progress toward this goal, we will analyze the correlations between a rich profile of states and traits and the learning-reward network before treatment.
Individual patients diagnosed with PTSD who have a Clinically Administered PTSD Scale for DSM-5 (CAPS) score greater than or equal to 26 were enrolled. We then gathered data on each subject using the CSSR-S, PHQ-9, three modules of the PROMIS (physical function, sleep disturbance, fatigue), the Anxiety Sensitivity Index (ASI), STAI, Intolerance of Uncertainty Scale, Anxiety Control Questionnaire (ACQ-r), Perceived Stress Scale (PSS), Fagerstrom Nicotine Dependence Inventory, WHOQOL-Bref, PCL-5, Life-Events Checklist (LEC-5), and TLFB alcohol and drug.
Patients were then scanned using functional MRI (fMRI) while performing an emotional face processing task, fear conditioning and extinction learning task (Milad and Quirk, 2012), and an aversive decision-making task. While all tasks were being performed, skin conductance data were also collected. Finally, we gathered salivary cortisol and alpha-amylase both pre and post-scan. Forthcoming correlational analyses should provide novel linkages and lay the groundwork for a better understanding of treatment mechanisms.
This work was supported by the Glenn Greenberg and Linda Vester Foundation.

Disclosures: R. Walters: None. **B. Shen:** None. **E. Tell:** None. **B. Moallem:** None. **L. Doan:** None. **C.M. Raio:** None. **K. Louie:** None. **M.R. Milad:** None. **P.W. Glimcher:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.05

Topic: G.07. Post-Traumatic Stress Disorder

Support: ACTRI

Title: Effect of Chronic Cannabis use on PTSD symptomatology.

Authors: *B. CUCCURAZZU[1,2], C. D. NAPAN[1,2], D. T. ACHESON[1,2], V. B. RISBROUGH[1,2], D. M. STOUT[1,2];
[1]Univ. of California San Diego, San Diego, CA; [2]VA Ctr. of Excellence for Stress and Mental Hlth., San Diego, CA

Abstract: Post-traumatic stress disorder (PTSD) is a common psychiatric disorder manifested as hyperarousal, anxiety, depressive symptoms, and sleep disturbances. Evidence-based treatments for PTSD have only partial efficacy (with remission rates reported to range from 20% to 30%) and can result in numerous side-effects and early discontinuation. In the past 10 years, a growing interest has developed in the use of cannabis and synthetic cannabinoids as medications for PTSD. However in rodents and healthy humans chronic use of cannabinoid receptor 1 agonists can cause downregulation of these receptors, with loss of critical functions such as fear extinction. Loss of fear extinction could severely effect natural recovery from trauma as well as interfere with extinction-based therapies for PTSD. Here we evaluated whether cannabis use modulates fear extinction in PSTD. Ninety-four volunteers grouped according to their cannabis use and PTSD diagnosis were tested in a fear conditioning and extinction task. On day 1, the volunteers were assessed for PTSD symptoms using the Clinician-Administered PTSD Scale for DSM-5 (CAPS-5), and cannabis use was recorded using a modified version of the Customary Drinking and Drug Use Record (CDDR). Subjects then underwent fear learning acquisition and extinction sessions. On day 2, subjects returned for assessment of extinction recall. Participants were screened for acute intoxication before testing to ensure behavioral effects were not caused by acute cannabis use. There was no effect of cannabis use or PTSD on fear learning. However there was a significant interaction ($P<0.01$) between PTSD symptoms and cannabis use on extinction recall. Individuals with PTSD plus high cannabis use (1g/day for at least 90 days) (N=28) had worse recall compared to all other groups (No PTSD/No cannabis, N=29; No PTSD/Cannabis, N=18; PTSD/No cannabis, N=20). PTSD symptom severity was also reduced in the PTSD/Cannabis group compared to the PTSD/No cannabis group. Non-aversive memory was differentially associated with PTSD and cannabis use, with the No PTSD/Cannabis group showing worse memory performance compared to No PTSD/No cannabis and PTSD/Cannabis group. These results indicate that despite improvements in PTSD symptom severity, chronic cannabis use may reduce fear extinction in PTSD patients.

Disclosures: B. Cuccurazzu: A. Employment/Salary (full or part-time):; University California San Diego. **C.D. Napan:** A. Employment/Salary (full or part-time):; University of California San Diego. **D.T. Acheson:** A. Employment/Salary (full or part-time):; VA San Diego Healthcare System. **V.B. Risbrough:** A. Employment/Salary (full or part-time):; University of California San Diego, VA San Diego Healthcare System. **D.M. Stout:** A. Employment/Salary (full or part-time):; University of California San Diego.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.06

Topic: G.07. Post-Traumatic Stress Disorder

Support: NIH Grant R01MH124761
NIH Grant UH3NS107673
NIH Grant F31MH127922

Title: Electrophysiological responses in the human amygdala during individual trauma reminders in treatment-resistant post-traumatic stress disorder

Authors: *J. A. SCHNEIDERS[1,2], J. L. GILL[1], M. STANGL[1], S. HILLER[1], M. VALLEJO[1], U. TOPALOVIC[1], C. S. INMAN[3], V. R. RAO[4], M. VICK[5], N. R. HASULAK[5], S. E. KRAHL[2], J. W. CHEN[2], M. S. FANSELOW[1], M. G. CRASKE[1], N. A. SUTHANA[1], J.-P. LANGEVIN[1,2], R. J. KOEK[1,2];
[1]Univ. of California Los Angeles, Los Angeles, CA; [2]VA Greater Los Angeles Healthcare Syst., Los Angeles, CA; [3]Univ. of Utah, Salt Lake City, UT; [4]Univ. of California, San Francisco, San Francisco, CA; [5]NeuroPace Inc., Mountain View, CA

Abstract: Post-traumatic stress disorder (PTSD) is an incapacitating psychiatric disorder that can be treatment-resistant especially after severe traumatic experiences such as combat trauma. For people with treatment-resistant PTSD (tr-PTSD), traditional therapy approaches, such as medication or trauma-focused psychotherapy, do not lead to sufficient symptom improvement. A promising novel approach for tr-PTSD is responsive neurostimulation, which aims to directly target abnormal neural activity through stimulation of target brain regions with electrical current. Responsive neurostimulation, however, requires a reliable biomarker (i.e., a symptom-related neural signature in a specific brain region), that can be continuously monitored and that can trigger stimulation upon its detection. The present study aims to identify a suitable biomarker for responsive neurostimulation to mitigate tr-PTSD symptoms. We recorded intracranial electroencephalographic (iEEG) data in two individuals with intracranially implanted electrodes in the amygdala for a clinical trial to investigate the effectiveness of responsive neurostimulation in tr-PTSD. Oscillatory activity in the amygdala was recorded during exposure of both participants to individualized trauma reminders. Specifically, we used a script-driven imagery task during which the participants listened to audio-recorded narratives of their most severe trauma, and of a personal pleasant experience as a control condition. Participants reported increased PTSD symptom severity and anxiety during traumatic compared to pleasant individual reminders. Traumatic reminders were further accompanied by a significant increase of oscillatory power in the amygdala within the theta frequency band (5-9 Hz). These results suggest that increased theta activity in the amygdala could serve as a biomarker of an enhanced fear state during acute tr-PTSD symptoms. Future studies will investigate the suitability of this biomarker for responsive neurostimulation to mitigate acute and long-term PTSD symptoms and to measure treatment response of affected individuals.

Disclosures: **J.A. Schneiders:** None. **J.L. Gill:** None. **M. Stangl:** None. **S. Hiller:** None. **M. Vallejo:** None. **U. Topalovic:** None. **C.S. Inman:** None. **V.R. Rao:** F. Consulting Fees (e.g.,

advisory boards); NeuroPace, Mountain View, CA. **M. Vick:** A. Employment/Salary (full or part-time):; NeuroPace Inc., Mountain View, CA. **N.R. Hasulak:** A. Employment/Salary (full or part-time):; NeuroPace Inc., Mountain View, CA. **S.E. Krahl:** None. **J.W. Chen:** None. **M.S. Fanselow:** None. **M.G. Craske:** None. **N.A. Suthana:** None. **J. Langevin:** None. **R.J. Koek:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.07

Topic: G.07. Post-Traumatic Stress Disorder

Title: Neurophysiological underpinnings of long-term exposure to war trauma.

Authors: *J. V. PINTO[1,2];
[1]Univ. of Sydney, Sydney, Australia; [2]Sync Body-Brain Hlth., Brisbane, Australia

Abstract: War and conflict impact millions of people worldwide, but the neurophysiological consequences of long-term exposure to trauma are unknown. A few studies have examined biomarkers of posttraumatic stress disorder (PTSD) in quantitative electroencephalography (qEEG), in most instances documenting unique patterns of neurophysiological brain activity, but results are inconsistent. This study investigated whether war refugees with and without clinical and subclinical PTSD would show abnormal qEEG activity. Moreover, it examined associations between qEEG markers and PTSD symptom severity. The hypothesis is that qEEG differences will be found between the PTSD group and controls, and that PTSD symptoms will predict observed differences. African war refugees in Australia were recruited for this study. Participants were assigned to a clinical and subclinical PTSD group (n = 35) and a trauma-exposed healthy control group (n=35). The final sample (n = 70) included Liberian (n = 22), Sudanese (n = 25), and Congolese (n = 23) refugees, consisting of 31 males and 39 females aged 18 - 54 years (M = 33.64, SD = 10.54). In the PTSD group, 28.6% reported experiencing PTSD symptoms for the past 6-15 years, 65.7% for the past 16-30 years, and 5.7% reported persistent symptomatology for over 30 years as a result of long-term traumatic exposure. All participants completed the Clinician-Administered PTSD Scale for DSM-5 (CAPS-5) and resting-state EEG recordings. Scalp EEG was recorded with the Geodesic's 32-Channel HydroCel net according to the standard 10-20 international system. Recordings were plotted into a frequency power spectrum using Fast-Fourier Transform (FFT) for all bands and brain regions. Independent t-tests and regression analyses were used to identify potential biomarkers and symptom associations. Results support the hypothesis of distinct EEG patterns among the PTSD group, which were observed in frontal, parietal, occipital, and temporal regions. Increased theta and beta activity in PTSD ($p < .05$) support previous studies, but findings in higher alpha power do not. Intrusion, avoidance, and negative mood and cognition symptoms strongly predicted higher theta and alpha power ($p < .005$). No symptom associations were found for higher beta activity, however, this marker was higher among females in the PTSD group. This is the first study to demonstrate how

long-term exposure to trauma and chronic PTSD may impact brain activity among African war refugees. These results may serve as parameters to inform the development of portable precision diagnostic tools for resource-poor regions worldwide impacted by war and other emergencies.

Disclosures: J.V. Pinto: None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.08

Topic: G.07. Post-Traumatic Stress Disorder

Support: CatalystGrant100718

Title: Assessment of Oxidative Stress and Neuronal Activity Affected in PTSD Subjects

Authors: *A. SUSNJAR, G. NOSSA, U. DYDAK, J. RISPOLI, M. ISAAC LAM;
Purdue Univ., West Lafayette, IN

Abstract: The applications of MRS to investigate the metabolic window on a wide range of biochemical processes are extremely diverse. MRS was the first tool that demonstrated biological changes in mental health patients, namely imbalances in brain metabolism, changing the stigma of mental health. However, research evidence for altered in vivo metabolite levels across post-traumatic stress disorder (PTSD) are lacking. This study evaluates edited and unedited single voxel spectroscopy (SVS) of ten clinically diagnosed PTSD participants and their age and gender-matched healthy controls. Four brain regions (anterior cingulate cortex (ACC), dorsolateral prefrontal cortex (DLPFC), insula, hippocampus related to the underlying PTSD symptoms have been investigated for assessment of alterations in cognitive processing, such as lack of extinction of the fear response, flashbacks, heightened physiological responses to trauma, and general hyperarousal symptoms and hypervigilance. Fifth brain region, the amygdala, was only investigated using unedited MRS due to difficulty in acquiring usable and reliable edited MRS. Unedited SVS was obtained using Point RESolved Spectroscopy (PRESS), processed in Osprey and analyzed using LC Model. Edited MRS was obtained using HERMES and analyzed using Gannet. To overcome acquisition downfalls, we have conducted critical optimization for each voxel of interest (VOI) by finding the optimal voxel rotation and slice order selection to enhance data quality and reproducibility. Amygdala is a core component in neurobiological models of stress-related pathologies. We found statistically significant changes in PTSD participants compared to healthy controls indicating neurotrauma with elevated glutamate-glutamine (Glx), and creatine (tCr) values. Additionally, a decrease in choline (tCho) seems to indicate neurotrauma related metabolic changes in the brain region in charge of “flight or fight” state. Statistically significant decrease is observed in tCho, a cell membrane turnover marker, in four out of five brain regions. Edited MRS depicted the important role of three brain regions in PTSD. Lower concentration of GABA in the hippocampus is a biological marker of vulnerability

for development of mood disorders, anxiety, depression, and insomnia which are common symptoms of PTSD. While the findings for Glx levels are somewhat inconsistent, a meta-analysis suggests that Glx concentration are increased in frontal brain regions in neurological disorders, confirmed by our findings in the ACC. Glutathione, an antioxidant, is decreased in DLPFC of PTSD subjects due to an active protection against toxicity.

Disclosures: A. Susnjar: None. **G. Nossa:** None. **U. Dydak:** None. **J. Rispoli:** None. **M. Isaac Lam:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.09

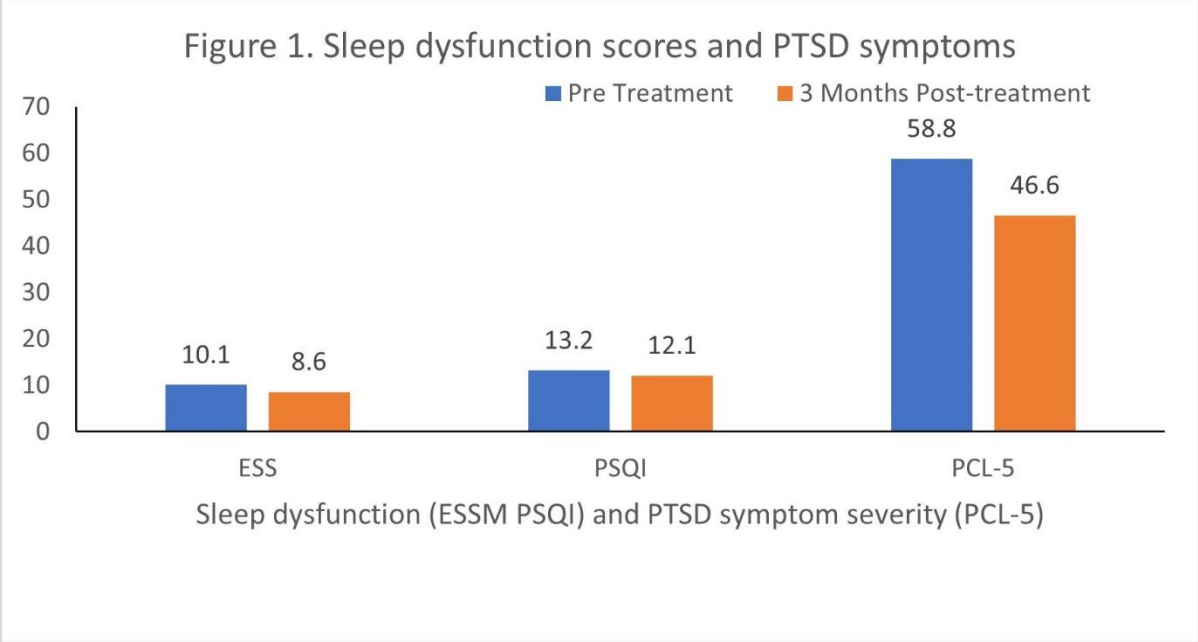
Topic: G.07. Post-Traumatic Stress Disorder

Title: Treatment of sleep disorders leads to amelioration of clinical severity and pro-inflammatory cytokines in combat veterans with Post Traumatic Stress Disorder (PTSD): A small cohort study

Authors: *V. SINGH[1,5], H. THAKUR[2], A. DE[3], S. MAALOUF[3], J. ZHOU[3], V. BOINPELLY[3], M. CHEN[3], P. THAPA[3], D. BURNS[3], Y. VAYSBERG[3], R. SHARMA[4], M. SHARMA[3]; [1]Kansas City VA Med. Ctr., [2]Psychiatry, [3]Neurobio. Res. Lab., [4]Res. Service, Kansas City VA Med. Ctr., Kansas City, MO; [5]Neurol., Univ. of Kansas Med. Ctr., Kansas City, KS

Abstract: Introduction: PTSD and sleep dysfunction are very common in US combat veterans with a bidirectional relationship. Chronic stress response associated with PTSD and sleep disturbance leads to immune endocrine changes and increased production of cytokines. **Aims:** To determine correlations between the severity of PTSD and sleep dysfunction symptoms and circulating levels of inflammatory cytokines and to evaluate if treating sleep dysfunction in treatment-naïve PTSD patients with standard sleep agents is associated with decreases in the circulating levels of cytokines and severity of PTSD symptoms. **Methods**: Nineteen treatment naïve veterans with PTSD and concurrent sleep disturbance were recruited for treatment with sleep agents for 3 months. Patients with sleep apnea, preexisting inflammatory conditions and on immunomodulatory drugs were excluded. Severity of sleep dysfunction was assessed before and at the end of treatment period using Epworth Sleepiness Score (ESS) and Pittsburgh Sleep Quality Inventory (PSQI) questionnaires and PTSD severity was assessed using PTSD Check List (PCL-5). Blood samples were obtained to assess serum cytokine levels using human cytokine plex-65 discovery assay. **Results:** Initiation of sleep agents led to improvement in sleep measures and clinically significant improvement in PTSD measure as demonstrated by decrease in scores (Figure 1). Out of 65 cytokines measured by multiplex assay, 22 cytokines were downregulated, and one was upregulated (p-value <0.05) Table 1.

Table 1: Effect of sleep promoting agents on serum cytokine panel	
Cytokines tested using plex-65 discovery assay	**Cytokines showing significant decrease at the end of the 3-month period (p value <0.05)**
EGF, FGF, Eotaxin-1, TGF-a, G-CSF, Flt-3L, GM-CSF, Fractalkine, IFNa2, IFNy, GRO, alpha, IL-10, MCP-3, IL-12P40, MDC, IL-12P70, PDGF-AA, IL-13, PDGF-BB, IL-15, sCD40L, IL-17A, IL-1RA, IL-1a, IL-9, IL-1B, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IP-10, MCP-1, MIP-1a, MIP-1B, RANTES, TNFa, TNFB, VEGF-A, IL-18, Eotaxin, MCP-2, BCA-1, MCP-4, I-309, IL-16, TARC, 6CKine, Eotaxin-3, LIF, TPO, SCF, TSLP, IL-33, IL-20, IL-21, IL-23, TRAIL, CTACK, SDF-1a+B, ENA-78, MIP-1d, IL-28A.	GM-CSF, Fractal K, MCP3, IL-13, IL-15, IL-9, IL-1b, IL-3, IL-5, IL-6, MIP1a, MIP1b, TNFb, I309, 6CKine, SDF1a+b, G-CSF, IL-1A, LIF, IL-20, IL-21, IL-28A.



Disclosures: **V. Singh:** None. **H. Thakur:** None. **A. De:** None. **S. Maalouf:** None. **J. Zhou:** None. **V. Boinpelly:** None. **M. Chen:** None. **P. Thapa:** None. **D. Burns:** None. **Y. Vaysberg:** None. **R. Sharma:** None. **M. Sharma:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.10

Topic: G.04. Emotion

Support: 7R01MH118239-03
1R01DA052453-01A1

Title: Investigating miRNAs' role in the neuropathology of Major Depression in a large sample of postmortem brains.

Authors: *Z. TAYLOR[1], J. DRAKE[1], A. DENHAM[1], S.-A. BACANU[2], J. SHIN[3], J. KLEINMAN[3], T. HYDE[3], V. I. VLADIMIROV[4];
[1]Texas A&M Univ. Syst. Hlth. Sci. Ctr., Bryan, TX; [2]Virginia Inst. for Psychiatric and Behavioral Genet., Virginia Commonwealth Univ., Richmond, VA; [3]Lieber Inst. for Brain Development, Johns Hopkins Univ., Baltimore, MD; [4]Psychiatry, Texas A&M Univ., College Station, TX

Abstract: Major Depression (MD) is a debilitating disorder characterized by low mood and anhedonia that affects roughly one out of every six adults worldwide. Several large GWAS studies have already identified genome-wide significant variants associated with MD. However, little can be said about the functional impact of these variants from the GWAS alone. A complementary approach to understanding the underlying neuropathology of MD involves identifying transcriptome changes associated with major depression in postmortem brain tissues. MicroRNA (miRNA), a class of small non-coding RNA with important gene regulatory functions and high expression in the brain, has been studied in relation to neuropsychiatric phenotypes. In this study, we used miRNA-Seq to compare the expression of 947 miRNAs between 150 MD patients and 150 matched controls in the subgenual anterior cingulate cortex (sACC) and in the amygdala. We applied statistical and bioinformatic analyses, e.g., single miRNA differential expression analysis (DEA), weighted gene co-expression analysis (WGCNA), expression quantitative trait loci (eQTL), and miRNA/mRNA correlation-based analyses to detect a set of miRNAs with a converging role in the etiology of MDD. In the sACC We identified 50 miRNAs whose expression was associated with MD (at FDR of 5%). These included miRNAs that were both previously associated with MD and unique to this study as well as miRNAs associated with neurodevelopment or other psychiatric illness. The network analyses detected two significant miRNA modules associated with MD at the Bonferroni corrected p &lt 0.05, the module eigengenes (ME) of which were also negatively correlated with mRNA modules significantly associated with MD. Our eQTL analysis identified 86 significant cis- and 8 trans-eQTLs modulating the expression of these miRNAs. We further showed the eQTLs for these miRNAs are enriched for signals in GWAS of MD. Lastly, we note A to I RNA editing for a number of our top miRNAs highlighting additional biological mechanisms by which miRNA contribute to neuropathology of MD. In conclusion ours is the largest to date postmortem brain miRNA expression study of major depression, and our ongoing analyses provides solid evidence of the importance of miRNA as a contributing factor to the development of MD.

Disclosures: **Z. Taylor:** None. **J. Drake:** None. **A. Denham:** None. **S. Bacanu:** None. **J. Shin:** None. **J. Kleinman:** None. **T. Hyde:** None. **V.I. Vladimirov:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.11

Topic: G.06. Anxiety Disorders

Support: NRF-2019M3C7A1032262
NRF-2021M3E5D9025026

Title: The sex-different effect of oxytocin receptor gene methylation on cerebral gray matter structures in patients with panic disorder

Authors: S. CHUNG[1], H.-J. KIM[2], H.-Y. JUNG[2], Y.-G. HWANG[2], C. PARK[2], B. KIM[2], K. LEE[2], T. CHOI[2], S.-H. LEE[2], ***M. BANG**[2];
[1]CHA Univ. Sch. of Med., Pocheon, Korea, Republic of; [2]Dept. of Psychiatry, CHA Bundang Med. Ctr., Seongnam, Korea, Republic of

Abstract: Background: Panic disorder (PD) is sexually dimorphic in its clinical manifestation and disease course. Recent evidence suggests that sex differences in PD may result from the different brain responses to fear and anxiety between men and women. Considering oxytocin's role in the regulation of socio-emotional behaviors, epigenetic alterations in the oxytocin receptor gene (*OXTR*) could be one possible mechanism underlying sexual dimorphism in PD. We aimed to investigate the sex difference in cerebral gray matter (GM) volume and its association with *OXTR* methylation in patients with PD. **Methods:** Forty-six patients with PD (24 women) and 55 healthy controls (HCs; 31 women) underwent a high-resolution T1-weighted magnetic resonance imaging scan and provided peripheral blood samples at baseline. Cerebral GM volume differences were examined using FreeSurfer's surface-based general linear model analysis. *OXTR* methylation was performed using the pyrosequencing method. The severity of PD symptoms was assessed using the Panic Disorder Severity Scale (PDSS) and Albany Panic and Phobia Questionnaire (APPQ). **Results:** Patients with PD showed smaller GM volumes in the right frontal pole, post-central gyrus, superior temporal gyrus, and parahippocampal gyrus compared to HCs independent of sex. An interaction effect of sex and PD diagnosis was found in the left precuneus; its volume was higher in women than men in patients with PD and vice versa in HCs. *OXTR* methylation, which was decreased in patients with PD than in HCs irrespective of sex ($F = 37.2$, $p < 0.001$), showed a significant negative correlation with the precuneus volume in women ($r = -0.329$, $p = 0.014$), but not in men ($r = 0.191$, $p = 0.204$). Higher precuneus volume was correlated with more severe agoraphobia in women with PD ($\rho = 0.448$, $p = 0.042$), whereas an inverse correlation was found in men with PD ($\rho = -0.566$, $p = 0.009$). **Discussion:** Our findings suggest that epigenetic alterations of *OXTR* contribute to structural brain changes in the precuneus underlying the sexual dimorphism in PD. Furthermore, the inverse association

between the precuneus volume and severity of agoraphobia in women and men with PD may provide a clue to understanding the neurobiology of the sex-different manifestation of PD. Further research is required to figure out the pathogenesis of PD with an integrated picture of the oxytocin system in men and women.

Disclosures: S. Chung: None. **H. Kim:** None. **H. Jung:** None. **Y. Hwang:** None. **C. Park:** None. **B. Kim:** None. **K. Lee:** None. **T. Choi:** None. **S. Lee:** None. **M. Bang:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.12

Topic: G.06. Anxiety Disorders

Support: NIH ZIA-MH002781
NIH ZIA-MH002782

Title: Naturalistic fear induction reveals aberrant fear-circuitry function during threat anticipation in anxious youth

Authors: *L. JETT[1], G. RINGLEIN[1], J. GALBRAITH[1], A. HARREWIJN[2], S. QI[1], A. ZUGMAN[1], A. WINKLER[1], D. PINE[1], R. ABEND[1];
[1]NIH, Natl. Inst. of Mental Hlth. (NIMH), Bethesda, MD; [2]Erasmus Univ. Rotterdam, Rotterdam, Netherlands

Abstract: Anxiety disorders are characterized by aberrant threat anticipation. While most research on aberrant responses in anxiety uses trial-based tasks, naturalistic paradigms may be more ecologically valid for studying psychopathological mechanisms. Such research is particularly needed in youth with anxiety, prior to later compounding psychopathology. We used a child-appropriate scary movie to evoke a threat anticipation state during functional neuroimaging to assess fear circuitry function in youth with anxiety disorders relative to healthy youth. 54 subjects, including 25 anxiety patients and 29 healthy controls [F/M=30/24, age M(SD)=14.43(2.26) years] underwent a baseline resting-state scan, followed by a 6-minute scary movie. Cognitive (self-reported fear; assessed between scans) and physiological (skin conductance, SC; recorded continuously) fear responses were recorded. Functional connectivity data were preprocessed with fMRIPrep and analyzed using SPM12-based CONN. Based on prior translational research, we defined a fear circuitry encompassing cortical and subcortical regions of interest (ROIs). We performed network-based connectivity statistics (multivariate pattern analysis omnibus test; $p < .005$ edge-wise and $p < .05$ FDR cluster-level thresholds) in a mixed design (group as between-subjects factor; scan as within-subject factor), controlling for age and sex. Results indicated that the movie elicited greater cognitive and physiological fear responses in anxious relative to healthy youth. We found no significant group differences in connectivity during the baseline resting-state scan, suggesting comparable intrinsic connectivity patterns. In

contrast, significant group differences emerged in the change in connectivity between baseline and movie. Anxiety patients showed a greater increase in connectivity within a distributed cortical-subcortical network that included the bed nucleus of the stria terminalis and central and basolateral amygdala, and cortical regions such as posterior cingulate cortex and ventromedial prefrontal cortex, and additional regions. Nodes in this network have been implicated in extensive fear and defensive response research. Here, we used naturalistic threat-anticipation induction and network statistics to characterize patterns of aberrant function in this circuitry in pathological pediatric anxiety. These findings can guide translational neuroscience and clinical research on psychopathological mechanisms in anxiety and potential treatment approaches.

Disclosures: **L. Jett:** None. **G. Ringlein:** None. **J. Galbraith:** None. **A. Harrewijn:** None. **S. Qi:** None. **A. Zugman:** None. **A. Winkler:** None. **D. Pine:** None. **R. Abend:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.13

Topic: G.06. Anxiety Disorders

Support: National Research Foundation of Korea(NRF) grant funded by the Korea government (MSIT) (No. 2019R1A2C1086581)

Title: Increased cortical thickness in a frontoparietal network and decreased thickness in the mesial structure in social anxiety disorder

Authors: D. LEE[1], Y.-H. JUNG[1], S. KIM[2], J. JANG[3], U. YOON[2], ***S.-H. CHOI**[3]; [1]Dept. of Psychiatry, Seoul Natl. Univ. Hosp., Seoul, Korea, Republic of; [2]Daegu Catholic Univ., Daegu, Korea, Republic of; [3]Seoul Natl. Univ. Col. of Med., Seoul, Korea, Republic of

Abstract: Social anxiety disorder (SAD) is characterized by fear and avoidance of social situations. Although functional changes of the limbic area for emotional hyper-reactivity and emotional dysregulation are well documented, prior neuroimaging studies on structural changes have shown mixed results. A total of 35 patients with SAD and 42 matched healthy controls underwent structural magnetic resonance imaging. A high-resolution anatomic whole-brain scan was acquired using a T1-weighted inversion recovery 3-dimensional fast-spoiled gradient echo sequence. For a comparison of vertex-based analysis, we calculated local averaged cortical thickness of each subject using automatic parcellation, based on an automated surface registration algorithm and a surface template on which regional boundaries were defined manually. Cortical thickness was increased in the left insula, left superior parietal lobule, left superior temporal gyrus, and left frontopolar cortex in patients with SAD than healthy controls. Cortical thinning was shown in the left medial prefrontal cortex and the left fusiform gyrus in patients with SAD (FDR corrected $p < .05$, brain size entered as a covariate). Partial correlation analyses with brain size and age as covariates revealed that earlier age of disease onset was

associated with smaller volumes of the left medial prefrontal cortex (r = .411) and left fusiform (r = .431) in patients (all, FDR uncorrected p < .05). These findings are in line with prior functional and structural alterations of salience and frontoparietal networks for attentional functions, and autobiographical and social structures for socioemotional processing in patients with SAD.

Disclosures: D. Lee: None. **Y. Jung:** None. **S. Kim:** None. **J. Jang:** None. **U. Yoon:** None. **S. Choi:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.14

Topic: G.06. Anxiety Disorders

Title: Mental distress among the Mongolian general population

Authors: T. AMARTUVSHIN[1], T. HIRAMOTO[2], B. LKHAGVASUREN[3], ***M. DASHTSEREN**[1];
[1]Mongolian Natl. Univ. of Med. Sci., Ulaanbaatar, Mongolia; [2]Dept. of Psychosomatic Medicine,, Fukuoka Natl. Hospital, Natl. Hosp. Organization, Fukuoka, Japan; [3]Intl. Univ. of Hlth. and Welfare Narita Hosp., Narita, Japan

Abstract: Mental disorders cause the high burden of diseases associated with disability in developing countries. There has been no study on the prevalence of mental disorders in the general population of Mongolia since 2005. Therefore, this study aimed to determine the prevalence of mental distress and its risk factors in the Mongolian population. The study was conducted between July and October 2020, as a part of a nationwide multicenter, interdisciplinary, prospective, population-based cohort study that investigates brain-related disorders in the general population. 646 participants from 64 sampling sites spread through Ulaanbaatar and rural regions were approached. To assess potential mental distress in the general population, we used a combination of a 42-item self-report questionnaire and a 13-item structured psychiatric interview. Vital function tests and a survey on the quality of life were administered to each participant. Among the participants (mean age=38.0±15.4; 57.6% women), 35.9% were evaluated as having mental distress. There was a difference in residence location and the brain overactivity scale (BOS) total score between distressed and normal people (p=0.011 and p=0.024, respectively). Multiple linear regression showed that an increased in the BOS total score was associated with those who were young, living in urban areas, and have high quality of life. These results suggest that the prevalence of mental distress is 35.9% among the general population in Mongolia. The use of the BOS questionnaire along with a psychiatric interview can provide an effective screening tool in low-resource settings.

Disclosures: T. Amartuvshin: None. **T. Hiramoto:** None. **B. Lkhagvasuren:** None. **M. Dashtseren:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.15

Topic: G.06. Anxiety Disorders

Support: NIH Intramural ZIA-MH002781
NIH Intramural ZIA-MH002782

Title: Can resting state network efficiency improve diagnostic prediction of a structured psychiatric assessment?

Authors: *C. ANTONACCI[1], A. ZUGMAN[1], G. RINGLEIN[2], D. PINE[1];
[1]Natl. Inst. of Mental Hlth., Bethesda, MD; [2]Section on Develop. and Affective Neurosci., NIH, NIMH, Rockville, MD

Abstract: The relationship between neuroimaging measures and clinical/behavioral phenotypes is an important avenue of research in psychiatry and neuroscience. However, recent evidence questions the usefulness of neuroimaging markers, as they may lack validity and reliability important for clinical relevance. In this study, we explored whether global efficiency of the amygdala - a region highly implicated in internalizing psychopathology - improves the diagnostic performance of the Development and Well-Being Assessment (DAWBA) in a pediatric anxiety cohort. The DAWBA is a package of online questionnaires, lay interviews, and rating techniques for assessing psychiatric diagnoses in children and adolescents. Twenty-four participants (14 with a diagnosed anxiety disorder and 10 healthy controls) completed the Kiddie-Schedule for Affective Disorders and Schizophrenia for School Aged Children (KSADS) with a trained clinician, after which they completed the DAWBA and a resting state fMRI scan. For each disorder, the DAWBA produces a band score indicative of an individual's risk for diagnosis; here, we used the highest band score across all anxiety disorders as a predictor. Global efficiency measures were calculated using the CONN Toolbox. We used logistic regression to analyze the relationship between the maximum DAWBA band score, amygdala global efficiency, and a clinician's diagnosis via the KSADS. We tested two models, one full model with amygdala efficiency and the maximum DAWBA band score as predictors, along with age and sex as covariates. For comparison, we also tested a clinical model using only the DAWBA band score as a predictor, along with age and sex as covariates. The full model with the amygdala and the DAWBA band score showed that amygdala efficiency was not significant in predicting a clinician diagnosis ($\beta = 0.87$, $p = .38$, 95% CI (-0.83, 3.27)). The DAWBA band score, however, was a significant predictor in both models. The full model presented a marginally higher AIC (22.66) compared with the clinical model without the efficiency predictor (AIC: 21.58). The DAWBA alone was a strong predictor of diagnosis ($\beta = 3.64$, $p = .03$, 95% CI (1.41, 8.59)). Despite the limited sample size, our results suggest that the DAWBA alone is a very sensitive, if not specific, predictor of a clinician's diagnosis while amygdala global efficiency does not produce any detectable improvement in diagnostic prediction. Aligned with

the extant literature, our results suggest that using neuroimaging markers as indicators of risk for anxiety disorders may be limited due to small effect sizes.

Disclosures: C. Antonacci: None. **A. Zugman:** None. **G. Ringlein:** None. **D. Pine:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.16

Topic: G.06. Anxiety Disorders

Support: SR/SATYAM/33/2016

Title: Mind body techniques-based amelioration of stress and cognition through neurogenic molecular mechanism

Authors: *K. SHARMA, A. ANAND, S. GOEL;
PGIMER, Post Grad. Inst. of Med. Educ. and Res., Chandigarh, India

Abstract: The developing society has increasing incidence of stress and anxiety-based disorders due to the current lifestyle and work-life balance. Stress and anxiety can lead to cognitive decline and influence adult neurogenesis. Mind-body techniques like Yoga have been known to reduce stress and related complications. Through this exploratory study, we wanted to understand the molecular mechanism through which Mind-Body technique-based yoga protocol helps in amelioration of lifestyle stress-based cognitive decline, we recruited 2 groups: 60 healthy adults between the age of 18-55 years who had a sedentary lifestyle, and they were given 45 minutes of Yoga intervention for 3 months for 5 days/week, the control group comprised of 50 healthy adults (no intervention was given). Assessment of stress and anxiety was done through STAI questionnaire, and their general health and cognitive functioning were also measured using neuropsychological scales, i.e., General Health Questionnaire (GHQ), Digit Span test (DST), Trail Making Test (TMT A & TMT B) and Digit Symbol Substitution Test (DSST) before and after the intervention. To understand the molecular influence of the inclusion of Yoga based lifestyle intervention on the molecular and biochemical parameters, blood samples of the participants were taken at baseline, after 1 month, and after 3 months of the intervention, and assessment of serum levels of BDNF, VEGF, Angiogenin, and Amyloid-beta was done by ELISA, Anti-apoptotic markers (BCl 2 and Bcl xL) were also measured by qRT PCR. Also, the Lipid profile of the participants was measured. The results show that after 3 months, the yoga group showed a significant decline in stress and anxiety, showed an overall improvement in general health, showed an increase in HDL (the good cholesterol), BDNF, VEGF, Angiogenin, and Amyloid-beta showed a pattern of change after 1 and 3 months of yoga practice. Also, anti-apoptotic markers i.e., BCl 2 and B Cl xL showed an increase after yoga practice for 3 months. These results ascribe that the inclusion of yoga practice in daily lifestyle can help maintain a

stress-free life with improved cognition, and the effects observed are through the molecular effects on the angiogenesis and neurogenesis pathways.

Disclosures: **K. Sharma:** None. **A. Anand:** None. **S. Goel:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.17

Topic: G.06. Anxiety Disorders

Title: Medial and lateral orbitofrontal cortex volume in the course of behavior therapy in unmedicated patients with OCD

Authors: *B. ZUROWSKI[1], A. RUBART[1], K. WAHL[2], A. WAHL-KORDON[3], T. FREYER[4]; [1]Dept. of Psychiatry, Univ. of Luebeck, Luebeck, Germany; [2]Dept. of Psychology, Univ. of Basel, Basel, Switzerland; [3]Oberberg Kliniken, Hornberg, Germany; [4]Oberberg Kliniken, Schlangenbad, Germany

Abstract: Structural abnormalities in the (orbito)fronto-striato-thalamo-(orbito)frontal loops in patients with obsessive-compulsive disorder (OCD) are among the most reliable systems neuroscience findings in psychiatric disorders. The orbitofrontal cortex (OFC) has been determined as a crucial cortical structure both, for evaluation of positive and negative outcomes of actions in healthy subjects and for the pathophysiology of OCD (1). The aim of our study was to determine, whether and which volumetric abnormalities persist or change, respectively, in the course of cognitive behavior therapy (CBT) of OCD. Voxel-Based Morphometry (VBM) allows for detection of gray matter (GM) changes reflecting neuroplasticity as early as after one week of training. Thirty-five unmedicated inpatients were scanned twice, at admission and after 3 months of structured CBT. Accordingly, we scanned healthy matched control subjects twice, using a Siemens Trio 3-Tesla scanner. T1-images were analyzed with SPM12 software using customized procedures for preprocessing of longitudinal VBM data (http://dbm.neuro.uni-jena.de/vbm). Symptom severity scores and neuropsychological profiles were obtained from patients at both time points of scanning. We analyzed GM volume for effects of group, time, and their interaction, respectively. In comparison to healthy controls, we found an enlargement of *lateral* prefrontal and *lateral* orbitofrontal cortex in OCD patients. These differences remained unchanged despite a highly significant reduction of OCD symptoms after CBT. In contrast, *medial* OFC/orbital gyrus and amygdala were enlarged in patiens after therapy as compared to the first measurement at admission. Other regions showed time-independent differences between group. Left anterior and posterior insula/operculum GM volume was *increased* in patients. Patients showed a persistingly reduced GM volume of the substantia nigra/nucleus subthalamicus. The observed enlargement of lateral prefrontal and orbitofrontal cortex in patients with OCD is consistent with functional neuroimaging findings of over-recruitment of these regions in rest and during cognitive demands. The lateral OFC seems preferably involved in

processing information about (potential) punishment, whereas medial orbitofrontal cortex activation is rather important for the evaluation of (potential) reward in healthy subjects. The present finding of medial OFC and amygdala GM volume increase in the course of CBT is both, consistent with the medial-lateral functional distinction hypothesis and clinically plausible given 'reward deprivation' in OCD.

Disclosures: B. Zurowski: None. **A. Rubart:** None. **K. Wahl:** None. **A. Wahl-Kordon:** None. **T. Freyer:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.18

Topic: G.06. Anxiety Disorders

Title: Evaluation of the effect of musical stimulation with alpha, beta and theta binaural pulses under stress, anxiety and concentration conditions.

Authors: *O. ROMERO PORTALES[1,2], C. SOLORIO ALVARADO[3], C. ALBA BETANCOURT[4], J. CONTI GONZÁLEZ[5];
[1]Univ. Virtual del Estado de Guanajuato, Guanajuato, Mexico; [2]Pharm., UNIVERSIDAD DE GUANAJUATO, Guanajuato, Mexico; [3]CHEMISTRY, [4]PHARMACY, [5]BIOLOGY, UNIVERSIDAD DE GUANAJUATO, GUANAJUATO, Mexico

Abstract: SFN 2022 Binaural pulsesEvaluation of the effect of musical stimuli with alpha, beta and theta binaural pulses under stress, anxiety and concentration conditions(Romero, O., Betancourt, C., Conti, J.)
Music is considered one of the elements that cause pleasure in life; it improves health through neurochemical systems for reward, motivation and pleasure. Binaural pulses (BB) - stimulation of sensory information - provide potential information of altered consciousness to modify the states of excitement, attention and level of consciousness. They can be classified in theta (4 to 7.9 Hz), alpha (8 to 13.9 Hz) and beta (14 to 30 Hz) waves. Theta waves are often associated with information consolidation processes or long-term memory tasks, alpha with cognitive improvement and beta with executive functioning.The release of dopamine induced by listening to music, provides a sense of calm; however, the musical genre can alter the results between different individuals because the stimuli that provide pleasure is different for each person.During the SARS-COV-2 pandemic, distance education was implemented, so university students used different concentration strategies to perform the tasks assigned by their teachers. In this work we evaluated the effect of BB generated by a compilation of songs that emit BB alpha, beta and theta, and how stress and concentration were modified when performing tasks that require logical-mathematical skills.A total of 200 participants were enrolled in the study. We found that University students got better results while listening to BB theta including on reggaeton and regional mexican band, compared to the exam resolved while listening to BB alpha and beta.

Since BB alpha/beta are related to cognitive improvement and executive functioning, it is quite surprising that students got more concentrated with BB theta than alpha or betha. This case could be explained because of the personal music taste that prevail among students, in which BB theta music predominates, even mentioning the adaptation of participants with BB theta on a regular basis.

Disclosures: O. Romero Portales: None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.19

Topic: G.06. Anxiety Disorders

Support: NIH Grant R15MH110951

Title: Frontal and striatal volumes at baseline predict changes in anxiety symptomology following attention bias modification

Authors: *S. LYONS, J. M. CARLSON, L. FANG;
Psychological Sci., Northern Michigan Univ., Marquette, MI

Abstract: Attentional resources are allocated to emotional stimuli within the environment. An attentional preference for one type of valenced stimuli is defined as an attentional bias. It is thought that attentional biases toward negative stimuli underlie cognitive maintenance and vulnerability to affective disorders such as anxiety. Attentional biases have been linked to frontal and subcortical areas thought to mediate affective regulation and processing, such as the lateral and superior prefrontal cortex, anterior cingulate cortex (ACC), and the amygdala. In the early 2000s, attention bias modification (ABM) was developed as a potential treatment for targeting this type of cognitive bias. However, ABM does not appear effective for everyone, underlying the importance for improving the understanding of which factors predict treatment success. The current investigation was aimed to discover which brain regions at baseline predict changes in anxiety symptoms following an ABM intervention. Thirty-one participants (N_{Female} = 22; M_{Age} = 21.5) who scored 40+ on the State-Trait Anxiety Inventory (STAI; M = 52.29, SD = 6.59) and had attentional bias scores greater than 7 ms completed attentional training away from negative images. Prior to receiving training, structural T1-weighted scans were obtained using a 1.5 T MRI. Six-weeks of ABM were administered using a mobile application installed on participants' phones. The STAI was administered again following ABM training. Trait subscores of the inventory were used to assess the relationship between baseline gray matter volumes and intervention-related changes in trait anxiety. Structural MRI images were processed using CAT12 (version 12.8). A voxel-based morphometry (VBM) regression model including changes in trait anxiety as a covariate and total intracranial volume as a nuisance variable was created in SPM12. Pre-session trait anxiety was marginally higher than the post-session (M = 51.68, SD =

8.23). We observed that lower levels of baseline gray matter volume in a large cluster encompassing the subgenual ACC and the ventral striatum (VS) significantly predicted decreases in trait anxiety ($t(28) = 4.76$, $p_{FWE} = 0.003$, $k = 690$). A similar relationship was observed in another cluster including the posterior cingulate cortex (PCC), but did not reach statistical significance when controlling for multiple-comparisons ($t(28) = 4.62$, $p_{uncorr} = 0.021$, $k = 174$). Results from this investigation suggest that the gray matter volume of brain regions involved in affective control and processing are important when determining who may most benefit from ABM.

Disclosures: S. Lyons: None. **J.M. Carlson:** None. **L. Fang:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.20

Topic: G.06. Anxiety Disorders

Title: Neuroanatomical Correlates of Climate Change Anxiety

Authors: *J. FOLEY[1], L. FANG[2], J. M. CARLSON[2];
[2]Psychological Sci., [1]Northern Michigan Univ., Marquette, MI

Abstract: Previous structural MRI studies have demonstrated that people who exhibit high levels of trait anxiety as well as those with an anxiety disorder tend to display lower gray matter volume(compared to healthy controls) in regions of the frontal cortex. Beyond traditional sources ofanxiety, climate change is an important global threat that can lead to elevated levels of anxiety. Indeed, studies have shown that people who care about the impacts of climate change tend toexperience heightened anxiety related to the phenomenon. Accordingly, a newly developingconstruct of climate change anxiety has emerged in the last few years, but little is known aboutits cognitive and neural correlates. Here, in an initial investigation, we assessed voxel-wise differences in gray matter volume across the brain in relation to climate change anxiety.Twenty-one participants (16 females, age: $M = 20.4$ years, $SD = 2.16$, range = 18 - 28 years) completed the climate change anxiety questionnaire ($M = 23.6$,$SD = 9.51$, range = 13 - 43), thenthey later received structural T1-weighted MRIs using a 1.5 T scanner within a week of completing the questionnaire. Using the CAT 12 toolbox in SPM 12 within the MatLab suite, a voxel-based morphometry linear regression was run using climate change anxiety scores as well as total intracranial volume as covariates. The analysis showed that lower gray matter volumewas associated with higher climate change anxiety in clusters located within the dorsolateral prefrontal cortex ($t = 4.50$, $p < 0.001$, $k = 113$) and posterior parietal cortex ($t = 4.95$, $p < 0.001$, $k = 205$). Results from this analysis add insight intothe neuroscience of climate change anxiety. Inparticular, our findings suggest that anxiety related to climate change is also linked to morphological differences in the frontal lobe, and posterior parietal cortex, as observed in research into general/high trait anxiety. The dorsolateral prefrontal cortex is associated with

attention guidance and emotion regulation, which can be affected in anxiety. Lower grey matter volume in these areas may decrease the ability to utilize regulatory cognitive control processesthat in turn may contribute to symptoms of anxiety. Future research will be needed to determine the exact functional significance of these gray matter volume differences; yet, our results provide initial evidence that climate change anxiety is linked to gray matter volume differences in regions implicated in cognitive control.

Disclosures: **J. Foley:** None. **L. Fang:** None. **J.M. Carlson:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.21

Topic: G.06. Anxiety Disorders

Support: NIH Grant R15MH110951

Title: Reduced intrinsic connectivity between the dorsal anterior cingulate and medial prefrontal cortices predicts anxiolytic benefits of attention bias modification training in high trait anxious individuals

Authors: L. GENTRY[1], S. LYONS[2], B. KWAK[1], D. KASSEL[1], B. DAVIS[1], L. FANG[2], *J. CARLSON[1];
[2]Psychological Sci., [1]Northern Michigan Univ., Marquette, MI

Abstract: Attention bias modification (ABM) is an intervention aimed at reducing anxiety by training individuals with an attentional bias toward threatening stimuli to redirect their attention toward non-threatening stimuli. Previous research found changes in amygdala, anterior cingulate cortex (ACC), and lateral prefrontal cortex (PFC) activity following ABM with effectiveness increasing with neuromodulation of the dorsolateral PFC. Although ABM has been found to be effective for some individuals, it is not effective for everyone. Identifying predictors of ABM outcomes could help determine who is most likely to benefit from the training. This investigation aimed to identify differences in intrinsic functional connectivity that predicted successful reduction in anxiety following ABM. Participants were initially screened using the state-trait anxiety inventory trait scale (STAI-T) for scores greater than 40 to be included in the study. 50 participants were assigned to complete 36 ABM training sessions across six weeks where attention was trained away from negative stimuli (including fearful faces and emotionally negative words). Resting-state functional MRIs were obtained before and after ABM training using a 1.5 T scanner. Participants who did not complete ABM training or had excessive motion during the MRI scan were excluded, leaving $N = 21$ participants ($N_{Female} = 15$) in the final analyses. In the post-session, the STAI-T was completed to assess changes in trait anxiety as a result of the ABM training. ROI-to-ROI resting-state functional connectivity analyses were performed utilizing seeds from the salience, frontoparietal, and default mode networks involved

in anxiety-relevant processes including affective, cognitive control, and self-referential processing, respectively. Post-training STAI-T scores were descriptively lower (M = 52.05, SD = 8.04) compared to before training (M = 53.43, SD = 6.34). We observed that decreased functional connectivity between the dorsal ACC (salience network) and medial PFC (default mode network) at baseline predicted greater improvements in anxiety following ABM training ($t(17) = -3.59$, $p_{FDR} = 0.03$). This finding suggests that weaker functional connectivity between networks involved in salience detection (ACC) and self-referential processing (mPFC) at baseline predict greater symptom change following ABM. Both the ACC and mPFC are involved in emotion appraisal and regulation. Weaker connectivity between these structures at baseline could indicate poorer ability to process and regulate negative emotion, thus leading individuals to experience greater changes over the course of ABM training.

Disclosures: L. Gentry: None. **S. Lyons:** None. **B. Kwak:** None. **D. Kassel:** None. **B. Davis:** None. **L. Fang:** None. **J. Carlson:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.22

Topic: H.04. Executive Functions

Support: NIH-NINDS NS111019

Title: Real-time regulation of arousal and performance in healthy non-human primates using the DyNeuMo2 closed-loop DBS device

Authors: *J. L. BAKER[1], R. TOTH[2], A. DELI[2], M. ZAMORA[2], J. E. FLEMING[2], M. BENJABER[2], J.-W. RYOU[1], K. P. PURPURA[1], N. D. SCHIFF[1], T. DENISON[2];
[1]Weill Cornell Med., Cornell University: Weill Cornell Med. Col., New York, NY; [2]Univ. of Oxford, Oxford, United Kingdom

Abstract: Application of closed-loop approaches in systems neuroscience and deep brain stimulation (DBS) holds great promise for revolutionizing our understanding of the brain and for developing novel neuromodulation strategies to restore lost function. The mammalian central thalamus (CT) is hypothesized to underlie *arousal regulation of the cortex*, a mechanism critical to supporting performance during wakefulness. Dysfunction of arousal regulation is hypothesized to contribute to the persistent deficits in executive attention, working memory, and chronic mental fatigue experienced in a variety of neurological disorders, most prominently in patients following traumatic brain injury (TBI). Previously, we showed that central thalamic DBS (CT-DBS) produced robust improvement in arousal and recovery of cognitive and motor function in a very severe TBI patient. In a recently completed feasibility study (NS095554), we demonstrated in five moderate to severe TBI (msTBI) patients that CT-DBS can restore executive attention and working memory, and in three patients, reduced mental fatigue. Despite

these promising results, our current mechanistic understanding of arousal regulation is limited by our inability to chronically record, analyze, and modulate, *in real-time, arousal regulation of the cortex* in behaving animals. Here we present data collected from two healthy non-human primates (NHP), *Macaca mulatta*, as they performed visuomotor reaction time tasks, while using the Picostim-DyNeuMo2 (Oxford and Bioinduction, Ltd. Bristol, UK) bi-directional, closed-loop research platform in order to modulate, *in real-time*, arousal regulation and performance. Chronic epidural ECoG signals collected over medial frontal areas were used to develop subject-specific algorithms to identify periods of low arousal and alertness. The NHPs' task performance and drowsiness, the latter quantified using eye blink duration and partial-to-full eye closures, were measured. The DyNeuMo2 embedded classifier detected the presence of low arousal/alert states and initiated stimulation CT-DBS until the ECoG signals return to an 'alert' state. Preliminary results from CL-DBS experiments show correlation above 0.7 between loss of alertness (eyes closed) and stimulation state changes; optimization of the stimulation algorithm is ongoing. These results represent proof-of-concept and a first step in our goal to design and test novel closed-loop devices that are capable of monitoring the circadian dynamics of arousal regulation in TBI patients, in order to effectively restore arousal regulation and thereby improve executive attention, working memory and reduce mental fatigue.

Disclosures: **J.L. Baker:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); receipt of intellectual property rights/patent holder. **R. Toth:** None. **A. Deli:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); receipt of intellectual property rights/patent holder. **M. Zamora:** None. **J.E. Fleming:** None. **M. Benjaber:** None. **J. Ryou:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); receipt of intellectual property rights/patent holder. **K.P. Purpura:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); receipt of intellectual property rights/patent holder. **N.D. Schiff:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); receipt of intellectual property rights/patent holder. **T. Denison:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock, stock options, royalty, receipt of intellectual property rights/patent holder.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.23

Topic: F.04. Neuroimmunology

Support: UCSD ACTRI Pilot Award
Cota-Robles Fellowship
National Science Foundation GRFP

Title: Longitudinal assessment of circulating anti-nmdar1 autoantibodies in service members before and after a combat deployment

Authors: *M. N. VAUGHN[1], V. B. RISBROUGH[1,2], X. ZHOU[1,3];
[1]Psychiatry, Univ. of California San Diego, La Jolla, CA; [2]Ctr. of Excellence for Stress and Mental Hlth., [3]Mental Illness Res. Educ. and Clin. Ctr., San Diego Veteran's Affairs, La Jolla, CA

Abstract: Amongst the general population, an estimated 5-10% contain low levels of anti-NMDAR1 autoantibodies in their peripheral blood stream. High titers of anti-NMDAR1 autoantibodies in the brain can cause anti-NMDAR1 encephalitis that displays psychiatric and neurological symptoms. We have previously shown that mice with chronic low titers of anti-NMDAR1 autoantibodies exhibit impairments in working memory, fear extinction learning and recall. NMDAR signaling in prefrontal cortex plays a key role in fear extinction and post-traumatic stress disorder (PTSD) development; and circuits underlying fear learning, extinction and recall are highly conserved across rodents and humans. Therefore, we hypothesized that anti-NMDAR1 autoantibodies in humans may also disrupt these fear processes and thereby confer increased risk for PTSD. In this study, we aim to investigate the prevalence and consistency of anti-NMDAR1 autoantibodies in a high-risk human population, and explore associations between autoantibody levels and multiple cognitive measures. We leveraged 143 banked blood samples and symptom data from a prospective longitudinal study of PTSD development in Active Duty Marines, which provides us with blood plasma samples collected before and after a 7 month combat deployment to Afghanistan. Subjects were age-, gender-, trauma exposure (Life Events Checklist “witnessed” or “happened to me” total scores) and ethnicity-matched. We utilized our previously developed novel, highly sensitive One-Step immunoassay to semi-quantify the staining intensities of 286 samples. We find that NMDAR1 autoantibodies are present in 16.7% of all participants, with 9.8% containing autoantibodies both before and after their one-year deployment. ~5% of participants developed autoantibodies during their deployment period, and 2% displayed a cessation of autoantibody production. Participants were also assessed for PTSD symptom severity at both time points, as well as fear extinction performance. Thus far, our exploratory analysis suggests associations between NMDAR1 autoantibodies and memory performance. Together, our findings suggest that circulating NMDAR1 autoantibodies in humans are persistent, and have the potential to influence cognitive deficits in trauma-exposed populations.

Disclosures: M.N. Vaughn: None. **V.B. Risbrough:** F. Consulting Fees (e.g., advisory boards); Jazz Pharmaceuticals, Engrail Therapeutics, Farallon Capital. **X. Zhou:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.24

Topic: F.04. Neuroimmunology

Support: Herman Dana Foundation

Title: Long term neuroendocrine immune alterations accompanying chronic Post-Traumatic Stress Disorder following exposure to suicide bomb terror incidents during childhood.

Authors: *R. SEGMAN[1,2], T. GOLTSER[1,2], A. SHALEV[2], F. BENARROCH[2], D. PEVZNER[2,1], L. CANETTI[1], C. SALONER[1], E. GALILI-WEISSTUB[2];
[1]Mol. Psychiatry Lab. - Dept. of Psychiatry, Hadassah - Hebrew Univ. Med. center, Jerusalem, Israel; [2]The Herman-Danna Div. of Pediatric Psychiatry, Dept. of Psychiatry, Hadassah - Hebrew Univ. Med. Center; Jerusalem Israel, Jerusalem, Israel

Abstract: Background and Aim: Long term neuroendocrine immune alterations have been proposedto play a mechanistic role in Post-Traumatic Stress Disorder (PTSD) as well as in itsassociated increase in physical morbidity and mortality. Better characterization of alteredimmune function may help identify diagnostic and prognostic biomarkers and potentiallytargets for preventive intervention.**Methods:** As part of a prospective cohort ongoing study, we conducted a preliminary casecontrol comparison of resting mononuclear cell profile between terror victims treated inchildhood at the Hadassah Medical Center emergency department over the previousdecade, who developed chronic PTSD upon long term follow up or recovered, and healthynon traumatized controls.**Results:** Our results point to childhood trauma related childhood long term alterations in theexpression of genes related to cortisol reactivity, including the glucocorticoid receptor aswell as its trans-activator Spindle And Kinetochore Associated Complex Subunit 2 (SKA2)among resting mononuclear cells in young adults.Conclusion: Chronic resetting of the neuroendocrine reactivity of immune cells mayparticipate in inflammatory activation, as well as modulate immune - neuro-endocrinefeedback.Better characterization and understanding of these findings may point todiagnostic and prognostic biomarkers and potentially elucidate mechanistic involvement ofimmune activation in childhood trauma related chronic PTSD.

Disclosures: R. Segman: None. **T. Goltser:** None. **A. Shalev:** None. **F. Benarroch:** None. **D. Pevzner:** None. **L. Canetti:** None. **C. Saloner:** None. **E. Galili-Weisstub:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.25

Topic: F.03. Stress and the Brain

Support: Natural Sciences and Engineering Research Council of Canada (NSERC)

Title: Trait anxiety predicts visual N1 ERP amplitude of visual distractors in the presence of tactile stimuli

Authors: *M. V. FAERMAN, K. A. EHGOETZ MARTENS, W. R. STAINES;
Kinesiology & Hlth. Sci., Univ. of Waterloo, Waterloo, ON, Canada

Abstract: Evidence suggests that individual differences in trait anxiety affect cognition, but its effect on sensorimotor function remains unknown. Sensorimotor function involves goal-directed attentional control to influence perception and generate action. Attentional Control Theory (ACT) postulates that high trait anxiety impairs attentional processing efficiency but does not affect behavioural performance. This study sought to assess the relationship between trait anxiety and sensorimotor function by examining early cortical event-related potentials (ERPs) in response to visual-tactile stimuli and behavioural performance cost incurred by crossmodal distractors. Trait anxiety was assessed using the State-Trait Anxiety Inventory trait scale (STAI-T). Electroencephalography was used to acquire early tactile (P50, N70, P100, N140) and visual (P1, N1, P2) ERP peak amplitudes from 27 healthy young adult participants (aged 18-33) during the task. Stimuli were presented as either visual or tactile alone or bimodally for each block. Visual stimuli were vertical bars of varying heights and tactile stimuli were vibrations to the left index finger. Participants responded by gripping a rubber bulb with force corresponding to stimulus amplitudes. Instructions were given to attend and respond to only either visual or tactile stimuli (alone or with a distractor) in each block. Based on ACT, it was hypothesized that trait anxiety would show reduced distractor suppression as shown by a positive relationship with early ERP amplitudes in response to distracting stimuli, but no relationship to behavioural distractor cost. Regression analysis revealed that trait anxiety showed a positive relationship with the reduction of visual distractor N1 ERP magnitudes when presented simultaneously with tactile stimuli ($p=0.003$). Tactile and visual behavioural distractor costs showed no relationship to trait anxiety ($p>0.05$). Evidence suggests that the magnitude of the visual N1 ERP corresponds to selective attention. These results indicate increased attentional suppression of visual distractors when presented with tactile stimuli. The behavioural results match ACT, as trait anxiety did not impact performance. In contrast to our hypothesis, higher trait anxiety was associated with more prominent attentional suppression of visual distractors with tactile stimuli. These findings show that high trait anxiety benefits early crossmodal selective attention processes. Future work should explore this concept in contexts with higher perceptual load and in different sensory modalities.

Disclosures: M.V. Faerman: None. **K.A. Ehgoetz Martens:** None. **W.R. Staines:** None.

Poster

559. Stress, Trauma, and Anxiety Related Disorders: Human Studies

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 559.26

Topic: H.02. Perception and Imagery

Title: Prevalence of stress, anxiety, depression during the pandemic in individuals with Multiple sclerosis - A Rasch analysis

Authors: *B. BALAKRISHNAN[1], M. GANESAN[1], V. KRISHNAN MUTHAIAH[2], H. GARG[3];
[1]Univ. of St. Augustine for Hlth. Sci., San Marcos, CA; [2]Univ. at Buffalo, New York, NJ; [3]Rocky Mountain Univ. of Hlth. Professions, Provo, UT

Abstract: The impact of the COVID 19 pandemic on mental health issues such as stress, anxiety, and depression are a growing concern in the United States and worldwide. A preexisting neurological condition may heighten the risk for mental health disorders during COVID 19. Hence, it is vital that the health care system be informed about the effect of COVID 19 on neurological conditions such as Multiple sclerosis (MS) to prioritize and integrate mental health concerns within the treatment plan. To address this issue, the current study aimed to check the prevalence of mental health issues using DASS21 and to check the construct validity of the DASS21 in individuals with MS during the pandemic. Further to explore the prevalence of stress, anxiety, and depression in individuals with MS after removing the participants with an extreme response. A survey monkey link consisting of demographics, and a DASS-21 questionnaire was sent to three centers including Bennett Rehabilitation Institute NY, Brain Center at Miami, FL, and the MS Physical Therapy and wellness center at Rocky Mountain University of Health Professions at Provo, Utah. We have obtained responses from 24 individuals with MS using 21 items DASS questionnaire during a pandemic. The DASS-21 consists of 21 questions which contain 3 subscales and each subscale with 7 questions named stress, anxiety, and depression; each subscale contains 7 questions. Participants rated a scale of 0-3 for each question. The rating scale “0” denotes does not apply to me at all, “1” applied to me to some degree or some of the time, “2” applied to me to a considerate degree or a good part of the time, and “3” applied to me very much or most of the time. The raw score is multiplied by 2 for each subcomponent. For each component, the score can range from 0 to 42. Rasch model was used to analyze construct validity and extreme responses of DASS 21 items and 24 individuals with MS. Extreme responses from four individuals with MS were identified using outfit statistics. After removing the extreme responses, we found that the person reliability improved to 88% with a separate index of 2.69. After removing the outfit, we found the prevalence of stress was 30%, anxiety was 20% and depression was 45%. The application of the psychometric Rasch model helps for the meaningful interpretation of determining the target attributes in multiple sclerosis. We report that individuals with MS exhibit a high prevalence of mental health issues during the pandemic

Disclosures: B. Balakrishnan: None. **M. Ganesan:** None. **V. Krishnan Muthaiah:** None. **H. Garg:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.01

Topic: G.07. Post-Traumatic Stress Disorder

Support: Aptinyx, Inc.
NIH Grant R01MH113007

Title: Positive allosteric modulator of the NMDA receptor, NYX-783, reverses repeated stress-induced reduction of exploration and reduces fear to unpredictable threats in rats

Authors: *J. DABROWSKA[1], J. ROSENKRANZ[2], R. CHUDOBA[3], S. OLSON[4], C. STICKLING[4];
[1]Chicago Med. Sch. RFUMS, North Chicago, IL; [2]Cell. and Mol. Pharmacol., Rosalind Franklin Univ. of Med. and Sci. Chicago Med. Sch., Evanston, IL; [3]Rosalind Franklin Univ. of Med. and Scien, North Chicago, IL; [4]RFUMS, North Chicago, IL

Abstract: Posttraumatic stress disorder (PTSD) is a trauma- and stressor-related disorder. Hallmarks include hyperarousal, avoidance behavior, and high fear reactivity to unpredictable threats. Although current pharmacotherapy for PTSD is limited, modulators of glutamatergic NMDA receptors (NMDAR) have received substantial attention. A recent phase II clinical trial (ClinicalTrials.gov: NCT04044664) demonstrated the efficacy of NYX-783 (Aptinyx, Inc., Evanston, IL), a positive allosteric modulator of NMDAR, in subjects with PTSD. Here, we investigated the effects of NYX-783 in rat models of the sensitization of acoustic startle reflex (SS), fear reactivity to unpredictable threats (anxiety-potentiated startle, APS), and repeated stress-induced reduction in exploration in the elevated plus-maze (EPM). Following baseline startle testing, male rats (n=12/group) were tested for foot-shock induced SS one hour after saline or NYX-783 (1 or 10 mg/kg i.p.). Despite significant foot-shock induced SS in all rats ($F(1,33) = 35.42$, $P<0.0001$), there was no effect of NYX-783 on SS ($F(2,33)=0.9954$, $P=0.3804$, two-way ANOVA). To probe fear to unpredictable threats, we used APS, in which startle is potentiated by un-paired cues and foot-shocks following fear conditioning. Female rats received saline or NYX-783 (1, 10, 30, or 100 mg/kg, n=10/group) before fear recall. All rats demonstrated significant APS during fear recall ($F(1.471, 51.47) = 13.10$, $P=0.0001$) with no treatment effect ($F(3,35)=1.670$, $P=0.1913$). However, one-way ANOVA of percentage change of non-cued fear showed a significant treatment effect ($F(3,35) = 3.355$, $P=0.0297$), with a trend between saline and 30 mg/kg of NYX-783 ($P=0.0710$). Finally, there was a significant difference between saline and 30 mg/kg of NYX-783 ($t=2.469$, $df=17$, $P=0.0244$, unpaired *t*-test). To measure effects of repeated stress on EPM, male rats (n=12/group) underwent daily resident-intruder stress for 5 consecutive days, or control handling. On the testing day, rats received saline or NYX-783 (1,10, or 30 mg/kg) one hour prior to testing on the EPM. There was no effect of NYX-783 in the control group. After stress there was a dose-dependent increase in time spent in the open arm ($P<0.05$, unpaired *t*-test at 30 mg/kg, $t=2.5$, $df=22$), and distance traveled in the open arm ($P<0.05$, unpaired t-test at 30 mg/kg, $t=3.5$, $df=22$), but no significant effect on total distance traveled in the EPM ($P>0.05$, unpaired *t*-test, $t=0.52$, $df=22$). Overall, our results show that NYX-783 at 30 mg/kg reverses repeated stress-induced reduction of exploration, acutely reduces fear to unpredictable threats, and demonstrates pharmacotherapeutic potential for PTSD.

Disclosures: J. Dabrowska: None. **J. Rosenkranz:** None. **R. Chudoba:** None. **S. Olson:** None. **C. Stickling:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.02

Topic: G.07. Post-Traumatic Stress Disorder

Support: T32 AA007456
AA027700
AA017447
AA029841
AA021491
AA006420

Title: Chemogenetic inhibition of central amygdala CRF neurons reduces alcohol drinking and trauma-related behaviors in a model of post-traumatic stress and alcohol use disorder

Authors: *B. CRUZ[1], A. E. SNYDER[1], V. VOZELLA[3], P. J. GANDHI[1], P. C. BIANCHI[1], D. KIRSON[4], R. O. MESSING[5], M. ROBERTO[2];
[1]The Scripps Res. Inst., San Diego, CA; [2]Dept. of Mol. Med., The Scripps Res. Inst., LA Jolla, CA; [3]Mol. Med., Scripps Res. Inst., LA JOLLA, CA; [4]Dept. of Pharmacology, Addiction Science, and Toxicology, Memphis, TN; [5]Neurosci., The Univ. of Texas at Austin, Austin, TX

Abstract: The central nucleus of the amygdala (CeA) plays an important role in mediating stress-induced excessive alcohol drinking. However, little is known about the neurocircuitry underlying enhanced alcohol drinking produced by post-traumatic events. We used a chemogenetic approach to examine the role of CeA neurons that produce the stress peptide, corticotropin-releasing factor (CRF). These neurons regulate anxiety-like behavior, traumatic fear responses, and alcohol drinking. Adult male and female Crh-Cre rats (Wistar background) that express Cre recombinase under control of the Crh promotor, received foot shock stress on two occasions separated by 48 hrs. This experience produces inhibitory avoidance. Subsequently, all rats underwent two-bottle choice (2BC; 20% v/v; 2hrs) voluntary alcohol drinking for four weeks. One week-later, fear overgeneralization and irritability-like behaviors were measured during alcohol abstinence. Chemogenetic inhibition of CeA CRF neurons generally reduced alcohol intake. Inhibition of CeA CRF neurons did not alter fear overgeneralization. In females, inhibition of CeA CRF neurons increased defensive signs measured by the bottle brush irritability test. Collectively, our findings reveal that CeA CRF neurons mediate some comorbid signs of post-traumatic stress including vulnerability to increased alcohol drinking, and in females, irritability-like symptoms. These findings suggest that CRF neuronal population in the CeA is a potential target for reducing phenotypic behaviors associated with comorbid post-traumatic stress and alcohol use disorder.

Disclosures: B. Cruz: None. **A.E. Snyder:** None. **V. Vozella:** None. **P.J. Gandhi:** None. **P.C. Bianchi:** None. **D. Kirson:** None. **R.O. Messing:** None. **M. Roberto:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.03

Topic: G.07. Post-Traumatic Stress Disorder

Title: L-acetylcarnitine supplementation reduces ptsd symptom severity and promotes hepatic fatty acid oxidation which drives ptsd resilience in male mice

Authors: *G. PRESTON, T. KOZICZ;
Clin. Genomics, Mayo Clin., Rochester, MN

Abstract: Mitochondrial dysfunction has been strongly implicated in several psychopathologies, including depression, anxiety, schizophrenia, and posttraumatic stress disorder. We recently demonstrated a profound multisystem metabolic reprogramming in mice associated with trauma exposure and PTSD-like behavior. Most significantly we observed evidence reduced hepatic fatty acid oxidation (FAO) flux following trauma exposure, and reduced plasma acetylcarnitine in mice displaying PTSD-like behavior. We therefore hypothesized that supplementation with L-acetylcarnitine (LAC) would promote hepatic fatty acid oxidation, ameliorate PTSD-like symptomatology and promote resilience to PTSD-like behavior in trauma-exposed mice. 48 male C57Black/6J mice aged 10 weeks were exposed two decontextualized sessions of inescapable electric foot shock over two consecutive days. Subsequently half of the cages received either 0.6% LAC in drinking water or vehicle. Mice were assessed for PTSD-like behaviors - anxiety, risk taking behavior, hyperarousal, and sleep disruption - and PTSD-like and resilient animals were identified. Each animal also performed a restrained stress test to assay corticosterone response. Finally, each animal was sacrificed, and liver and plasma were collected and flash frozen. ELISA was performed to measure restraint stress plasma corticosterone response. Targeted metabolomics for acylcarnitines was performed on liver and plasma. RT-qPCR was performed on liver cDNA to assay expression changes in genes involved with FAO, and mitochondrial electron transport chain complex activities were assayed in liver tissues. LAC supplementation reduced the severity of PTSD-like behaviors, as well as the restraint stress corticosterone response. LAC supplementation increased the abundance of plasma short chain acylcarnitines, and hepatic free carnitine and medium chain acylcarnitines in the liver, which drove PTSD-resilience. LAC-supplemented resilient animals displayed significantly increased expression of the FAO genes Pgc1a, Cpt1a, and Cpt1b, and increased activity of hepatic mitochondrial electron transport chain complex II, which receives electrons from the reducing equivalent $FADH_2$, which is produced in large quantities through FAO. These data further implicate the role of multisystem energy metabolism in PTSD vulnerability and symptomatology and indicate that improving hepatic mitochondrial FAO flux and subsequent bioenergy metabolism through dietary supplementation can ameliorate PTSD-like symptomatology and drive PTSD-resilience.

Disclosures: G. Preston: None. **T. Kozicz:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.04

Topic: G.07. Post-Traumatic Stress Disorder

Support: GR-2019-12369216
ERA-NET NEURON Cofund JTC 2017 (Top-down PTSD)

Title: Behavioral and molecular characterization of PTSD-like susceptible and resilient lines of rats

Authors: *M. MORENA[1], G. F. MANCINI[1], E. BLASI[1], E. RICCARDI[1], A. PISANESCHI[1], O. C. MEIJER[2], P. CAMPOLONGO[1];
[1]Sapienza Univ. of Rome, Rome, Italy; [2]Leiden/Amsterdam Ctr. for Drug Res., Leiden, Netherlands

Abstract: After experiencing a traumatic event, stress coping mechanisms may become dysfunctional leading to the development of psychiatric disorders, such as post-traumatic stress disorder (PTSD). However, only a subset of subjects who experience trauma develops PTSD and factors conferring vulnerability to the neurobiological sequelae of trauma exposure remain largely unknown. Identifying mechanisms of susceptibility to develop PTSD has been challenging due to the lack of proper research tools, which is reflected by the scarcity of effective treatments for PTSD. We have previously developed a rat model for PTSD-like symptomatology that allows early post-trauma behavioral profiling to enable the differentiation between susceptible and resilient phenotypes to the development of long-lasting trauma-induced behavioral alterations. However, to date, the available PTSD-like susceptible (SUS) or resilient (RES) animal models are primarily obtained based on post-trauma behavioral profiling, thus they do not allow for examination of neurobiological determinants of vulnerability (or resilience) before trauma exposure. The aim of the present study was to generate 2 populations of rats SUS or RES to develop PTSD-like symptoms and perform a behavioral and molecular characterization (transcriptomic analyses of fear-related brain regions) to identify markers of vulnerability and resilience to develop PTSD-like symptomatology.By selective breeding of rats, screened based on post-trauma behavioral indicators of PTSD-like vulnerability (or resilience) (Colucci et al. 2020 Transl Psychiatry 10:243), we generated two distinct lines of PTSD-like SUS and RES rats. Data collected so far, on the 9th generations, indicated a marked distinction between the two phenotypes in terms of behavioral indices of traumatic memory (i.e., increased traumatic memory consolidation, and recall and reduced extinction in SUS vs RES) and post-trauma sociability (i.e., reduced sociability in SUS vs RES). Furthermore, profiling of anxiety- and depressive-like behavior, stress-reactivity and social interaction indicated overall marked baseline differences between the two phenotypes in both sexes, assessed in naïve rats never exposed to a traumatic experience.The availability of such SUS and RES lines of rats represents an unprecedented tool to determine mechanisms of PTSD vulnerability or resilience and identify new targets for prophylactic and/or therapeutic interventions for the treatment of trauma-related disorders.

Disclosures: **M. Morena:** None. **G.F. Mancini:** None. **E. Blasi:** None. **E. Riccardi:** None. **A. Pisaneschi:** None. **O.C. Meijer:** None. **P. Campolongo:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.05

Topic: G.07. Post-Traumatic Stress Disorder

Support: Bradley University O'Grady Fund

Title: Characterization of sex differences among adolescent rats with post-traumatic stress disorder

Authors: *A. MABIE, J. O'RUSSA, B. HUERTA, M. DILLERUD, T. E. KOELTZOW; Bradley Univ., Peoria, IL

Abstract: Sex differences associated with Post-traumatic Stress Disorder (PTSD) are commonly reported, but not well understood. Although women are diagnosed with PTSD at twice the rate of men, the vast majority of research is on male subjects (Kessler et al., 2017). Sex differences in onset, symptom expression, and treatment response highlight the need for a more thorough examination of animal models of PTSD.The present study compared the impact of the single-prolonged stress (SPS) model of PTSD in adolescent (postnatal day 25) male and female Sprague-Dawley rats. The SPS procedure consisted of two hours of forced restraint, twenty minutes of forced swimming, and a CO_2-induced loss of consciousness (Lisieski et al., 2018). The SHAM controls were exposed to the same contexts but without the stress-inducing stimuli. Immediately after SPS, rats were placed in automated locomotor activity chambers for twenty hours. After a 10 day incubation period, rats were subjected to a test battery: open field (10 minutes), elevated plus maze (EPM; 10 minutes), forced swim (10 minutes), and sucrose preference (72 hours). Six weeks later, rats were inescapably placed on an open arm of the EPM for five minutes. Rats were subsequently assessed on a second test battery. The sucrose preference test was expanded (48 hours with 2% and 72 hours with 1%). Upon completion of behavior testing, dopamine in prefrontal cortex and ventral striatum were measured (data in progress).Preliminary data analysis revealed robust sex differences across several measures. A Greenhouse-Geiser corrected RMANOVA revealed a statistically significant Time X Sex interaction (F=2.44; $p < 0.05$) as the female rats displayed hyperactive locomotor activity during the first hour of the automated locomotor activity chamber. A one-way ANOVA also revealed a statistically significant effect of stress (F= 13.31; $p < 0.001$) as the SPS-exposed rats showed increased locomotor activity during the second hour of the test. The open field test resulted in a statistically significant increase in center crosses (F= 27.68; $p < 0.001$), total crosses (F= 11.69; $p < 0.002$), and rearing (F= 36.37; $p < 0.00$) among female rats. The elevated plus maze revealed statistically significant increases in open arm time (F= 20.47; $p < 0.001$), closed arm time (F= 7.53; $p < 0.01$) and open arm entries (F= 34.79; $p < 0.001$) among female rats. Surprisingly, SPS

tended to further increase exploration of the open arms of the EPM (p= 0.06; d=0.67). In both males and females, SPS produced a reliable increase in rearing compared to Sham rats. Taken together these findings emphasize the importance of evaluating sex differences in the SPS model.

Disclosures: A. Mabie: None. **J. O'Russa:** None. **B. Huerta:** None. **M. Dillerud:** None. **T.E. Koeltzow:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.06

Topic: G.07. Post-Traumatic Stress Disorder

Support: VA Merit Award #BX004693
VA Merit Award #BX004646
NIMH F31-MH127890-01A1
NINDS T32-NS082145

Title: Inhibition of hippocampal and thalamic inputs to the nucleus accumbens reverses aberrant dopamine system function in a rodent model used to study PTSD and comorbid psychosis

Authors: *H. B. ELAM, A. M. MCCOY, A. M. BOLEY, D. J. LODGE;
UT Hlth. San Antonio, San Antonio, TX

Abstract: Post-traumatic stress disorder (PTSD) is a psychiatric illness that afflicts approximately 8% of the United States population. In addition to core symptoms of the disorder, patients with PTSD commonly present with a comorbid diagnosis, including psychosis. Symptoms of psychosis are thought to be driven by increased mesolimbic dopamine transmission, however, no clear histopathology has been identified within these cells. Rather, hyperactivity in upstream brain regions that regulate ventral tegmental area (VTA) dopamine neuron activity contributes to psychosis-like behavior. Two such regions are the paraventricular nucleus of the thalamus (PVT) and ventral hippocampus (vHipp), which synergistically regulate VTA dopamine neuron activity through a multisynaptic circuit that begins with convergent inputs to the nucleus accumbens (NAc). We have previously demonstrated that activation of PVT-NAc or vHipp-NAc projections significantly increases VTA dopamine neuron population activity, defined as the number of neurons firing spontaneously. These data suggest that hyperactivity in the PVT or vHipp, which are stress-sensitive brain regions, may contribute to psychosis-like behavior in a rodent model used to study PTSD. Interestingly, hippocampal and thalamic regulation of VTA population activity requires simultaneous activity from both regions, suggesting that targeting either region may be a novel site of intervention for comorbid psychosis related to PTSD. In this study, we induced stress-related pathophysiology in male Sprague Dawley rats using the two-day inescapable foot shock procedure. Following foot shock, we observed deficits in sensorimotor gating, as measured by prepulse inhibition of startle (PPI) and

an increase in VTA dopamine neuron population activity. Interestingly, chemogenetic inhibition of either PVT-NAc projections or vHipp-NAc projections, following stress, reversed deficits in PPI and restored dopamine system function. These results suggest that the PVT or vHipp may be novel therapeutic targets for decreasing psychosis symptoms observed in PTSD.

Disclosures: H.B. Elam: None. **A.M. McCoy:** None. **A.M. Boley:** None. **D.J. Lodge:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.07

Topic: G.07. Post-Traumatic Stress Disorder

Support: CIHR

Title: Neural activities in the anterior pretectal nucleus is associated with fear generalization in a mouse model of posttraumatic stress disorder

Authors: *X. QIN[1], X. YANG[2], Y. WANG[3];
[1]Dept. of Neurosci., Univ. of British Columbia, Vancouver, BC, Canada; [2]Shenzhen Inst. of Advanced Technol., Shenzhen, China; [3]Univ. British Columbia, Vancouver, BC, Canada

Abstract: Generalization of fear response towards innocuous stimuli is a highly prevalent symptom of PTSD. Although generalization of fear memories can protect animals by helping them avoid potential dangers, overgeneralization of fear can lead to anxiety-related mental disorders, including PTSD. Previous studies on fear generalization have heavily focused on the role of fear-related circuitry including the amygdala, prefrontal cortex, and hippocampus. Modulating these regions can reduce fear generalization, but it also increases the risk of being unable to elicit the correct fear response when encountering an actual threat. Previous researches have shown that stimulation of the Zona Incerta (ZI) could reduce fear generalization generated by auditory cues paired with high intensity foot-shocks. In this study, we aimed to determine if the APtN, a midbrain structure that has dense projections to the ZI, also plays a role in fear generalization. Our C-FOS images did not support a direct involvement of the APtN in neither acquisition of a fear memory nor elicitation of a proper behaviour response when the animals were placed in a traumatized environment. However, we did observe a linear relationship between the neural activities in the APtN and the animals' freezing rate in a fear generalization test 24 hours after the initial fear acquisition. Moreover, the observed relationship is validated in a long-term fear generalization paradigm, in which we trained the mice to associate high intensity foot-shocks with a contextual box and tested the fear response in a chamber that was visually similar to the training context after two months to mimic the real-world condition in human PTSD patients. Our results strongly suggest that APtN may play an important role in fear generalization. Together with the fact that it conveys sensory information to the thalamic and

subthalamic brain regions, the APtN may represent a novel therapeutic target for attenuating fear generalization without affecting fear learning and fear expression.

Disclosures: X. Qin: None. **X. Yang:** None. **Y. Wang:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.08

Topic: G.07. Post-Traumatic Stress Disorder

Support: Startup fund from the medical college of Georgia at Augusta University

Title: Establishment of the female mouse model of social avoidance induced by female-directed female aggression

Authors: *J. KIM, K. POKHAREL, S. MICHAEL, **C. KIM**;
Dept. of Neurosci. & Regenerative Med., Augusta Univ., Augusta, GA

Abstract: Although women are more likely than males to suffer from stress-related disorders such as posttraumatic stress disorder and depression, the majority of preclinical research on the impact of stress has been conducted on male subjects. Chronic social defeat stress (CSDS) is a rodent model of psychosocial stress that causes social avoidance and anhedonia. However, this model has been challenged in female mouse studies since neither male nor female resident mice attack intruder females. In this study, we aim to establish the female mouse model of social avoidance. We used ovariectomized (OVX) CD-1 female mouse with single- or a pair-housed setting with male CD-1 mouse. Given that the selection of aggressor is the critical step for the CSDS model, we first determined the attack latency and the number of attacks during the selection of aggressor. In the single-housed OVX mice, 34 percent of mice met the criterion of the selection of aggressor. There was a significant negative correlation between attack latency and the number of attacks (R^2=0.5660, P=0.009), suggesting that attack latency can be a good indicator of the number of attacks. However, single-housed OVX mice did not show reliable, aggressive behaviors (e.g., attack bites) during the CSDS. As a result, we did not find any behavioral changes in the social interaction test, elevated plus-maze test, and spontaneous alternation Y-maze test, suggesting the importance of aggressive behavior toward the intruder. In contrast, during the selection of aggressor, 42 percent of OVX mice in a pair-housed condition with male CD-1 mice satisfied the criterion and displayed consistently aggressive behaviors. CSDS produced susceptible (67 %) and resilient (33 %) phenotypes during the social interaction test. In addition, both susceptible and resilient mice showed anxiogenic-like behaviors in the elevated plus-maze test. Our study suggests that OVX CD-1 mice in a pair-housed setting show reliable, territorial aggression toward female intruders, producing susceptibility and resilience to social avoidance.

Disclosures: J. Kim: None. **K. Pokharel:** None. **S. Michael:** None. **C. Kim:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.09

Topic: G.07. Post-Traumatic Stress Disorder

Support: VA I01 BX005118

Title: Increased hypervigilance, startle response, and fear memory retention in an adolescent mouse reminders model of traumatic stress

Authors: *M. T. WATSON[1], J. G. TASKER[2];
[1]Tulane Brain Inst., [2]Cell and Mol. Biol., Tulane Univ., New Orleans, LA

Abstract: Post-traumatic stress disorder (PTSD), a trauma-related disorder affecting approximately 8% of the population, is associated with hyperactivity of the amygdala, a key region for adaptive stress and fear processing. Amygdala hyperactivity is suggested to be the cause of hyperarousal symptoms of PTSD, which include avoidance of cues related to trauma, hypervigilance, and increased startle response. This traumatic stress disrupts the excitatory and inhibitory balance within the basolateral nucleus of the amygdala (BLA), resulting in long-term maladaptive stress processing. This exploratory study is to determine how a chronic reminder of traumatic stress impacts hyperarousal symptoms and fear learning in an adolescent mouse model. Five-week-old male C57Bl6/J mice were used and a baseline acoustic startle response (ASR) to 15 110 dB white noise bursts delivered at a 30-s inter-tone interval (ITI) was recorded on day 1. On days 2-8, the mice were exposed to an adapted Traumatic Experience with Reminders of Stress (TERS) model which consisted of a 5-minute exposure to a pre-shock context followed by a 10-second, 2 mA foot shock in a secondary context. Subsequently, mice were reminded of the shock experience by exposure to the pre-shock context for 60-seconds once a day for another five days. The mice were retested on day 9 for changes to ASR. A 4-day fear conditioning protocol began on day 14, and on day 18 a final ASR was recorded. TERS-exposed mice had significantly higher motion ($p<0.0001$), time freezing ($p<0.01$), and freezing episodes ($p<0.001$) upon final reminder exposure compared to controls. While no significant increase to average ASR was observed, individual scores relative to baseline showed significant increases for TERS mice compared to controls ($p<0.01$), and this change increased in intensity after the final ASR ($p<0.0001$). Dramatic differences were observed in active escape behaviors from TERS mice during fear conditioning acquisition ($p<0.0001$). Differences in number and duration of freezing episodes were also observed throughout recall (day 15, $p<0.01$) and extinction (days 16-17, $p<0.001$). TERS-exposed mice showed greater fear memory acquisition ($p<0.01$) and a delayed extinction ($p<0.001$) compared to control mice. These results indicate that this adapted TERS model induces a hyperarousal phenotype, increases fear memory retention, and delays fear memory extinction in the adolescent mouse. This model will be used to further assess circuit level and cellular mechanisms underlying these behavioral phenotypes induced by traumatic stress.

Disclosures: **M.T. Watson:** None. **J.G. Tasker:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.10

Topic: G.07. Post-Traumatic Stress Disorder

Support: VA grant I01BX001075
NIH grant F32MH117913
NIH grant K99AA029168

Title: Angiotensin receptor signaling mediates fear-relevant behaviors via a subfornical organ to prefrontal cortex circuit

Authors: *K. M. J. MCMURRAY[1,2], R. SAH[3,2];
[1]Univ. of Cincinnati, Cincinnati, OH; [2]Veterans Affairs Med. Ctr., Cincinnati, OH; [3]Pharmacol. & Systems Physiol., Univ. of Cincinnati, Cincinnati, OH

Abstract: PTSD associates with dysregulated fear processing. Most prior fear-related research has focused on responses to externally evoked stressors. Yet mounting evidence suggests PTSD patients are more sensitive to homeostatic stressors such as CO2 inhalation which triggers acidosis and evokes fear-associated defensive behaviors in clinical and preclinical models. Studies suggest this sensitivity may exist prior to PTSD pathology. Therefore, investigating the relationship between homeostatic stressors like CO2 inhalation and externally evoked stressors may improve our understanding of PTSD vulnerability. We recently developed a mouse model examining the effect of CO2 inhalation on later PTSD relevant behaviors. We found CO2 exposure increased later shock-evoked fear extinction deficits. This associated with reduced neuronal activation within the infralimbic (IL) cortex which plays a primary role in fear extinction, suggesting a convergence of homeostatic CO2-sensing nodes and forebrain extinction circuits. We previously reported the subfornical organ (SFO) as a key homeostatic site regulating CO2-evoked fear. Here, we identified direct SFO to IL projections and used an intersectional chemogenetic approach to test the hypothesis that inhibiting SFO-IL projections during CO2 inhalation attenuates CO2-evoked defensive responding and delayed CO2-associated fear extinction deficits. We found inhibition of the SFO-IL circuit during CO2 inhalation reduced CO2-evoked defensive behaviors and attenuated CO2 evoked extinction deficits 1 week later. We then sought to identify specific cell types within the SFO mediating these effects. The SFO is known to mediate central responses to renin-angiotensin system (RAS) signaling and mounting evidence supports a role for RAS in fear regulation. Therefore, we investigated the role of RAS in our model. We identified a population of SFO-IL projecting neurons containing the angiotensin-type II receptor (AT1R). Inhibiting this population also reduced CO_2-evoked freezing and attenuated the delayed CO_2-associated fear extinction deficits one week later. Together these data point to a specific role for an AT1R associated SFO-IL circuit in threat

responding. Collectively, these data highlight AT1R SFO-IL projections as a key circuit facilitating the convergence of homeostatic threat sensing and long-term fear memory that may contribute to PTSD vulnerability.

Disclosures: K.M.J. McMurray: None. **R. Sah:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.11

Topic: G.07. Post-Traumatic Stress Disorder

Support: National Institute on Drug Abuse - psilocybin

Title: Psilocybin prevents symptoms of hyperarousal and enhances novel object recognition memory in rats exposed to the single prolonged stress paradigm

Authors: *C. R. DEL VALLE, H. R. SPARKMAN, M. M. NAYLOR, C. M. CRUEA, R. E. RICE, C. E. MILLER, B. E. BRAMLAGE, L. P. PUPPEL, M. L. BROWN, A. K. AL-OLIMAT, E. S. DIETZ, P. R. ZOLADZ;
Psychology and Neuroscience, The Sch. of Hlth. and Behavioral Sci., Ohio Northern Univ., Ada, OH

Abstract: Pharmacotherapy for stress-related psychological disorders remains inadequate. Patients who are treated with conventional pharmacological agents frequently report negligible symptom reduction, and, in most cases, less than 50% experience full remission. Clearly, there is a need for additional pharmaceutical research into both established and novel approaches to alleviate these conditions. Over the past several years, there has been a renewed interest in the use of psychedelics to aid in the treatment of psychological disorders. Several studies have reported promising results in patients with major depression, anxiety disorders, and post-traumatic stress disorder (PTSD) following treatment with psychedelic agents such as lysergic acid diethylamide (LSD), 3,4-methylenedioxymethamphetamine (MDMA), ayahuasca, ketamine, and psilocybin. However, the precise behavioral and neurobiological mechanisms for these effects remain unclear. Thus, we aimed to develop an animal model of PTSD that involved prophylactic treatment with psilocybin, a 5-HT_{2A} agonist, that could be used to further understand the mechanisms underlying the benefit of psychedelic substances in treating these disorders. Adult male and female Sprague-Dawley rats were subjected to the single prolonged stress (SPS) paradigm, including 2 hours of physical restraint, 15 minutes of forced swim, and ether vapor exposure until loss of consciousness. Five minutes following ether-induced loss of consciousness, the rats were intraperitoneally injected with vehicle (0.9% saline) or psilocybin (1 mg/kg). One week later, the rats underwent a battery of behavioral tests, including the elevated plus maze (EPM), startle response assessment, open field testing, and novel object recognition (NOR) testing. No effects of SPS or psilocybin were observed for EPM behavior. SPS led to

enhanced startle responses in males, but not females, which was prevented by psilocybin. SPS also increased locomotor activity in the open field in males, but not females, and this effect was not prevented by psilocybin. SPS had no impact on NOR memory in males, but enhanced memory in females. Interestingly, psilocybin administration, alone or in combination with SPS, enhanced NOR memory in males only. These findings support a complex interaction between the administration of psilocybin and the prevention of stress-induced behavioral sequelae that depends on both sex and the type of behavioral task.

Disclosures: **C.R. Del Valle:** None. **H.R. Sparkman:** None. **M.M. Naylor:** None. **C.M. Cruea:** None. **R.E. Rice:** None. **C.E. Miller:** None. **B.E. Bramlage:** None. **L.P. Puppel:** None. **M.L. Brown:** None. **A.K. Al-Olimat:** None. **E.S. Dietz:** None. **P.R. Zoladz:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); National Institute on Drug Abuse.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.12

Topic: G.07. Post-Traumatic Stress Disorder

Support: DARPA N66001-15-2-4057

Title: Timing of Vagus Nerve Stimulation During Fear Extinction Determines Efficacy in a Rat Model of PTSD

Authors: *R. R. SOUZA[1], M. B. POWERS[3], R. L. RENNAKER[1], S. A. HAYS[2], C. K. MCINTYRE[1], M. P. KILGARD[1];
[1]Sch. of Behavioral and Brain Sci., [2]Bioengineering, The Univ. of Texas at Dallas, Richardson, TX; [3]Baylor Univ. Med. Ctr., Dallas, TX

Abstract: The first-line psychotherapies used in the treatment of Posttraumatic Stress Disorder (PTSD) are exposure-based therapies, such as Prolonged Exposure (PE). PE aims to extinguish conditioned fear through repeated in vivo or imaginal exposure to reminders of the traumatic experiences, reducing the maladaptive fear and attenuating other symptoms in PTSD. However, large clinical trials indicate suboptimal response rates, and the current research agenda has shifted to strategies that can augment the efficacy of PE. We have demonstrated that vagus nerve stimulation (VNS) during extinction trials enhances extinction learning in different rat models for PTSD, suggesting that VNS could be used as an effective adjuvant to exposure therapies. Here we investigated if precisely pairing VNS with the presentation of the conditioned stimulus (CS) is critical for VNS enhancement of extinction learning. Adult Sprague-Dawley rats were exposed to an intense stress followed by fear conditioning training to produce extinction-resistant conditioned fear. The rats were then implanted with a cuff electrode around the left vagus. After recovery, the animals underwent extinction training paired with VNS (0.5 s, 0.8 mA, 100 μs, and

30 Hz) or with Sham VNS (0 mA). VNS rats were randomized into the following subgroups: During VNS (delivered during presentations of the CS), Between VNS (delivered between CS presentations), Continuous VNS (delivered during the entire extinction session), and Dispersed VNS (delivered at longer inter-stimulation intervals across the extinction session). Sham VNS rats failed to extinguish the conditioned fear response over five days of repeated exposure to the CS. Rats that received Between or Dispersed VNS showed modest improvement in conditioned fear at the retention test. During and Continuous VNS groups displayed the greatest reduction in conditioned fear. These findings indicate that delivering VNS paired precisely with CS presentations or continuously throughout extinction promotes the maximum enhancement in extinction learning. Translational insights into the use of VNS to augment the efficacy of PE therapy in individuals with PTSD will be discussed.

Disclosures: **R.R. Souza:** None. **M.B. Powers:** None. **R.L. Rennaker:** None. **S.A. Hays:** None. **C.K. McIntyre:** None. **M.P. Kilgard:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); MicroTransponder.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.13

Topic: G.07. Post-Traumatic Stress Disorder

Support: Internal NYMC Funds

Title: Potential mediators of divergent response to single prolonged stress model of PTSD in female rats

Authors: *A. TANELIAN, B. NANKOVA, A. CHERIYAN, C. ARENS, F. HU, E. L. SABBAN;
New York Med. Col., New York Med. Col., Valhalla, NY

Abstract: Exposure to traumatic stress is a major risk factor for development of neuropsychiatric disorders in a sub-population of individuals, while others remain resilient. The contributing factors differentiating between these phenotypes are still unclear. The majority of the studies assessing the potential factors predisposing to host susceptibility or resilience to stress induced psychopathologies have been conducted using male animals. Here, we aimed at characterizing the microbial, immunological and molecular differences of stress susceptible and resilient Sprague Dawley female rats before and after exposure to traumatic stress. Animals were randomly divided into unstressed controls and experimental group exposed to Single Prolonged Stress (SPS), an animal model for PTSD. Fourteen days later, all rats underwent a battery of behavioral tests and were sacrificed one day later to collect cecum, ventral hippocampus (Vhipp), and medial prefrontal cortex (mPFC). Stool and urine samples were collected both

before and after SPS. The behavioral analyses on the elevated plus maze (EPM) test revealed that about half of the animals displayed high anxiety-like behavior, whereas the other half behaved like the unstressed controls. Due to this divergent response, the SPS group was further subdivided into SPS-Resilient (R) and SPS-Susceptible (S) subgroups. The SPS-S subgroup showed impaired social interaction (SI) relative to the SPS-R subgroup, and the time spent interacting on SI correlated significantly ($r=0.83$, $p<0.0001$) with Anxiety Index on the EPM. Interestingly, the levels of urinary epinephrine at baseline and during SPS were higher in SPS-S subgroup. Fecal 16S rDNA sequencing also revealed differences in the gut microbial composition between the subgroups both before and after SPS, and the analysis of the cecal short chain fatty acids showed significantly higher levels of isobutyrate, isovalerate and valerate in SPS-S subgroup relative to the SPS-R subgroup. In line with the observed distinct behavioral phenotypes, the levels of inflammatory cytokines, Il-1b and Il-6 were significantly higher in the Vhipp of SPS-S subgroup relative to the controls and the SPS-R subgroup. In contrast, the expression levels of tight junction protein claudin-5 in the Vhipp and mPFC were higher in SPS-R subgroup relative to the SPS-S and the controls, indicating lower blood-brain-barrier permeability. The results reveal potential contribution of gut-brain axis in the susceptibility or resilience to traumatic stress in females. Further characterization of these factors will be crucial to understand susceptibility and foster resiliency.

Disclosures: **A. Tanelian:** None. **B. Nankova:** None. **A. Cheriyan:** None. **C. Arens:** None. **F. Hu:** None. **E.L. Sabban:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.14

Topic: G.07. Post-Traumatic Stress Disorder

Support: UT Health San Antonio Center for Biomedical Neuroscience Pilot Project Grant

Title: Neurophysiological and behavioral effects of mPFC tDCS in a rodent model used to study PTSD

Authors: *A. BOLEY, S. M. PEREZ, D. MORILAK, C. L. STRAUD, D. J. LODGE, F. R. CARRENO;
UT Hlth. San Antonio, San Antonio, TX

Abstract: Approximately 16 million people in the U.S. will experience a traumatic event and eventually develop posttraumatic stress disorder (PTSD). Symptoms of PTSD can be debilitating and include fear, anxiety, and uncontrollable negative thoughts about the event. Unfortunately, current interventions for PTSD include pharmacological and behavioral therapies, which have limited efficacy due to the lack of treatments that target the pathophysiology associated with PTSD. The anatomical structures involved in processing of fear related information are well

known and include the prefrontal cortex (PFC), amygdala, and the hippocampus. The PFC is central to cognitive flexibility, extinction of conditioned fear, and volitional regulation of negative emotion – all of which are altered in PTSD. Importantly, hypoactivity of the PFC is a consistent observation in functional imaging studies and in rodent models used to study PTSD. Advances in non-invasive brain stimulation allow for a novel approach to modulate PFC activity. Specifically, transcranial direct current stimulation (tDCS) is an innovative approach to neuromodulation that targets superficial areas of the brain. Anodal stimulation can cause depolarization and increase neural excitability, which can increase prefrontal cortical function. We posit that anodal tDCS will reverse PFC hypoactivity and may serve as a novel therapeutic approach for the treatment of PTSD. Here, we apply tDCS to the PFC of rats to investigate the neurophysiological alterations in brain activity. We observed an increase in c-fos expression in rats given tDCS demonstrating activation of the PFC. In addition, preliminary data revealed that combining tDCS with a sub-effective extinction protocol, enhanced the efficacy of extinction alone in reversing stress-induced deficits in set shifting, a measure of cognitive flexibility. Further, we investigated a downstream, stress sensitive brain region and found that tDCS increased dopamine neuron activity in the ventral tegmental area (VTA), where decreases are thought to underlie anhedonia. Taken together, these data provide initial evidence supporting the effects of PFC tDCS to treat symptoms of PTSD.

Disclosures: A. Boley: None. **S.M. Perez:** None. **D. Morilak:** None. **C.L. Straud:** None. **D.J. Lodge:** None. **F.R. Carreno:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.15

Topic: G.07. Post-Traumatic Stress Disorder

Support: Northwestern University Office of Undergraduate Research

Title: Extinction of stress-enhanced fear in a genetic stress-hyperreactive rat model

Authors: *A. M. HARTER, M. NEMESH, M. T. JI, L. LEE, A. YAMAZAKI, C. S. KIM, E. E. REDEI;
Psychiatry and Behavioral Sci., Northwestern Univ., Evanston, IL

Abstract: Post-Traumatic Stress Disorder (PTSD) has a lifetime prevalence of 7 - 8% and is the fifth most common major psychiatric disorder in the United States. PTSD has been related to exaggerated fear memory. Prior stress is known to enhance fear conditioning and the Stress Enhanced Fear Learning (SEFL) model is used for modeling enhanced and prolonged fear memory. Because women are twice as likely as men to develop PTSD, we employed female rats of two nearly isogenic inbred strains with differing stress reactivity to study extinction of fear memory after SEFL. The inbred Wistar Kyoto (WKY) Less Immobile (WLI) and Wistar Kyoto

More Immobile (WMI) strains show genetic predisposition to less, and more stress reactivity and passive coping behaviors, respectively. Adult WLI and WMI females were randomly divided into a control group (not stressed) and a stressed group, the latter exposed to a two-hour restraint stress (stressed). 48 hours after the restraint stress, both groups underwent a Contextual Fear Conditioning (CFC) test using three one-sec foot shocks (one per minute), of 0.8 mA intensity. 24 hours later, the animals were returned to the CFC chamber for three minutes without shock and their freezing behavior observed. Extinction was carried out for a week by reintroducing the animals to the CFC chamber without shock. Fear memory, as measured by freeze duration during the second day of CFC, was significantly greater in WMI females regardless of prior stress compared to WLIs [strain, $F(1,26)=19.4$; $p<0.001$]. As expected, the inverse was observed for distance traveled, where WLIs were more active than WMI females [strain, $F(1,26)=13.1$; $p<0.001$]. During extinction, the same strain differences were preserved in the first two days, but disappeared in the subsequent days [days, $F(7,185)=42.4$; $p<0.001$; strain, $F(1,28)=6.4$; $p<0.05$; days x strain, $F(7,185)=5.6$; $p<0.001$]. Thus, the genetic differences in stress-responsiveness between the strains were more impactful than the restraint stress-induced differences in fear memory and extinction within the strain. This suggests that individual stress sensitivity contributes to the exaggerated fear memory component of PTSD but not the vulnerability to prolonged fear memory.

Disclosures: **A.M. Harter:** None. **M. Nemesh:** None. **M.T. Ji:** None. **L. Lee:** None. **A. Yamazaki:** None. **C.S. Kim:** None. **E.E. Redei:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.16

Topic: G.07. Post-Traumatic Stress Disorder

Support: NIH Grant DA048241
NIH Grant DA034721
NIH Grant DA053781
NIH Grant DA047054
NIH Grant DA007244

Title: The Role of Hippocampal Astrocytes in Heroin Withdrawal and Future Enhanced Fear Learning

Authors: *S. PAREKH[1], J. E. PANICCIA[2], L. O. ADAMS[1], G. BARKELL[1], K. J. REISSNER[1], D. T. LYSLE[1];
[1]Psychology and Neurosci., Univ. of North Carolina, Chapel Hill, NC; [2]Med. Univ. of South Carolina, Charleston, SC

Abstract: Converging evidence suggests the neural immune system is altered following opioid use and withdrawal and post-traumatic stress disorder (PTSD). Using an innovative rodent model that combines escalating heroin administration and spontaneous withdrawal with enhanced fear learning, we demonstrated that heroin withdrawal enhanced fear learning is driven by dorsal hippocampal (DH) interleukin-1 (IL-1) and tumor necrosis factor alpha (TNF-α) signaling, and withdrawal from escalating heroin administration induces both DH IL-1β and TNF-α expression. Additionally, the IL-1β and TNF-α involved in heroin withdrawal enhanced fear learning is primarily expressed in hippocampal astrocytes. Interestingly, we found that heroin withdrawal upregulates hippocampal glial fibrillary acidic protein (GFAP), a marker for astrocyte reactivity. Therefore, the goal of the current studies was to investigate the hypothesis that heroin withdrawal enhances fear learning through astrocyte activity, as well as alterations in astrocyte physiology. To demonstrate astrocyte activity is causally related to heroin withdrawal-induced fear learning, Experiment 1 employed G_i-coupled designer receptors exclusively activated by designer drugs (DREADDs) to manipulate astrocyte signaling *in vivo*. Male rats were infused with AAV8-GFAP-hM4D(G_i)-mCherry into the DH, and underwent the chronic escalating heroin administration procedure. Clozapine-n-oxide (3 mg/kg, s.c.) or vehicle was administered 0-hour, 24-hour, and 48-hour into heroin withdrawal, and animals were then exposed to the enhanced fear learning paradigm. To test if withdrawal from chronic heroin altered astroglial structure and synaptic interactions, Experiment 2 used an established method to visualize DH astrocytes in rich detail using a membrane-tagged GFP, advanced microscopy, and image analysis. Male rats were administered bilateral intra-DH infusions of AAV5-GFAP-Lck-GFP, and underwent the chronic, escalating heroin administration procedure. Tissue was collected 24-hour into heroin withdrawal. Using high resolution confocal microscopy and analysis of 3-dimensional cellular reconstructions, astrocyte surface area, volume, and colocalization with postsynaptic density-95 were quantified to assess the consequence of chronic heroin and spontaneous withdrawal on astrocyte structure and neuronal interactions. Together, these studies examine the role of hippocampal astrocytes in heroin withdrawal-enhanced fear learning.

Disclosures: S. Parekh: None. **J.E. Paniccia:** None. **L.O. Adams:** None. **G. Barkell:** None. **K.J. Reissner:** None. **D.T. Lysle:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.17

Topic: G.07. Post-Traumatic Stress Disorder

Support: NIH Grant DA048241
NIH Grant DA034721
NIH Grant DA053781
NIH Grant DA047054
NIH Grant DA007244

Title: Mdma administration attenuates stress-enhanced fear learning and severe stressor-induced il-1β immunoreactivity

Authors: *L. ADAMS, S. PAREKH, D. T. LYSLE;
Psychology and Neurosci., Univ. of North Carolina, Chapel Hill, NC

Abstract: Post-traumatic stress disorder (PTSD) is a chronic and devastating disorder resulting from a traumatic experience that is also associated with biological alterations in several immune system pathways. Our laboratory has investigated the mechanism of stress-enhanced fear learning (SEFL), an animal model of PTSD, and found that the severe stressor in SEFL paradigm induces a time-dependent region-specific increase in dorsal hippocampal (DH) interleukin-1β (IL-1β) in astrocytes. Further, we show that blockade of IL-1β attenuates this stress-enhanced fear learning, suggesting that IL-1β signaling is necessary for the development of enhanced fear learning. Recently, clinical trials have demonstrated 3,4-methylenedioxymethamphetamine (MDMA)-assisted psychotherapy is highly efficacious in individuals with severe PTSD, and that this treatment is safe and well-tolerated by patients. Interestingly, MDMA has a suppressive effect on cytokines such as IL-1β and tumor necrosis factor-alpha (TNF-α). Therefore, we hypothesized that MDMA may be effective for PTSD in part due to these immunosuppressive effects. In Experiment 1, we examined MDMA's effect on enhanced fear learning by administering three, 10mg/kg doses of either MDMA or saline at 0 hours, 24 hours, and 48 hours following the severe stressor in the SEFL paradigm. Excitingly, we demonstrated that MDMA administration following the severe stressor attenuated enhanced fear learning. In Experiment 2, we used fluorescent immunohistochemistry to examine MDMA's effect on the neuroimmune and cellular markers IL-1β, TNF-α, glial fibrillary acidic protein (GFAP), and ionized calcium-binding adaptor protein-1 (IBA- 1) following the severe stressor. Interestingly, MDMA administration attenuates IL-1β immunoreactivity following a severe stressor, but has no effect on TNF-α, GFAP, or IBA-1 immunoreactivity. Overall, these experiments indicate that MDMA inhibits the development of stress-enhanced fear learning, and suggest that it may function mechanistically by inhibiting IL-1β immunoreactivity.

Disclosures: L. Adams: None. **S. Parekh:** None. **D.T. Lysle:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.18

Topic: G.07. Post-Traumatic Stress Disorder

Support: EUROSTARS E! 113468 - DiSARM FEAR Special

Title: A novel Post Traumatic Stress Disorder (PTSD) model in the Jeju minipig

Authors: *J. H. LEE[1], K. H. PARK[2], T. H. KIM[1], B. Y. LIM[1], S. C. LEE[3], Y. J. SONG[3], P. J. SWEENEY[1], J. E. FRIEDMAN[1], L. C. PARK[1];

[1]Naason Sci., Cheongju-si, Korea, Republic of; [2]KNU / Naason, Cheongju, Korea, Republic of; [3]Cronex Inc., Cheongju-si, Korea, Republic of

Abstract: PTSD is a serious condition that can lead to intense fear, anxiety and depression. Childhood exposure to highly stressful events has been found to increase the possibility that the adult will be more prone to suffer from PTSD. Current medication is not very effective and has a large number of side effects. As a result, many people continue to suffer from PTSD, indicating a critical need for new treatments. One limiting factor in the development of treatment for PTSD is the lack of valid, translational models. Naason Science has been developing models of PTSD in the rat and in the minipig, based on multi-faceted, prolonged stress paradigms, so as to test the efficacy of new compounds. Behavioral profiling analysis is employed capturing individual differences in response to trauma and treatment. We also expose the animals to stress as juveniles in order to increase their susceptibility to PTSD as adults. Pigs are intelligent animals that not only closely share physiology with humans, such as the cardiovascular system, but demonstrably show emotions, including fear. This is different from many animals that do not overtly express fear nor discomfort, making pigs viable candidates for behavioral studies. Jeju minipigs from the same litter, mixed gender, were used, n=4-6 per group, one group being a control for the group that experienced stressors. Over a 5-week period, piglets are exposed to predator urine, restraint and brief isolation from the mother sow. Piglets 1 week old were separated from their mothers for 2 hours/day until weaning, restrained in a net for 10 min/day and exposed to predator urine and sound. Both sets of animals appeared to thrive as they experienced similar weight gain. Animal behavior was subsequently tested 2 times/week in an open field (OF) from week 6, and for 20 hours in their home cage at week 7. Minipigs were recorded and analyzed using our digitized video recording and AI analysis system. Both control and PTSD pigs occupied the same zones in the OF, but by day 4 of testing PTSD pigs only moved 20% of the distance that their control littermates moved over the same time period. PTSD pigs experienced longer periods of freezing, about 3-fold that of their control littermates. In their home cage, PTSD minipigs demonstrated significantly greater social distancing from their littermates compared to controls, both day (0.15 vs. 0.11m) and night (0.21 vs. 0.1m). Changes in MRI (volume) and MRS (metabolites) in brains of PTSD minipigs were also evaluated. This study demonstrates how early stress can affect simple behavior in juvenile pigs and can serve as a model system for the development of PTSD in large animals and possibly provide for a translational model for humans.

Disclosures: **J.H. Lee:** A. Employment/Salary (full or part-time):; Naason Science Inc. **K.H. Park:** A. Employment/Salary (full or part-time):; Naason Science. **T.H. Kim:** A. Employment/Salary (full or part-time):; Naason Science. **B.Y. Lim:** A. Employment/Salary (full or part-time):; Naason Science. **S.C. Lee:** A. Employment/Salary (full or part-time):; Cronex Inc. **Y.J. Song:** A. Employment/Salary (full or part-time):; Cronex Inc. **P.J. Sweeney:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Naason Science Inc. F. Consulting Fees (e.g., advisory boards); Naason Science Inc. **J.E. Friedman:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Naason Science. F. Consulting Fees (e.g., advisory boards); Naason Science. **L.C. Park:** A. Employment/Salary (full or part-time):; Naason Science. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Naason Science Inc.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.19

Topic: G.07. Post-Traumatic Stress Disorder

Support: EUROSTARS E! 113468 - DiSARM FEAR

Title: A novel Post Traumatic Stress Disorder (PTSD) model in the rat

Authors: *J. E. FRIEDMAN[1], J. LEE[1], B. Y. LIM[1], T. H. KIM[1], E. S. JOO[1], T. H. KIM[1], K. PARK[2], P. J. SWEENEY[1], L. C. PARK[1];
[1]Naason Sci., Cheongju-si, Korea, Republic of; [2]KNU / Naason, Cheongju, Korea, Republic of

Abstract: PTSD is a serious condition that can lead to intense fear, anxiety and depression, resulting in social distancing and other behavioral disorders. Childhood exposure to highly stressful events increases the likelihood that the adult will be more prone to suffer from PTSD. Current medication, usually antidepressants or anxiolytics, is unspecific, ineffective and has a number of side effects (sedation, sexual dysfunction). As a result, many people continue to suffer from PTSD in spite of current treatment options, indicating a critical need for new treatments. A limiting factor in the development of treatment for PTSD is the lack of valid translational models. Naason Science has been developing models of PTSD in the rat and the minipig based on a multi-faceted, prolonged stress paradigm so as to test the efficacy of new compounds. Animals are stressed as juveniles in order to increase their susceptibility to PTSD as adults. We have used Desipramine + Methylphenidate, Fluoxetine or Diazepam as controls in the rat. Juvenile male SD rats, n=12/group, are exposed to different stressors over a 2-day period, comprised of restraint (2h), swimming (1m), submersion (30s) in the Water Associated Zero Maze (WAZM) and cued electric shock (x5). This paradigm is repeated weekly for another 2 weeks. The behavior of the rats is recorded using our Home Cage Activity (HCA) monitoring system, recording the behavior of the mixed group housed animals 24/7. Behavior is automatically assessed using AI computerized systems. After being exposed to the 3-week PTSD paradigm subjects are treated for 5 days and then exposed to the external track of the WAZM and subsequently to cued fear conditioning (cFC), novel object recognition (NOR) and HCA monitoring. This is repeated after another 30 days. Upon terminating the study blood samples are collected. After 2 weeks, test subjects exhibited signs of anxiety and antisocial behavior, persisting for at least 60 days. In the WAZM, normal rats spent 34±9sec in the open arms with 5 entries, while PTSD rats entered for 17.8±6sec with 2 entries total. Fluoxetine (10mpk) had no beneficial effect. In the NOR test PTSD rats spent significantly less time exploring the novel object than did the unstressed rat (63.9±3sec vs. 73±2sec). Fluoxetine (10mpk) was effective in this case (72.3±5sec). Normal rats move significantly more than PTSD rats (78±3.7m vs 54±2.7m), unaffected by Fluoxetine. PTSD rats exhibited significantly less social (interactive) activity by 21%. The model presented here is a simple, robust model of PTSD that should

facilitate dissecting out various aspects of this disorder and provides a screening paradigm for new medical treatments.

Disclosures: **J.E. Friedman:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Naason Science. F. Consulting Fees (e.g., advisory boards); Naason Science. **J. Lee:** A. Employment/Salary (full or part-time):; Naason Science. **B.Y. Lim:** A. Employment/Salary (full or part-time):; Naason Science. **T.H. Kim:** A. Employment/Salary (full or part-time):; Naason Science. **E.S. Joo:** A. Employment/Salary (full or part-time):; Naason Science. **T.H. Kim:** A. Employment/Salary (full or part-time):; Naason Science. **K. Park:** A. Employment/Salary (full or part-time):; Naason Science. **P.J. Sweeney:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Naason Science. F. Consulting Fees (e.g., advisory boards); Naason Science. **L.C. Park:** A. Employment/Salary (full or part-time):; Naason Science. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Naason Science.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.20

Topic: G.07. Post-Traumatic Stress Disorder

Support: NIH R01-MH115678
Staglin Center for Brain and Behavioral Health

Title: Cfos expression during stressors significant and non-significant enough to produce stress enhanced fear learning

Authors: *S. Y. MARKOWITZ[1], L. LAUGHLIN[1], M. S. FANSELOW[2];
[1]Psychology, [2]UCLA, UCLA Chapter, Los Angeles, CA

Abstract: Approximately 10% of people who experience a highly stressful event, or trauma, will go on to develop Post-Traumatic Stress Disorder (PTSD). As the majority of people who experience intense stress do not develop PTSD, an important question is what makes a stressful event “significant” enough to cause this disorder to manifest. It has previously been established in an animal model of PTSD called Stress Enhanced Fear Learning (SEFL) that not all stressors are significant enough to cause PTSD-like symptoms, even if both experiences cause comparable levels of fear. Specifically, rats that experienced 15 presentations of (1s, 1mA) Shock Stress showed future SEFL, while rats that experienced 15 presentations of (1s, 120 dB) Noise Stress did not. Additionally, only Shock Stress animals exhibited a panic behavior response to stressor presentations. Shock animals exhibited a flight response to each stressor, followed by significantly more movement and thigmotaxis; Noise animals exhibited a simple startle response

to each stressor. We concluded that shock is a Significant enough stressor to cause SEFL, while noise was Nonsignificant. The periaqueductal grey (PAG) is a key brain region for defense response behavior. It is well established that activity in the PAG is both necessary and sufficient for both fear and panic response toward external stressors. We hypothesized that there would be a difference in activity levels in the PAG between animals experiencing Significant or Nonsignificant stress. Rats were first exposed to either 15 presentations of Shock Stress or 15 presentations of Noise Stress. 30-45 minutes after the final stressor presentation, these experimental rats, as well as homecage controls, were perfused. Brain slices from central PAG stained for c-Fos expression showed a significant difference in activity in the lateral PAG (lPAG) and ventral-lateral PAG (vlPAG) between Shock Stress and Noise Stress animals. Thus, we have begun to characterize which specific subregions of the PAG correlate to exposure to "Significant" and "Non Significant" stress, which in turn may better help us understand the underlying neural mechanisms of PTSD.

Disclosures: S.Y. Markowitz: None. **L. Laughlin:** None. **M.S. Fanselow:** F. Consulting Fees (e.g., advisory boards); Neurovation Inc.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.21

Topic: G.07. Post-Traumatic Stress Disorder

Support: NIMH R01-MH115678
NIDA T32-DA024635
NIDA F31-DA054792

Title: Stress Enhanced Fear Learning enhances excitatory synaptic transmission in Basolateral Amygdala neurons

Authors: *J. E. MONDELLO[1], C.-W. CHANG[1], J. M. TROTT[2], A. ANAYA[1], S. SOLORIO[1], L. TRAN[1], M. S. FANSELOW[1];
[1]Psychology, [2]Neurosci., Univ. of California Los Angeles, Los Angeles, CA

Abstract: Post-traumatic stress disorder (PTSD) is marked by maladaptive learning processes that lead to inappropriately exaggerated fear responses following a traumatic experience. In order to recapitulate some of the symptomology of PTSD, our laboratory developed a rodent stress model, termed Stress Enhanced Fear Learning. In this stress model, a single traumatic stressor (10 footshocks across 60 min) leads to heightened fear learning and defensive behaviors. Our lab and others have shown that stress leads to hyperexcitability and neural plasticity in the basolateral amygdala (BLA), leading to an enhanced propensity for future associative learning. In the present set of studies, we further tested the functional impact of traumatic stress on BLA synapses. We conducted whole-cell voltage clamp recordings of BLA neurons in acute brain

slices from adult male and female C57BL/6J mice in order to compare miniature excitatory postsynaptic currents (mEPSCs) between animals that received traumatic stress or no stress. While there was no difference in the amplitude of the mEPSCs, there was increased frequency of mEPSCs in the neurons of stressed mice. Next, we utilized a novel activity-dependent labeling AAV (AAV9-RAM-d2TTA:TRE-hM4Di-mCherry-WPREgamma, "RAM" virus) that fluorescently tags neurons that have expressed c-fos and/or Npas4, immediate-early genes associated with neuronal activity. This custom viral vector allows for robust and temporally specific labeling and inhibition of previously activated neurons, i.e. "engram" cells. BLA neurons were tagged during traumatic stress in male and female mice and then whole-cell voltage clamp recordings were performed one day after the stress. We confirmed in stressed mice that there was increased mEPSC frequency, but not amplitude, in tagged neurons vs. untagged neurons. Taken together, these data suggest that trauma promotes excitatory input onto BLA neurons, likely through either increased presynaptic neurotransmitter release and/or number of synapses. Our findings additionally provide partial validation of the novel RAM virus and indicate we can further dissect functional difference between different neuronal populations based on their prior activity history.

Disclosures: **J.E. Mondello:** None. **C. Chang:** None. **J.M. Trott:** None. **A. Anaya:** None. **S. Solorio:** None. **L. Tran:** None. **M.S. Fanselow:** Other; Neurovation Labs, Inc..

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.22

Topic: G.07. Post-Traumatic Stress Disorder

Support: MOST109-2320-B-001-010

Title: Early- and late-phase alternations of the brain wave activity and early-phase intervention of neuromodulation in the post-traumatic stress disorder rat model

Authors: *S. CHANG[1], F. SHAW[2], B. SHYU[1];
[1]Inst. of Biomed. Sci., Academia Sinica, Taipei, Taiwan; [2]Dept. of Psychology, Natl. Cheng Kung Univ., Tainan, Taiwan

Abstract: Background Post-traumatic stress disorder (PTSD) is a complex syndrome, which may occur after exposure to life-threatening events. Fear memory with abnormally enhanced consolidation and impaired extinction have been indicated a vital role in PTSD. Besides, the expression of fears has also been found strongly correlated with the theta (4Hz) activities in animal models. However, the relation between longitudinal fear formation and potential brain wave features after traumatic stress exposure still remains largely unknown.**Aims & Objectives**We hypothesized that after traumatic stress exposure, the longitudinal dynamic changes of abnormal fears in PTSD models could be reflected by local field potentials (LFPs)

and could be further differentiated from the conditioned fear model.**Methods**In our study, we use a well-established modified single prolonged stress (SPS&FS) PTSD rat model and compared it with a conditioned fear model and control (each group n=5). In the PTSD model, animals were restrained for 2 hrs and followed by 20 mins forced swimming, after that, rats were exposed to diethyl ether until lose consciousness, then are kept in a conditioning chamber until they awake, foockshock (2 shocks (1.5 mA) trains of 1 s / per minute (total 5 min, 10 shocks)) was applied after awaking. Microwires for LFPs recording were implanted in the medial prefrontal cortex, the amygdala and the ventral hippocampus, brain waves were recorded after traumatic stress exposure at the early (10 mins, 30 mins, 2,4, 6 hrs) and late phases (day 1, 3, 7, and 14) during context re-exposure. All data were presented as Mean ± SEM, and statistical results were calculated by one-way analysis of variance (ANOVA) (behavioral indexes) and two-way ANOV (indexes of brain wave activities). **Results**The SPS&FS rats showed heterogenous PTSD phenotypes (anxiety, depression and anhedonia) with sustained fears. And in the PTSD model, time-dependent higher theta (5-8 Hz) and gamma (>30 Hz) activities were observed only in the early phases (10 mins and 30 mins post SPS&FS) and started to shift to continuous lower delta (0.5-4Hz) from the time points of 2-4 hrs to the late phase (day 1-14), the patterns could also be differentiated from the conditioned fear model. Later, the PTSD phenotypes could be alleviated with the application of transcranial direct current stimulation (tDCS). **Conclusion**Our results revealed the time-dependent inter-regional changes after traumatic stress exposure, the understanding of these longitudinal changes could help find the important biomarkers at specific phases after traumatic stress exposure.**Keywords:** PTSD, brain synchrony, traumatic memory, fear, temporal development, tDCS.

Disclosures: **S. Chang:** None. **F. Shaw:** None. **B. Shyu:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.23

Topic: G.07. Post-Traumatic Stress Disorder

Support: University of Oslo, Forskerlinjen
National Institute of Mental Health Grant R01MH098003
National Institute of Neurological Disorders and Stroke Grant R01NS085200
National Institute of Mental Health Grant RF1MH114224
National Institute of Mental Health Grant R37MH058883

Title: Spine density in the anterior cingulate cortex (ACC) is dependent on trait anxiety after trauma in a rat model of Post-Traumatic Stress Disorder (PTSD)

Authors: *S. DAVANGER[1], E. SANDERS[2], M. HANSEN[3], A. SAADAT[2], D. DOPFEL[6], P. D. PEREZ[7], P. LAAKE[4], R. HOLST[5], S. HUSSAIN[1], B. S. MCEWEN[8], N. ZHANG[9];
[2]Div. of anatomy, [3]Dept. of Psychology, [4]Dept. of Biostatistics, [1]Univ. of Oslo, Oslo, Norway;

[5]Univ. of Oslo, Department of Biostatistics, Norway; [6]Bioengineering, [7]Pennsylvania State Univ., University Park, PA; [8]Lab. of Neuroendocrinology, Rockefeller Univ., New York, NY; [9]The Pennsylvania State Univ., University Park, PA

Abstract: The anterior cingulate cortex (ACC) is involved with emotional processing and attentional regulation. It has been shown to decrease in size after PTSD. Others have shown, however, that synapse density may increase in the ACC in the long term after trauma. We wanted to determine if the increase in spine density is linked to trait anxiety, using a rat model of post-traumatic stress disorder (PTSD). Adult male Long Evans rats were exposed to a single episode of predator odor in an inescapable environment. After exposure, the rats were returned to their home cage for six days, before Elevated Plus Maze (EPM) testing was performed to measure levels of trait anxiety. Rats were placed in the middle of the maze facing an open arm, and then letting them freely explore the EPM for five minutes. The percentage of time spent in the open arms [open/(open + closed)] was calculated, where low EPM scores signifies high levels of anxiety. We perfusion fixed 33 male rats 3-4 days after their EPM testing with a mixture of formaldehyde and glutaraldehyde, before processing the right hemisphere of each rat for Golgi-Cox staining with the Hito Golgi-Cox Optimstain TM kit. The left hemisphere was processed further with freeze substitution and embedding in Lowicryl HM 20 for later ultrathin sectioning and post-embedding immunogold labelling for electron microscopy (EM). Golgi-stained sections were imaged with a Leica DM5500 B microscope with a 100x objective, preparing z-stacks for analysis with NeuronStudio and preparation of 3-D models for quantification of dendritic spine densities. Three different regions of interest were chosen: ACC, hippocampus CA1 region (CA1), and the primary somatosensory cortex (S1). Dendritic spine density measurements were performed on primary branches from apical dendrites of pyramidal neurons (layer 5 in ACC and S1). Linear mixed effects model analysis with R showed that spine density was a significant function of anxiety levels (EPM) ($p = 0.008$) in ACC, but not in CA1 or S1. When broken down in different morphological spine types, the significance increased to $p<0.001$ for mushroom spines (mature synapses), but not for the other spine types. EM immunogold analysis showed that concentration of GluN2B-subunit containing NMDA receptors was significantly lower in the PSD of high anxiety rats compared to both low-anxiety rats and a control group which was not exposed to trauma. In conclusion, high anxiety levels in rats six days after being subjected to trauma, predicts high synaptic density in ACC, a prominent part of the default mode network, with important functions in emotional regulation.

Disclosures: S. Davanger: None. **E. Sanders:** None. **M. Hansen:** None. **A. Saadat:** None. **D. Dopfel:** None. **P.D. Perez:** None. **P. Laake:** None. **R. Holst:** None. **S. Hussain:** None. **B.S. McEwen:** None. **N. Zhang:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.24

Topic: G.06. Anxiety Disorders

Support: NIH R01 MH083862

Title: Contribution of the opioid system to the antidepressant effects of fluoxetine

Authors: *P. NGUYEN;
Columbia Univ., New York, NY

Abstract: Background: Selective serotonin reuptake inhibitors such as fluoxetine have a limited treatment efficacy. The mechanism by which some patients respond to fluoxetine while others do not remains poorly understood, limiting treatment effectiveness. We have found the opioid system to be involved in the responsiveness to fluoxetine treatment in a mouse model for anxiety- and depressive-like behavior.**Methods:** We analyzed gene expression changes in the dentate gyrus of mice chronically treated with corticosterone and fluoxetine. After identifying a subset of genes of interest, we studied their expression patterns in relation to treatment responsiveness. We further characterized their expression through in situ hybridization and the analysis of a single-cell RNA-Seq data set. Finally, we behaviorally tested mu and delta opioid receptor knockout mice in the Novelty Suppressed Feeding test and the Forced Swim Test after chronic corticosterone and fluoxetine treatment.**Results:** Chronic fluoxetine treatment upregulates proenkephalin expression in the dentate gyrus, and this upregulation is associated with treatment responsiveness. The expression of several of the most significantly upregulated genes, including proenkephalin, is localized to an anatomically and transcriptionally specialized subgroup of mature granule cells in the dentate gyrus. We have also found that the delta opioid receptor contributes to some, but not all, of the behavioral effects of fluoxetine. **Conclusions:** These data indicate that the opioid system is involved in the antidepressant effects of fluoxetine, and this effect may be mediated through the upregulation of proenkephalin in a subpopulation of mature granule cells.

Disclosures: P. Nguyen: None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.25

Topic: G.06. Anxiety Disorders

Support: HDRF Grant RGA-13-003

Title: Contribution of Adult Hippocampal Neurogenesis to the Mechanism of Action of Electroconvulsive Stimulation

Authors: *J. CASTELLO SAVAL[1,2], V. LUNA[1,3], H. CHUNG[1,2], R. HEN[1,2];
[1]Columbia Univ., New York, NY; [2]Res. Fndn. for Mental Hygiene, Inc., New York, NY; [3]Temple Univ., Philadelphia, PA

Abstract: Major depressive disorder is the leading cause of disability worldwide and about one third of patients do not respond to current medications. In cases of treatment-resistant depression, electroconvulsive therapy (ECT) is an effective alternative with a fast onset of action. However, its mechanism of action is poorly understood. In the dentate gyrus of the hippocampus, a brain region highly vulnerable to stress, ECT increases the production of adult born granule cells (abGCs), through a process termed adult hippocampal neurogenesis. We have successfully established a mouse model of ECT, named electroconvulsive stimulation (ECS) and shown that abGCs are required for the behavioral effects of ECS. In this model, ECS stimulates the production of abGCs, increases the number of presynaptic boutons coming from abGCs in the molecular layer of the dentate gyrus and decreases c-Fos activation in the granule cell layer.In addition, optogenetic stimulation of abGCs induce an inhibitory current in mGCs mediated through mGluR2 receptors in ECS treated animals. Our overarching hypothesis is that ECS, through stimulation of neurogenesis induces an increase in the number of presynaptic boutons from abGCs which lead to the activation of postsynaptic mGluR2 receptors on mGCs. We propose that the resulting inhibition of the dentate gyrus is responsible for the antidepressant-like effects of ECS. Understanding the neurobiological substrates underlying the effects of ECT is therefore likely to reveal new targets that may lead to the development of novel fast acting antidepressants.

Disclosures: **J. Castello Saval:** None. **V. Luna:** None. **H. Chung:** None. **R. Hen:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.26

Topic: G.06. Anxiety Disorders

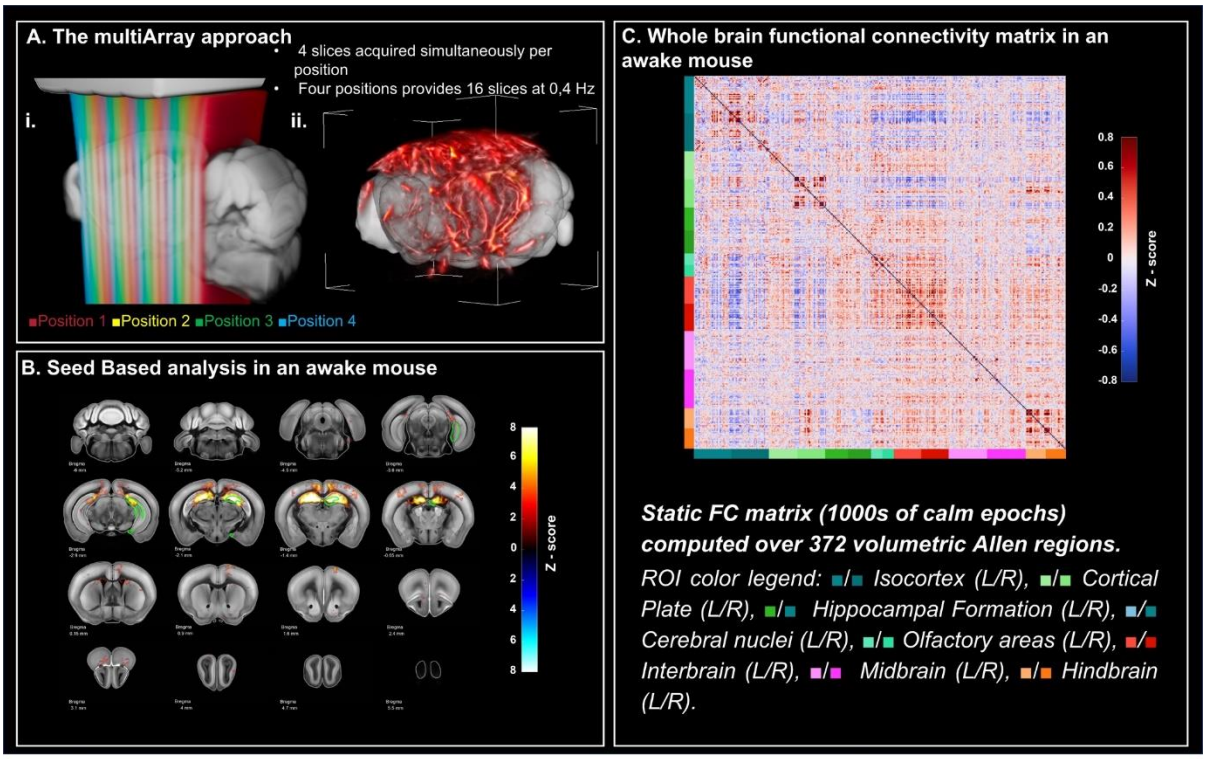
Support: Iconeus

Title: Whole-brain 3D transcranial functional connectivity of the mouse brain using a multiArray probe

Authors: *A. BERTOLO[1,2], J. FERRIER[2], S. CAZZANELLI[1,2], M. TANTER[1], B. OSMANSKI[2], M. PERNOT[1], T. DEFFIEUX[1];
[1]Physics for Med. Paris, Inserm U1273, ESPCI Paris, PSL University, CNRS FRE 2031, Paris, France; [2]ICONEUS, Paris, France

Abstract: Functional Ultrasound (fUS) recently allowed 2D or 3D transcranial functional imaging in awake mice thanks to motorized linear or matrix arrays [1]. However, the tradeoff between the field of view and temporal resolution introduced by motorized scanning prevents acquiring whole-brain resting state Functional Connectivity (rsFC) whereas the limited sensitivity of matrix arrays prevents transcranial imaging in mice. Here we propose a new hybrid approach dedicated to 3D FC in the whole brain. A 15 MHz MultiArray probe was developed

based on four high-sensitive compact linear arrays (4x64 transducers, pitch 110 µm, Iconeus, Paris, France), mounted on a 4-axis motor stage for automatic positioning and scanning both driven by the Iconeus One scanner. 3D rsFC was characterized in both anesthetized (n = 4) and head-fixed awake mice experiments (n = 4). During rsFC acquisitions (20 minutes) the power Doppler was acquired in 4 slices simultaneously during 0.4s (200 compounded frames at 500 Hz) for four positions, resulting in 16 contiguous slices (525 µm step) acquired in 2.4s. After automatic atlas-based registration and preprocessing (slice timing correction, temporal filtering, global mean regression), correlation matrices were computed with 372 Allen regions. Strong cortical and hippocampal bilateral connectivity patterns were found, and seed maps analysis allowed the quantification of long-ranged connectivity in the entire mouse brain. The MultiArray approach is a suitable solution for 3D functional imaging in awake and anesthetized mice without craniotomy. The wide field of view combined with the high sensitivity opens new opportunities to perform unbiased 3D functional connectivity measurements in the whole mouse brain.



A. The multiArray approach: i. Principle: The whole brain is covered in 16 slices, acquired at 4 positions. The resulting pressure field (simulated with field II) is overlayed to the 2-photon Allen template **ii. 3D Power Doppler angiography** registered on the Allen atlas. **B. Seed Based map** (right Dentate Gyrus) overlayed with the 2-photon Allen template after Resting State FC experiment in an awake mice reveals long ranged connectivity patterns. **C. Whole brain Resting State FC matrix in awake mice** computed after automated volumetric ROI extraction in 372 regions.

[1] A. Bertolo *et al.*, « Whole-Brain 3D Activation and Functional Connectivity Mapping in Mice using Transcranial Functional Ultrasound Imaging », *J. Vis. Exp.*, nº 168, p. 62267, févr. 2021, doi: 10.3791/62267.

Disclosures: **A. Bertolo:** A. Employment/Salary (full or part-time):; Iconeus. **J. Ferrier:** A. Employment/Salary (full or part-time):; Iconeus. **S. Cazzanelli:** A. Employment/Salary (full or

part-time):; Iconeus. **M. Tanter:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Iconeus. **B. Osmanski:** A. Employment/Salary (full or part-time):; Iconeus. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Iconeus. **M. Pernot:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Iconeus. **T. Deffieux:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Iconeus.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.27

Topic: G.06. Anxiety Disorders

Title: Exercise promotes sex-specific resilience to the effects of chronic stress

Authors: E. ELIAS, A. Y. ZHANG, A. WHITE, M. PYLE, ***M. T. MANNERS**;
St. Joseph's Univ., Philadelphia, PA

Abstract: Besides significant benefits to physical health, exercise promotes mental health, reduces symptoms of mental illness and enhance psychological development. Exercise can offset the impact of chronic stress, which is a major precursor to the development of mental disorders. The effect of exercise on chronic-stress-induced behaviors are contradictory in preclinical studies, primarily due to the lack of data and sex-specific investigations. We sought to evaluate the effects of exercise on chronic stress-induced behavioral changes in both male and female mice. Mice were subjected to an Unpredictable Chronic Mild Stress (UCMS) paradigm with accessibility to running wheels for 2 hours daily. Physiological and behavioral evaluations were conducted throughout the stress paradigm to determine if exercise blunted the effects of UCMS. Stressed mice spent more time running, with an overall increase in females compared to males. Chronic stress induced sex-specific differences in weight, and in food, water, and sucrose consumption. Exercise blunted the hyponeophagia effect of UCMS in the novelty suppressed feeding test. Results of the open field test and tail suspension test reflect a sex-specific exercise-induced amelioration in avoidance of the light zone and center zone. These results indicate that exercise concurrent with chronic stress can ameliorate some of the behavioral effects of chronic stress and baseline behavioral effects.

Disclosures: E. Elias: None. **A.Y. Zhang:** None. **A. White:** None. **M. Pyle:** None. **M.T. Manners:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.28

Topic: G.04. Emotion

Title: Exploring Natural Adaptations: Neurobiological comparisons of male and female laboratory and wild populations of Rattus norvegicus

Authors: J. JACOB[1], J. RICHARDSON[1], B. CROCKETT[1], P. LUBY[1], Y. SHATALOV[1], N. GONZALEZ[1], R. HUNTER[2], T. RICHTER[2], M. KENT[3], E. GOMEZ-SANCHEZ[4], C. GOMEZ-SANCHEZ[4], C. RIEGEL[5], T. MADERE[5], R. DENNY[5], O. HARDING[1], ***K. LAMBERT**[1]; [1]Univ. of Richmond, Richmond, VA; [2]Univ. of Massachusetts, Boston, Boston, MA; [3]Virginia Military Inst., Lexington, VA; [4]Univ. of Mississippi Med. Ctr., Jackson, MS; [5]New Orleans Mosquito, Termite and Rodent Control Board, New Orleans, LA

Abstract: Selective breeding strategies over the past century have produced laboratory rats that differ from their wild counterparts. Recently, we reported that stressed wild-trapped adult rats have higher corticosterone (CORT) fecal metabolite (FM) levels and larger adrenal glands than their weight-matched lab rat counterparts, suggesting that Hypothalamic-Pituitary-Adrenal (HPA) activation has been significantly impacted in lab rats. In the current study, comparative evaluations continued; specifically, 4 male and 11 female wild rats were trapped in Richmond, VA, and the same number of weight- and sex-matched Long-Evans lab rats (both *R. norvegicus*) were compared on several neurobiological measures. Concurrent with past findings, a 2x2 (sex x population) ANOVA indicated that stressed CORT FM levels were significantly higher in the wild cohort than the lab group [i.e., a nine-fold difference (p=.0001)] whereas dehydroepiandrosterone (DHEA), an endocrine marker of emotional resilience, levels were higher in the wild rats but not to the degree observed in the CORT data (p=.003). In an additional cohort of 10 wild adult male rats trapped in New Orleans, LA, baseline levels of CORT (in serum and fecal samples) were also observed to be higher than previously observed lab rats' baseline levels (i.e., approximately 10x higher). Focused on the brain, preliminary data suggest that the wild brains have significantly higher levels of non-specific staining of vascularization in the anterior cingulate cortex and hippocampus than the lab rat brains. Subsequent analyses using CD31-immunoreactivity, a platelet endothelial cell adhesion molecule 1 (PECAM-1) marker of vascular differentiation and angiogenesis, are ongoing. Cellular profiles in the dentate gyrus revealed no differences in the density of neurons (pyramidal and fusiform) and glial cells in lab and wild rats. Immunoreactive glucocorticoid and mineralocorticoid receptors are being quantified in the hippocampus to provide additional information about HPA activation in wild rats; additionally, levels of B2 SINE transposon RNA are being processed in several brain areas involved in emotion and cognition. Taken together, these results suggest that wild rats possess specific adaptations for enhanced vigilance and survival in the wild, including enhanced HPA responsivity and, based on preliminary data, increased markers of brain vascularization. Gaining more information about the similarities and differences between lab and wild *R. norvegicus* will inform the translational value of rodent preclinical models, especially related to stress responsivity-- a core component of many psychiatric illnesses.

Disclosures: **J. Jacob:** None. **J. Richardson:** None. **B. Crockett:** None. **P. Luby:** None. **Y. Shatalov:** None. **N. Gonzalez:** None. **R. Hunter:** None. **T. Richter:** None. **M. Kent:** None. **E. Gomez-Sanchez:** None. **C. Gomez-Sanchez:** None. **C. Riegel:** None. **T. Madere:** None. **R. Denny:** None. **O. Harding:** None. **K. Lambert:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.29

Topic: H.03. Decision Making

Support: Veterans Administration (BX003512).
NIMH (MH72672)
William and Ella Owens Foundation

Title: Ventral hippocampal input to the infralimbic cortex is necessary for the effects of extinction on set shifting after chronic stress

Authors: *D. PAREDES[1], D. A. MORILAK[2];
[1]Univ. of Texas Hlth. Sci. Ctr. San Anto, San Antonio, TX; [2]Pharmacol., UT Hlth. San Antonio, San Antonio, TX

Abstract: Psychiatric disorders such as post-traumatic stress disorder and major depressive disorder are characterized by deficits in cognitive flexibility. Exposure therapy can be effective in reversing cognitive deficits in these patients. Fear extinction in rodents bears similarity to exposure therapy. Extinction reverses chronic stress-induced deficits in cognitive flexibility on the attentional set-shifting test (AST), a medial prefrontal cortically-mediated executive process. Extinction requires the activity of pyramidal neurons in the infralimbic cortex, and BDNF-initiated signaling cascades to reverse stress-induced impairments in set shifting. However, the circuit mechanisms governing the extinction-mediated BDNF plasticity in the infralimbic are unknown. The ventral hippocampus plays a key role in regulating infralimbic activity during extinction learning, and plasticity in the ventral hippocampus is necessary for extinction memory consolidation. Thus, in these experiments we investigated the role of ventral hippocampal (vHipp) input to the infralimbic cortex (IL) for the effects of extinction after chronic stress in reversing cognitive deficits in male and female rats. Our results demonstrate that chemogenetically silencing pyramidal cell input from the vHipp to the IL prevents the effects of extinction in reversing stress-induced cognitive deficits. Further, we demonstrate that activating vHipp input in the IL, in the absence of extinction, is sufficient to reverse stress-induced deficits in set shifting. Importantly, the effects of activating vHipp terminals in the IL are dependent on BDNF signaling, as local infusion in the IL of a neutralizing antibody prevented these beneficial effects. These findings suggest vHipp-driven BDNF signaling in the IL is critical for extinction to counteract the deleterious cognitive effects of chronic stress.

Disclosures: **D. Paredes:** None. **D.A. Morilak:** None.

Poster

560. Stress and Trauma Related Disorders: Animal Models

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 560.30

Topic: I.07. Data Analysis and Statistics

Support: TriService Nursing Research Program (TSNRP)

Title: Effects of subanesthetic intravenous ketamine on brain glucose metabolism ([18F]FDG-PET) in female Sprague-Dawley rats

Authors: *S. KIM[1], C. SONG[2], S. JAISWAL[3], K. RADFORD[4], K. CHOI[5];
[1]Sch. of Medicine, Uniformed Services Univ. of Hlth. Sci., Bethesda, MD; [2]Natl. Intrepid Ctr. of Excellence, Walter Reed Natl. Military Med. Ctr., Bethesda, MD; [3]Radiology, Uniformed Services Univ. of Hlth. Sci., Bethesda, MD; [4]Uniformed Services Univ. of Hlth. Sci., Bethesda, MD; [5]Psychiatry, Uniformed Services Univ., Bethesda, MD

Abstract: Ketamine is a multi-modal anesthetic drug that is commonly used to treat pain caused by traumatic injury. However, there remain questions regarding the effects of ketamine administration on traumatic memory formation and subsequent long-term psychological outcomes, considering the drug's dissociative and hallucinogenic properties. To address these questions, this study investigated the effects of intravenous (IV) ketamine at subanesthetic doses on brain glucose metabolism in female Sprague-Dawley rats following fear conditioning. To analyze brain glucose metabolism, rats underwent two fluorodeoxyglucose (^{18F}-FDG)-positron emission tomography (PET) and computed tomography (CT) scans: (1) baseline scan 1 (Day 0, n=36), and (2) scan 2 immediately following auditory fear conditioning and ketamine infusion (Day 5, n=34). During fear conditioning, rats received three pairings of an auditory tone that co-terminated with a mild footshock (0.6 mA, 1 s) at the end of the tone. Immediately thereafter, rats received a single subanesthetic dose of IV ketamine infusion: saline (n=11), 10 mg/kg (n=11), and 20 mg/kg (n=12). FDG-PET images were pre-processed using Analysis of Functional Neuroimages (AFNI) and VivoQuant software. A voxel-wise analysis of brain glucose uptake was then performed using Statistical Parametric Mapping 12 (SPM12) and the Small Animal Molecular Imaging Toolbox (SAMIT). Our preliminary results showed that, compared to the baseline scan 1, 10 mg/kg IV ketamine at scan 2 increased glucose uptake in the left parietal association cortex, left secondary visual cortex (lateral area), and left secondary auditory cortex (dorsal area) (Family-Wise Error (FWE)-corrected $p<0.05$). Further, compared to the baseline scan 1, 20 mg/kg IV ketamine at scan 2 increased glucose uptake in the left secondary visual cortex (lateral area), left secondary auditory cortex (dorsal area), right alveus of the hippocampus, and right retrosplenial granular b cortex (FWE-corrected $p<0.05$). However, no significant changes in brain glucose uptake were observed when comparing baseline scan 1 and scan 2 of rats that received IV saline. Together, our results suggest that IV ketamine may

dose-dependently alter glucose uptake in key brain regions important for the processing and memory encoding of visuospatial, somatosensory, and auditory information. These preliminary findings warrant further investigation into the effects of ketamine infusion on brain function, memory formation, and behavior following the peri-trauma period.
Disclaimer: The views expressed are solely those of the authors and do not reflect the official policy or position of the US government.

Disclosures: S. Kim: None. **C. Song:** None. **S. Jaiswal:** None. **K. Radford:** None. **K. Choi:** None.

Poster

561. Addictive Drugs: Pharmacology and Neural Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 561.01

Topic: G.09. Drugs of Abuse and Addiction

Title: Effect of co-administration of modafinil with atomoxetine on motor activity and subsequent self-administration of the mixture of these substances in rats

Authors: *J. YEPEZ[1,2], J. JUAREZ[3];
[1]Univ. De Guadalajara, Univ. De Guadalajara, Guadalajara, Mexico; [2]Inst. de Neurociencias, Guadalajara, Mexico; [3]Univ. de Guadalajara, GUADALAJARA, Mexico

Abstract: The effects of psychostimulants involve higher availability of monoamines. While dopamine (DA) has been related to rewarding effects of drugs, noradrenergic system (NA) has been mainly related to peripheral signs. However, there is evidence that NA has an important action in brain and play an important role in the addictive effects of psychostimulants. Modafinil is a weak psychostimulant prescribed for sleep disorders that acts mainly on DA transporters and has a low affinity for NA transporters; at the same time, no addictive potential has been described for this drug. Based on the differential action on monoaminergic activity, it is possible that the co-administration of modafinil plus atomoxetine (a specific inhibitor of NA reuptake) could conferring or increase the incentive value of modafinil (MOD) and, at the same time, facilitate its subsequent self-administration of MOD and the mix MOD+ATX. Six groups of male rats 60-day old (n=12 each) were treated chronically (16 days) with, either 60mg/kg MOD, 2 and 4 mg/kg ATX or the combination of 60mg/kg MOD with 2 or 4mg/kg ATX before evaluating the motor activity and the exploratory behavior. In addition, oral MOD and MOD+ATX self-administration was assessed after 12h of liquid restriction. ANOVA test was used. Atomoxetine 4mg/kg+MOD decreases modafinil-induced exploratory behavior during pharmacological treatment. This group 60MOD+4ATX showed a higher subsequent oral consumption of modafinil that was not observed in the group pretreated with only 60MOD. Although the group 60MOD+4ATX did not present a higher subsequent oral consumption of this mixture, did present a greater exploratory behavior that was not observed during the initial treatment period. The co-administration of 60MOD+4ATX seem increase the incentive value of

modafinil without affecting motor activity; however, the increase in exploratory behavior suggest an effect, of this mixture, on behaviors more related to attentional process.

Disclosures: J. Yepez: None. **J. Juarez:** None.

Poster

561. Addictive Drugs: Pharmacology and Neural Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 561.02

Topic: G.09. Drugs of Abuse and Addiction

Title: Periadolescent toluene exposure leads to behavioral desensitization and neurochemical shifts in subsequent ethanol or cocaine challenges.

Authors: *C. J. DAVIDSON[1], S. A. PERRINE[1], S. E. BOWEN[2];
[1]Psychiatry and Behavioral Neurosciences, Wayne State Univ. Sch. of Med., [2]Psychology, Wayne State Univ., Detroit, MI

Abstract: Inhalants, such as toluene, rank fourth in lifetime use among 8th graders in the U.S. and drug use during adolescence is known to produce lasting neurobiological and behavioral consequences. Despite toluene's abuse during this period, there is a gap in understanding of the impact early adolescent toluene exposure causes on subsequent drugs of abuse. To address the gap, adolescent (PND 28-32) male Swiss-Webster mice (N=360) were exposed to 0, 2000, or 4000 parts per million (ppm) of toluene vapor for 30 min/day for 5 consecutive days using a static exposure chamber. After a delay period of 4- (PND36) or 12-days (PND44), a subset of mice (N = 180) had their locomotor activity recorded during a five injection (8 min ITI) cumulative dosing regimen of cocaine (0, 2.5, 5, 10, 20 mg/kg), ethanol (0, 0.5, 1, 2, 4 g/kg), or saline (5 control injections). Immediately following the last injection of the regimen and locomotor activity recording, animals were euthanized and brains processed for assessment of DA, NE, and 5-HT via High Pressure Liquid Chromatography (HPLC). The common metabolites DOPAC, HVA, 3-MT, & 5-HIAA were also assessed, and all HPLC analyses focused on four regions of the reward neurocircuitry, including medial prefrontal cortex (mPFC), nucleus accumbens (NAc), dorsal striatum (dSTR), and ventral tegmental area (VTA). Toluene exposure dose-dependently increased locomotor activity during the 5-day exposure. When challenged with cocaine, mice previously exposed to toluene produced significantly decreased activity after higher cocaine doses (10 and 20 mg/kg) as compared to air controls. Mice previously exposed to 4000 ppm toluene showed a non-significant downward shift in locomotor activity at 4 g/kg ethanol ($p = 0.101$). The HPLC analyses revealed subtle changes in neurochemical levels that varied by prior toluene exposure, brain region, and age tested (i.e., delay period 4 vs 12 days) with the greatest impact of prior toluene exposure observed in the dSTR. Together these results suggest that adolescent toluene exposure produces behavioral desensitization to subsequent cocaine (and possibly ethanol) exposure and that the striatum likely participates in these effects. This toluene-induced behavioral desensitization in our animal model

may be a mechanism during adolescence that impacts the propensity for continued drug use by requiring the individual to take more drugs during later drug use.

Disclosures: C.J. Davidson: None. **S.A. Perrine:** None. **S.E. Bowen:** None.

Poster

561. Addictive Drugs: Pharmacology and Neural Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 561.03

Topic: G.09. Drugs of Abuse and Addiction

Support: Friedman Brain Institute
Alkermes Grant 20-2833-00001-01
Brain and Behavior Research Foundation NARSAD 19-0612-00001-01

Title: Nos1 and NO in the interpeduncular nucleus mediate oxycodone tolerance

Authors: *K. NIBLO[1], M. JODEIRI-FASHBAF[2], Z. OKETOKOUN[2], C. FILLINGER[3], P. BALI[3], M. HEYER[3], P. J. KENNY[3], J. L. ABLES[2];
[1]Icahn Sch. of Med. At Mount Sinai Grad. Training Program In Neurosci., New York, NY; [2]Psychiatry, [3]Neurosci., Icahn Sch. of Med. At Mount Sinai, New York, NY

Abstract: Previously we demonstrated that disrupting nitric oxide synthase 1 (NOS1) in the IPN abolishes place preference for nicotine. Others have demonstrated that systemic administration of a NOS1 inhibitor can prevent the development of analgesic tolerance to opioids, reverse established tolerance, as well as prevent and reverse withdrawal to chronic opioids. We sought to determine if NOS1 in the IPN was necessary and/or sufficient for reward tolerance by knocking down NOS1 expression in the IPN using a shRNA virus (n=17). Control animals received a scrambled shRNA virus(n=20). After allowing 2-3 weeks for the virus to express, mice were divided into oxycodone-drinking and water-drinking groups. After allowing mice to habituate to their drinking condition over 1 week, a biased CPP paradigm was run conditioning mice to their least preferred side with oxycodone. As expected, the scramble injection group with regular drinking water(n=12) demonstrated robust preference for the chamber paired with 5 mg/kg oxycodone, while the scramble injection group given oxycodone (n=8) in the drinking water showed significantly less preference after chronic oxycodone, which is suggestive of reward tolerance. However, mice with NOS1 knocked down in the IPN did not show a significant difference in preference between the water (n=7) and oxycodone-drinking groups (n=10) which suggests that NOS1 in the IPN is necessary for reward tolerance to develop. Interestingly, it appears that the NOS1 shrna water group, although not statistically significant, was not able to develop as high a preference for oxycodone as the scramble group. This is suggestive that knocking down NOS1 in the IPN also reduces the rewarding aspect of acute oxycodone administration. This finding confirms previous studies showing similar behavior to other drugs of addiction such as nicotine. Understanding the mechanisms of nitric oxide production in the

IPN and the impact on drug tolerance behavior may lead to better treatment options for those struggling with opioid addiction and other substances impacted by nitric oxide and NOS1.

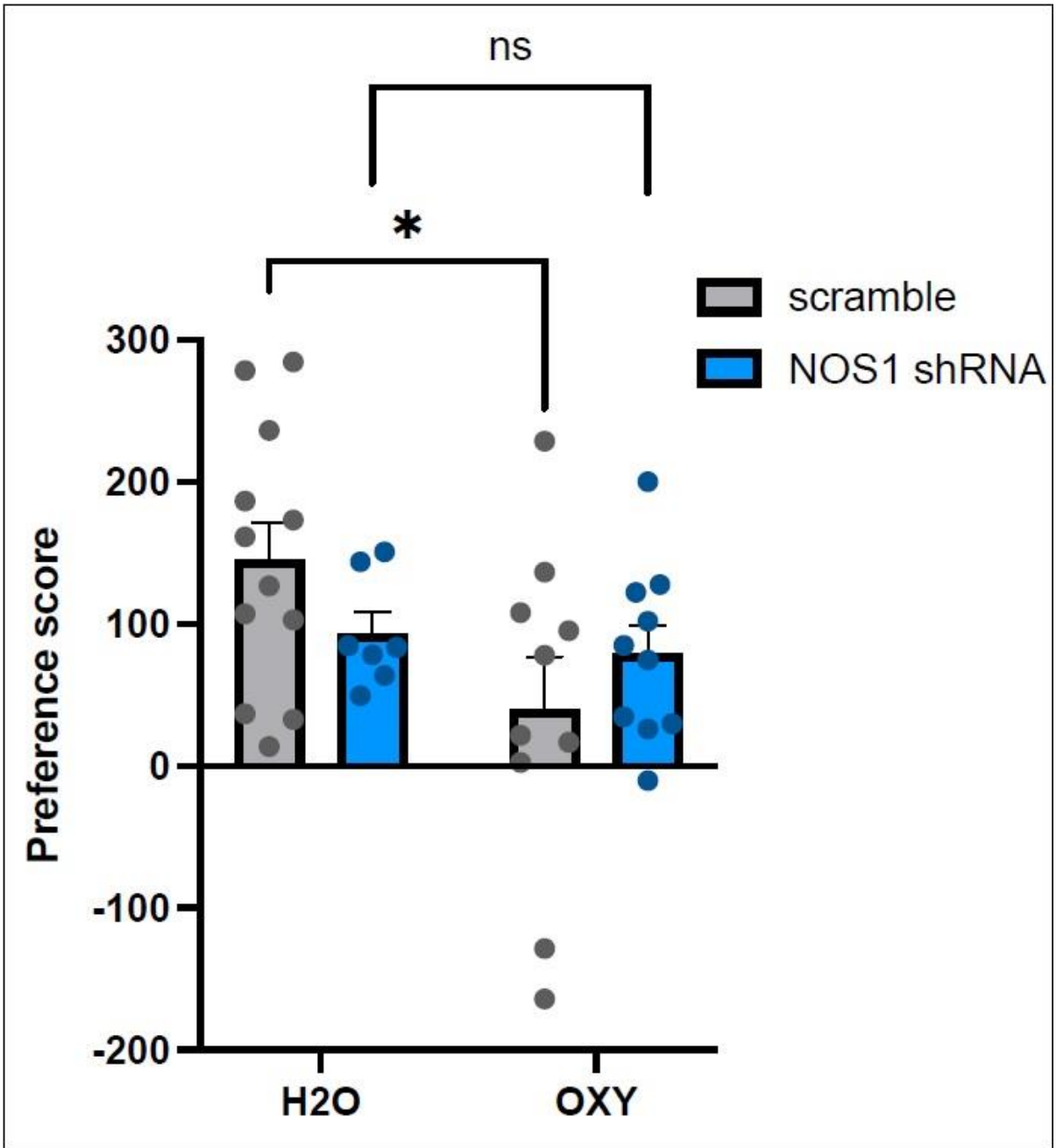


Figure 1: Oxycodone place conditioning in C57BL6J mice with *NOS1* knocked down in the IPN and chronically drinking oxycodone 0.05mg/mL (n=10) demonstrate attenuated conditioning. This is suggestive that tolerance has been ameliorated.

Disclosures: **K. Niblo:** None. **M. Jodeiri-Fashbaf:** None. **Z. Oketokoun:** None. **C. Fillinger:** None. **P. Bali:** None. **M. Heyer:** None. **P.J. Kenny:** None. **J.L. Ables:** None.

Poster

561. Addictive Drugs: Pharmacology and Neural Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 561.04

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH DA34749
NIH DA047157
NIH R00DA045795
NIH R01CA224366
NIH P01DA008227
NIH R01DA007359

Title: Transcriptional factor pCREB mediates NR2B gene expression in the PAG in morphine withdrawal in rats

Authors: H. YI[1], J. GU[1], P. HAMILTON[3], S. LIU[1], D. IKEGAMI[1], Y. KASHIWAGI[1], S. ZHU[2], K. HAYASHI[1], X. ZHU[1], D. PENG[4], G. ZHUANG[1], W. EL-RIFAI[4], R. C. LEVITT[1], E. NESTLER[3], ***S. HAO**[1];
[1]Anesthesiol., [2]Surgery, Univ. of Miami, Miami, FL; [3]Neurosci., Icahn Sch. of Med. at Mount Sinai, New York, New York, NY; [4]Surgery, Univ. of Miami,, Miami, FL

Abstract: Background: People with opioid dependence experience a physical withdrawal syndrome when tapering off opioids. Physical withdrawal is one factor that drives compulsive drug-taking behavior and short-term relapse. However, the molecular mechanisms of morphine withdrawal (MW) remain unclear. MW is characterized by persistent neuroadaptations in key brain regions, such as the midbrain periaqueductal gray (PAG). A transcription factor, phosphorylated cAMP response element binding protein (pCREB), is involved in the PAG in MW. However, the downstream mechanisms of pCREB in the PAG in MW is still not clear. Evidence strongly implicate NMDA receptor 2 B (NR2B) subunits in drug abuse. Here, we examined the molecular relationship of pCREB and NR2B in the PAG in MW using rats. **Methods**: Chronic escalating doses (10-50 mg/kg) of morphine given intraperitoneally for a period of 5 days in male rats. MW syndrome was precipitated by naloxone (4 mg/kg, IP) 1 h after the last morphine injection (day 5). Immediately after naloxone, withdrawal signs were evaluated for 30 minutes. Following MW, western blot was used to determine levels of pCREB and NR2B proteins in the ventrolateral PAG. For intracranial microinjection into the PAG, cannula implantation was carried out in a stereotaxic headholder. An antisense oligodeoxynucleotide against CREB (AS-CREB), or a NR2B inhibitor, was microinjected in MW rats. Chromatin immunoprecipitation (ChIP) assay was used to determine the *nr2b* gene promoter enrichment of pCREB. Over-expression of CREB was induced by microinjection of a recombinant herpes simplex virus (HSV) vector that encodes the CREB gene. **Results**: Chronic morphine withdrawal was precipitated by naloxone, with increased MW scores seen in rats. MW increased the expression of pCREB and NR2B protein in the PAG using western blots. Immunostaining showed co-localization of pCREB and NR2B in PAG neurons. Microinjection of AS-CREB, or of the NR2B inhibitor Ro25-6981, into the PAG blunted the MW syndrome.

Western blots showed that AS-CREB reduced the expression of NR2B in the PAG in MW. ChIP assay showed that AS-CREB reduced the enrichment of pCREB on the *nr2b* gene promoter region in the PAG in MW. Over-expression of CREB mediated by HSV vectors further increased MW scores and expression of NR2B protein in the PAG. **Conclusions**: Our results suggest that MW is associated with pCREB and induction of NR2B in PAG neurons, and that CREB mediates increased expression of NR2B in an epigenetic manner. Understanding the complex relationship between pCREB and NR2B will allow us to formulate novel epigenetic and molecular therapies.

Disclosures: **H. Yi:** None. **J. Gu:** None. **P. Hamilton:** None. **S. Liu:** None. **D. Ikegami:** None. **Y. Kashiwagi:** None. **S. Zhu:** None. **K. Hayashi:** None. **X. Zhu:** None. **D. Peng:** None. **G. Zhuang:** None. **W. El-Rifai:** None. **R.C. Levitt:** None. **E. Nestler:** None. **S. Hao:** None.

Poster

561. Addictive Drugs: Pharmacology and Neural Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 561.05

Topic: G.09. Drugs of Abuse and Addiction

Support: UG3 DA050322

Title: Preclinical evaluation of the NMDAR positive allosteric modulator NYX-783 for opioid use disorder

Authors: *R. TRINKO[1], E. FOSCUE[1], D. DIAZ[1], K. LEADERBRAND[2], J. R. TAYLOR[1], R. J. DILEONE[1];
[1]Yale Univ., New Haven, CT; [2]Aptinyx Inc, Evanston, IL

Abstract: Rates of opioid use disorder (OUD) and overdose deaths have increased dramatically in recent years. Currently approved medications for OUD include the opioid agonists methadone and buprenorphine, and the opioid antagonist naltrexone. However, relapse rates are high due to an impaired ability for addicts to control their urge to consume due to strong cravings, and the extreme severity of withdrawal symptoms. Additionally, the opiate nature of the agonists can lead to abuse of those compounds as well. Non-opiate targets, such as glutamate receptors, are potentially ideal for developing intervention strategies with the goal of reducing OUD, relapse, and overdose death. NYX-783 is a small molecule positive modulator for the glutamate receptor NMDAR. It has been shown to modulate learning and memory, both of which are impaired in drug addicts, and known play a role in relapse. Using mice, we have conducted preclinical studies to evaluate the potential for NYX-783 as a therapeutic for OUD with assessment of several outcomes: 1) respiratory depression, 2) consumption, 3) motivation for consumption, and 4) the development of aversion to withdrawal symptoms. For respiratory depression studies, mice were pretreated 1h prior with NYX-783 and respiratory rates were monitored for 15min

after each escalating dose of oxycodone. No effects were seen across all doses of NYX-783 tested. To test for effects on drug consumption, mice were trained to orally self-administer oxycodone and then treated every two days with different doses of NYX-783. No effect on intake at clinically-relevant doses was observed. Mice that were self-administering oxycodone were then evaluated on a progressive ratio paradigm and no effect of NYX-783 was observed on oxycodone rewards earned. We next evaluated withdrawal using two separate paradigms to test for effects of NYX-783 on, 1) somatic withdrawal symptoms, 2) aversion to the state of withdrawal. For somatic withdrawal, using higher doses of naloxone (1 mg/kg), NYX-783 did not attenuate jumping behavior. For aversion to withdrawal, three aversion pairings were completed that consisted of oxycodone treatment, followed by NYX-783 preceding a low dose of naloxone (0.1mg/kg ip) immediately before pairing in a specific context. These alternated with neutral (saline) pairings daily in an alternate context, resulting in place aversion (avoidance of oxycodone/naloxone paired side) on test day. We observed a significant reduction in place aversion in the NYX-783 treated mice. These data suggest a potential therapeutic use for NYX-783 in reducing the negative state of withdrawal that can drive relapse in OUD.

Disclosures: **R. Trinko:** None. **E. Foscue:** None. **D. Diaz:** None. **K. Leaderbrand:** A. Employment/Salary (full or part-time):; Aptinyx Inc. **J.R. Taylor:** None. **R.J. DiLeone:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Aptinyx Inc..

Poster

561. Addictive Drugs: Pharmacology and Neural Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 561.06

Topic: G.09. Drugs of Abuse and Addiction

Title: Animal models of addiction - Aiding the search for pre-clinical CNS drug discovery

Authors: Author Block needs to be regenerated

Abstract: Animal models of addiction have been crucial to our understanding of brain mechanisms that underlie reward functions and the dysregulation of these processes during addiction. Clinical features have been incorporated into these models to better capture the human condition, including the compulsive aspects of drug seeking and taking, as well as propensity to relapse. Here, we present three addiction-related behavioral assays that are routinely utilized for testing the abuse potential of novel therapeutics. Alternatively, these assays can also be implemented to assess efficacy of potential pharmacotherapies for addiction. Locomotor sensitization, an augmentation of locomotory response following exposure to drugs of abuse like cocaine and amphetamine, is mediated by molecular and cellular mechanisms that contribute to drug-induced pathophysiological emotional and motivational states characteristic of addiction. Conditioned place preference (CPP) paradigm is a well-established model utilized to test the reinforcing properties of drugs. A drug's rewarding or aversive properties are measured based on

associations formed between the administered drug and a contextual environment. Finally, the drug self-administration paradigm relies on operant conditioning and involves the subject to perform a response, such as pressing a lever, to receive a dose of a drug. Abuse potential or rewarding properties of the drug is indicated by its ability to support continued responding by the subject. In addition to their fundamental role in abuse liability testing, which forms an integral part of drug development, regulatory review, and market approval, these assays can also be used for testing efficacy of new medications being developed for the treatment of substance use disorders.

Disclosures: **U. Datta:** None. **M. Dudek:** None. **A. Sharma:** None. **J. Korkalainen:** None. **T. Huuhtanen:** None. **S. Bäck:** None. **J. Rytkönen:** None. **D. Miszczuk:** None.

Poster

561. Addictive Drugs: Pharmacology and Neural Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 561.07

Topic: G.09. Drugs of Abuse and Addiction

Support: CIC-UMSNH 26.10
CIC-UMSNH 2.36
CIC-UMSNH 30.2

Title: Toluene exposure modifies adrenergic responses in aorta in rats

Authors: A. CEJA-FERNANDEZ[1], D. GODINEZ-HERNANDEZ[2], L. F. ORTEGA-VARELA[3], *M. Y. GAUTHEREAU-TORRES[1];
[1]Facultad de Ciencias Medicas y Biologicas "Dr. Ignacio Chavez", [2]Inst. de Investigaciones Quimico Biologicas, [3]Facultad de Salud Publica y Enfermeria, Univ. Michoacana De San Nicolas De Hidalgo, Morelia, Mexico

Abstract: Solvent misuse constitutes a serious public health problem worldwide that mainly affects children and adolescents. Volatile solvents share pharmacological properties with central nervous system depressants, and they are commonly inhaled to achieve intoxicating states. The most misused solvent is toluene. It can be found as part of multiple products such as thinner, adhesives, gasoline, and cleaning products. There are several reports indicating that inhaling toluene can produce cardiac arrhythmias due to adrenergic sensitization of the heart and also lead to a phenomenon named sudden sniffing death, but the mechanism responsible is not completely understood. In this sense, it is unknown the effect of toluene exposure on adrenergic responses in arteries, such as aorta. The purpose of this study was to investigate the effect of subacute toluene exposure on alpha 1- and beta-adrenergic responses in aorta in rats. Male Wistar rats (250-300 g) were placed in a static exposure chamber and exposed to 6000 ppm of toluene or air (control group) during 30 minutes, twice a day, for a week. After this period, rats were euthanized with sodium pentobarbital (65 mg/kg) and the aorta was isolated, it was cut into rings and the

endothelium was removed from half of them. Arterial rings were bathed in a 10 ml chamber, filled with Krebs-Henseleit solution, and attached to the bottom of the chamber and to an isometric force displacement transducer. Aortic rings were subjected to an initial optimal tension of 3 g and were stimulated with a submaximal concentration of phenylephrine (1x10-7 M). Concentration-response curves to phenylephrine (alpha-1 adrenergic agonist: 1x10-9 - 1x10-5 M), and isoprenaline (beta adrenergic agonist: 1x10-9 - 1x10-5 M) were constructed. Our results showed that phenylephrine produced a contraction in a concentration-dependent manner both in control and toluene groups, and a diminished response was produced in aortic rings with endothelium compared with endothelium-denuded rings. On the other hand, in endothelium-denuded rings toluene evoked a lower response to phenylephrine compared to control group. In contrast, there were no significant differences between toluene and control groups when isoprenaline stimulation was made. In conclusion, our findings suggest that toluene modifies alpha adrenergic responses probably due to an endothelium-dependent mechanism.

Disclosures: A. Ceja-Fernandez: None. **D. Godinez-Hernandez:** None. **L.F. Ortega-Varela:** None. **M.Y. Gauthereau-Torres:** None.

Poster

561. Addictive Drugs: Pharmacology and Neural Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 561.08

Topic: G.09. Drugs of Abuse and Addiction

Support: DA044451
DA043799

Title: Identification of unique microbiome and metabolome profiles associated with vulnerability to substance use disorder risk in a population of heterogenous outbred stock of rats using machine learning.

Authors: *S. SIMPSON[1], A. AARON[2,4], R. GABRIEL[3], M. BRENNAN[1], L. MATURIN[1], G. DE GUGLIELMO[1], O. GEORGE[1];
[1]Psychiatry, [2]Collaborative Mass Spectrometry Innovation Ctr., [3]Anesthesiol. and Perioperative Bioinformatics, Univ. of California San Diego, La Jolla, CA; [4]Chem. and Biochem., Univ. of Colorado Denver, Denver, CO

Abstract: The increase in substance use in the last decade represents a significant major public health problem with a mounting need for a precision approach to the treatment and diagnosis of substance use disorders. Prediction of potential substance abuse prior to exposure remains an avenue of great interest. The identification of novel pathways and biomarkers are necessary to explore potential innovative treatments. The gut-brain axis has been implicated in substance use disorders, is metabolically active, and interacts with the whole body throughout the life of the host. While it has been hypothesized that the microbiome and metabolome may improve

diagnosis or identify individuals vulnerable to escalation of substance use, the tools are still being developed to address this question. The highly dimensional data generated from metagenomic sequencing and untargeted metabolomics is a challenge to analyze and integrate. To address this issue, we have applied modern machine learning and artificial intelligence approaches combined with class balancers and explainer models to reveal actionable features for future study. Shotgun metagenomics and untargeted metabolomics were carried out on fecal and plasma samples from the heterogenous stock rats (HS) that undergo self-administration of cocaine (n=50) or oxycodone (n=50). The rats were characterized for their addiction-like behaviors through a wide range of tests as well as during short/long access drug self-administration. Following the experiment, organs of interest, feces, and plasma were stored in the Cocaine (cocainebiobank.org) and Oxycodone biobanks (oxycodonebiobank.org). Machine learning approaches were implemented to analyze microbiome and metabolomic signatures of animals before and after exposure to either oxycodone or cocaine self-administration. Animals vulnerable to escalation were found to exhibit unique microbiome and metabolomic profiles from those that did not escalate intake during long-access drug self-administration. The HS rats exhibit heterogeneity in intake, leading to class imbalance issues (SMOTE) and the need for explainer models (SHAP) to identify features of interest. When applied, metabolic pathways related to inflammation, gene expression modification, and gut-brain axis signaling were identified as potential biomarkers of interest. Upon supplementation of microbial metabolites in the oxycodone group, the vulnerable animals decreased their drug intake. These data demonstrate that microbial metabolic activity and functional read-outs such as metabolomics are key in developing novel diagnostics and therapeutics for substance use disorders.

Disclosures: **S. Simpson:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); CEO BrilliantBiome, Inc.. **A. Aaron:** None. **R. Gabriel:** None. **M. Brennan:** None. **L. Maturin:** None. **G. de Guglielmo:** None. **O. George:** None.

Poster

561. Addictive Drugs: Pharmacology and Neural Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 561.09

Title: WITHDRAWN

Authors: *E. RODRIGUEZ[1], T. M. RODRIGUEZ[1,3], H. K. NAMBALLA[2], K. GRONDECKI[3], M. AGUILAR[1], B. YOO[2], W. W. HARDING[2], P. A. SERRANO[1,3]; [1]Psychology Dept., [2]Chem. Dept., Hunter College, CUNY, New York, NY; [3]Grad. Center, CUNY, New York, NY

Abstract: Effective pharmacological agents for the treatment of methamphetamine use disorder (MUD) have not been developed. The chronic use of methamphetamine (MA) alters dopamine (DA) levels, resulting in reward- and drug-seeking behaviors. Polypharmacological agents that simultaneously target DA D1, D2 and D3 receptors may be useful to remediate MUD. Ideally,

for such an agent to be therapeutically useful, it should be blood-brain barrier (BBB) penetrable upon oral administration. (-)-Stepholidine (SPD), is a tetrahydroprotoberberine alkaloid which acts as a DA D1 receptor partial agonist /D2 antagonist /D3 antagonist with anti-addiction and memory enhancing properties. Although the pharmacodynamic profile of SPD is promising as a potential MUD therapeutic, it suffers from poor oral bioavailability, limiting its therapeutic utility. Therefore, we sought to investigate SPD derivatives (potential prodrugs with predictably improved oral bioavailability and ability to deliver SPD to the brain), following voluntary oral administration. A mouse model of VOA was used to deliver the drugs – SPD, tetrahydropalmatine (THP) and stepholidine diacetate (SPDD). Liquid chromatography-tandem mass spectrometry (UPLC- MS/MS) was used to evaluate the permeability and concentration of the drugs in C57Bl/6J male mice brain. A single dose of SPD (10 mg/kg) was administered, and brains assessed at 15- and 30-min post VOA. For THP, a single 20 mg/kg dose was administered, and brains assessed at 15, 30, 60, and 120 min post VOA. We successfully detected THP at all time points in brain; however, there was no drug detection for SPD. Following a single oral dose of SPDD (20 mg/kg), we detected stepholidine 15 min post VOA in brain. These results indicate that SPDD improved the oral bioavailability and BBB permeability of SPD and similar investigations of other potential SPD prodrugs are warranted. Further evaluation of SPDD as an anti-MA addiction agent using a previously validated model of voluntary oral methamphetamine administration (VOMA) as well as characterization of underlying changes in DA receptor expression, are planned.

Disclosures: E. Rodriguez: None. **T.M. Rodriguez:** None. **H.K. Namballa:** None. **K. Grondecki:** None. **M. Aguilar:** None. **B. Yoo:** None. **W.W. Harding:** None. **P.A. Serrano:** None.

Poster

561. Addictive Drugs: Pharmacology and Neural Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 561.10

Topic: G.09. Drugs of Abuse and Addiction

Title: Neuroprotective role of magnesium sulfate in NMDA receptor-mediated excitotoxicity in the lateral habenula of ethanol dependent mice

Authors: *S. G. GAKARE, P. P. PATNI, R. R. UGALE;
Dept. of Pharm Sciences,, Nagpur, India

Abstract: Alcohol is the most commonly abused drug worldwide; it causes dependence and shows withdrawal symptoms upon cessation. Lateral habenula (LHb), rich in glutamatergic neurons, plays a significant role in alcohol addiction. Neurotoxicity following ethanol withdrawal has been shown to occur via NMDA receptors, which show a binding site for magnesium (Mg^{2+}) ions that reversibly block the passage of Ca^{2+}. Mg^{2+} deficiency is involved in alcohol dependence and withdrawal, while Mg^{2+} treatment decreases alcohol withdrawal

syndrome. The present study aimed to investigate the neuroprotective effect of magnesium on excitotoxic neurodegeneration induced by ethanol withdrawal in LHb. Adult Swiss albino mice were stereotaxically implanted with the cannula into LHb and were subjected to intragastric chronic binge ethanol administration (4g/kg, 20% w/v) for ten days; post-24 hr withdrawal behavioral testing was carried out. We found the systemic and intra-LHb treatment of magnesium sulfate ($MgSO_4$) decreased somatic withdrawal signs and reduced hyperlocomotion, grooming, rearing, and wall climbing compared to the vehicle-treated group. Further, in the EPM and LDB test, $MgSO_4$ treatment increased the number of entries and time spent in the open arm and light chamber, respectively, indicating an anti-anxiety effect. Furthermore, systemic and intra-LHb $MgSO_4$ treatment increased the NeuN positive cells and significantly decreased Caspase-3 positive cells compared to the vehicle-treated group. Thus, the present study suggests systemic and intra-LHb administration of $MgSO_4$ reduces ethanol withdrawal symptoms and associated excitotoxic degenerative changes in LHb in mice. Therefore, we conclude that $MgSO_4$ serves as an agent to prevent ethanol withdrawal and associated neurodegeneration in LHb.

Disclosures: S.G. Gakare: None. **P.P. Patni:** None. **R.R. Ugale:** None.

Poster

561. Addictive Drugs: Pharmacology and Neural Mechanisms

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 561.11

Topic: G.09. Drugs of Abuse and Addiction

Title: The co-presence of mind-altering substances in Mitragynine as a cause of death

Authors: *A. SURIAGA;
Epidemiology, Harvard Univ., Boston, MA

Abstract: During the initial year of the COVID-19 pandemic, deaths from mind-altering substances such as opioids and stimulants have increased substantially. However, deaths from Mitragynine received less attention. There is a dearth of research about the characteristics of people who died from Mitragynine, and its effect on the neurocognitive functioning. Also, research on the safety and efficacy of Mitragynine in its perceived use as a partial opioid agonist and its effect on cognitive functioning is limited. In this study, we described the characteristics of people with Mitragynine as a cause of death (COD) in Florida in 2020 using de-identified data from Florida Department of Law Enforcement in 2020. Mitragynine is the primary alkaloid of the plant Mitragyna speciosa, known as kratom, indigenous to Southeast Asian countries, such as the Philippines, Thailand, and Malaysia. While the neurobiology of Mitragynine seems unclear, particularly on how it affects the brain leading to a person's death, our study results indicated that most of the decedents who died from Mitragynine as a COD were not of Asian descent. Mitragynine was determined as a COD by the medical examiners when it played a causal role in the death of a person through autopsy, urine, and toxicology results. We use descriptive statistics to retrospectively describe these decedents. The co-presence of other mind-altering substances,

such as opioids (Fentanyl, Morphine, and Oxycodone) and stimulants (methamphetamine and amphetamine), cannabinoids, and ethanol was also examined. Results: A total of 166 people died from Mitragynine as a COD; age range (21 to 68 years), mean age of 37.28 (SD=8.884). More males were affected at 80.1%. 160 of 166 decedents were non-Hispanic whites (96.4%), Asian (n=1), Black (n=1), and Hispanic (n=3). Most decedents died in accidents (99.4%), with drug intoxications as main reason. The co-presence of mind-altering substances are as follows: opioids- fentanyl (n=129), morphine (n=21), and oxycodone (n=14); stimulants- amphetamine (n=14) and methamphetamine (n=20), ethanol (n=47), and cannabinoids (n=115). Only three decedents had Mitragynine as the sole substance found during autopsy and toxicology results. Results of this study informed our efforts to conduct an exploratory study on perceived Mitragynine use for pain, and its measured effects on neurocognitive functioning. Mitragynine-related death is indeed a legitimate health concern. As most people died from accidents while under the influence of Mitragynine, the public is warned about the harmful effects of using Mitragynine to reduce avoidable deaths.

Disclosures: A. Suriaga: None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.01

Topic: G.09. Drugs of Abuse and Addiction

Support: DA047858 (to AGH)
Indiana Addiction Grand Challenge Grant (to AGH)
DA041229 (to AM)
DA041229 (to LB, AGH, and AM)

Title: Synthetic CB1 agonists induce respiratory depression in awake adult mice - a potential role for beta arrestin

Authors: *J. WATKINS[1], V. IYER[1], C. ZAVALA[1], W. O'CONNOR[1], C. I. TSOUTSOUVAS[2], A. MAKRIYANNIS[2], C. JOHNSTON[3], L. BOHN[3], A. HOHMANN[1]; [1]Indiana Univ., Bloomington, IN; [2]Northeastern Univ., Boston, MA; [3]Univ. of Florida Scripps Biomed. Res., Jupiter, FL

Abstract: Clinical literature indicates that illicit synthetic cannabinoids, known as spice compounds, can induce acute respiratory depression requiring emergency medical care in humans. These compounds typically target cannabinoid receptor type 1 (CB1) and are used recreationally to mimic the psychoactive effects of cannabis, casting doubt on previously held conventions that CB1 agonists do not detrimentally affect respiration. The apparent insensitivity of these synthetic cannabinoids to traditional first line treatments for drug-induced respiratory depression (e.g. Naloxone) calls into question how cannabinoids interact with the respiratory

system and highlights the need for effective therapeutics. Our lab previously reported that LY2828360, a G protein-biased cannabinoid receptor type 2 (CB2) agonist, lacked intrinsic effects on respiratory parameters and attenuated fentanyl-induced respiratory depression. Here we examined the impact of two synthetic CB1 agonists, AM12059 and AM11250, on the respiratory parameters of awake mice. Whole-body plethysmography was used to assess each ligand's impact on respiratory minute volume, tidal volume, and frequency. Cannabimimetic effects and tolerance were evaluated via tetrad testing after once daily dosing. Both ligands induced respiratory depression at strikingly low doses in wild type mice, affecting both respiratory frequency and average tidal volume. Respiratory depression elicited by both ligands was fully blocked by the CB1 antagonist/inverse agonist AM251 and was entirely absent in CB1 knockout mice. Repeated dosing significantly attenuated the magnitude of respiratory effects but did not eliminate them. Respiratory depression induced by both ligands was unaffected by coadministration of the CB2 antagonist AM630, or the peripherally restricted CB1 antagonist AM6545. Furthermore, AM11250 and AM12059 differ markedly in efficacies for recruiting beta-arrestin, producing suprathreshold and minimal recruitment of beta-arrestin-2 while showing similar efficacies and potencies for inhibiting adenylyl cyclase in vitro. AM12059 elicited more extreme hypothermia, enhanced catalepsy, greater antinociception, and stronger respiratory depression than AM11250. These results suggest that CB1 receptors are expressed in CNS neurons that are responsible for the regulation of respiratory function. The susceptibility of these neurons to exogenous CB1 agonists may explain clinical observations of respiratory depression after the administration of synthetic cannabinoid drugs.

Disclosures: J. Watkins: None. **V. Iyer:** None. **C. Zavala:** None. **W. O'Connor:** None. **C.I. Tsoutsouvas:** None. **A. Makriyannis:** None. **C. Johnston:** None. **L. Bohn:** None. **A. Hohmann:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.02

Topic: G.09. Drugs of Abuse and Addiction

Support: INSERM U121519G grant
University of Bordeaux
CNRS
French MESRI
Aelis Farma (Bordeaux, France)

Title: Disruption of pregnenolone allosteric binding site on the type-1 cannabinoid receptor CB_1 triggers hypersensitivity to cannabinoids in a new mutant mouse line

Authors: *P.-L. RAUX[1], V. LALANNE[1], L. BELLOCCHIO[1], A. CATHALA[1], G. DRUTEL[1], V. ROULLOT-LACARRIÈRE[1], A. GREL[1], I. MATIAS[1], P.-V. PIAZZA[2], G. MARSICANO[1],

J.-M. REVEST[1], M. VALLÉE[1];
[1]INSERM U1215, INSERM U1215, BORDEAUX CEDEX, France; [2]Aelis Farma, Bordeaux, France

Abstract: The type-1 cannabinoid receptor (CB_1) participates in the regulation of a myriad of biological functions to help maintain homeostasis. Because CB_1 occupies such a key position in the central nervous system, the brain has set up endogenous regulatory mechanisms to prevent an imbalance in CB_1 activity that may lead to the onset of neuropsychiatric disorders. We have shown that pregnenolone (PREG), formerly considered an inactive precursor of steroid hormones, is a potent CB_1 negative allosteric modulator[1] and prevents against excessive CB_1 activation and toxic effects by Δ^9-tetrahydrocannabinol (THC) [1, 2], the main psychoactive ingredient of the plant *Cannabis sativa*. No study, however, has so far determined if this regulation loop is set up endogenously and participates in the modulation of CB_1-mediated physiological functions. To investigate this topic, we have developed a novel mutant mouse line (CB_1-E134G), carrying a missense mutation in the CB_1-encoding gene (*Cnr1*) aimed at disrupting the PREG-CB_1 allosteric binding site. We investigated the effects of cannabinoids in male and female CB_1-E134G mice (aged 3 to 4 months) to validate and identify the impact of the loss of PREG binding to CB_1 in our model. We show that CB_1-E134G mice exhibit increased behavioral and somatic efficiency of THC compared to wild-type (WT) animals. By addressing THC-induced PREG synthesis by gas chromatography-mass spectrometry tandem, we show that THC-induced PREG increase is similar between genotypes, indicating that higher efficacy of THC in CB_1-E134G mice is not related to an alteration in PREG brain levels. In addition, we performed *ex vivo* analyses on mitochondrial extracts to study the effect of the synthetic CB_1 agonist WIN-55,212-2 (WIN) on cellular respiration. A higher response to WIN is observed in samples collected from CB_1-E134G mice compared to those of WT mice, and this response is inhibited by PREG treatment in WT samples only. Our data strongly suggest that PREG cannot bind to CB_1 in our mouse model, leading to hypersensitivity to cannabinoids. This model provides a significant strategic opportunity to elucidate the role of the feedback loop between PREG and CB_1 *in vivo* and may point towards new therapeutic potentials for PREG in CB_1-related neuropsychiatric disorders.
References:[1]Vallée et al. 2014 ; [2]Busquet-Garcia et al. 2017

Disclosures: **P. Raux:** None. **V. Lalanne:** None. **L. Bellocchio:** None. **A. Cathala:** None. **G. Drutel:** None. **V. Roullot-Lacarrière:** None. **A. Grel:** None. **I. Matias:** None. **P. Piazza:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Aelis Farma. **G. Marsicano:** None. **J. Revest:** None. **M. Vallée:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.03

Topic: G.09. Drugs of Abuse and Addiction

Title: The effects of cannabinoid agonism on auditory discrimination

Authors: D. NYKANEN[1], H. STIFFLER[2], M. BAY[1], C. GOLDIE[1], ***N. SWALVE**[1];
[1]Alma Col., [2]Alma Col., Alma, MI

Abstract: Cannabis use alters audition in a variety of ways. In humans, there is an increased appreciation of musical stimuli, impaired auditory signal detection and filtering of irrelevant stimuli during cannabis intoxication. Disruptions in sensorimotor gating are also seen in both humans and mice. The mechanisms behind the alterations in both basic auditory processing such as sensorimotor gating/processing and more complex processes like musical appreciation are currently unknown. The goal of the current study was to examine the effects of a selective cannabinoid agonist on the CB1 receptor (WIN 55,212-2) on auditory discrimination. Twenty-four Sprague Dawley rats were tested in an auditory go/no-go task using two different frequency tones. After training, rats were assigned to one of three groups: low dose WIN (1.2 mg/kg), high dose WIN (3 mg/kg) or control. Rats were tested for number of errors made and overall responding after drug exposure. WIN dose-dependently altered responding in the go/no-go task with little to no effect on activity. This study furthered the literature on how the CB1 receptor plays a role in basic auditory processes related to auditory learning. This information can then be used to further examine the various cannabinoids in cannabis to examine the short- and long-term impact of cannabis on audition in rodents and humans.

Disclosures: D. Nykanen: None. **H. Stiffler:** None. **M. Bay:** None. **C. Goldie:** None. **N. Swalve:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.04

Topic: G.09. Drugs of Abuse and Addiction

Support: ETSU Internal Funds, RDC Major

Title: Reinforcement enhancing effects of Δ-9 tetrahydrocannabinol (THC) in male and female rats.

Authors: K. B. WALSTON, C. D. R. JOHNSON, A. F. RADFORD, B. SCHMEICHEL, *M. I. PALMATIER;
East Tennessee State Univ., Johnson City, TN

Abstract: Rationale. Cannabis is widely consumed by humans for pharmacological effects that are mediated by THC. However, THC supports low rates of behavior in non-humans. We hypothesized that THC may have reinforcement enhancing effects comparable to other drugs (e.g., nicotine and caffeine) which are also widely consumed by humans, but difficult to establish as primary reinforcers in non-human animals. Objective. We sought to determine whether THC

is a reinforcement enhancer, in male (M) and female (F) rats in three experiments. Method. In Experiment 1, rats were shaped to lever press for a reinforcing saccharin solution (0.2% w/v) in standard operant chambers equipped with infrared beams to monitor locomotor activity. In Experiment 2, rats were shaped to press a nose-key for 0.2% saccharin - the nose key was used to reduce the likelihood that size differences influenced the amount of effort required to earn saccharin. In Experiments 1 and 2 rats were tested under a progressive ratio (PR) schedule of reinforcement. In Experiment 3 rats were shaped to enter a receptacle for a visual stimulus (VS; 1 min extinction of houselights). Results. In Experiment 1 there was a significant sex difference for active lever responses and reinforcers earned ($F<M$, $p<0.05$) but not activity. Pretreatment with 1.5 and 3 mg/kg THC significantly reduced active lever presses, reinforcers earned, and activity in both M and F rats ($p<0.05$). Pretreatment with 0.75, 0.5, and 0.35 mg/kg THC systematically increased active lever responding (but not activity), and saccharin reinforcers earned in M, but not F rats, relative to baseline (0 mg/kg). In Experiment 2 using the nose-key operant response eliminated sex differences in responding for saccharin reinforcement, there were no differences in active responses or reinforcers earned ($p>0.05$). Also, 0.5 mg/kg THC increased responding for saccharin in both M and F rats ($p<0.05$). In Experiment 3 there were no sex differences in responding for VS, THC pretreatments are currently being tested for their effects on responding for the VS. Conclusions. THC enhances the motivation for saccharin in M and F rats. The putative sex differences observed in Experiment 1 may be more accurately described as size differences, as using a response that was agnostic to rat size eliminated the sex difference in Experiment 2. Finally, the reinforcement enhancing effects of THC were observed at lower doses than anticipated based on prior research. These findings should also inform the range of doses and procedures (e.g., inclusion of visual reinforcers) that may facilitate future investigation of THC self-administration.

Disclosures: K.B. Walston: None. **C.D.R. Johnson:** None. **A.F. Radford:** None. **B. Schmeichel:** None. **M.I. Palmatier:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.05

Topic: G.09. Drugs of Abuse and Addiction

Support: CIHR Grant

Title: The effect of prenatal THC and CBD exposure on behaviour in the adult rodent

Authors: *M. DRUPALS[1,2], J. HICKS[1], B. E. STEINBERG[1,2], M. W. SALTER[1,2];
[1]Hosp. for Sick Children, Toronto, ON, Canada; [2]Physiol., Univ. of Toronto, Toronto, ON, Canada

Abstract: Existing human and rodent studies have found some convergent and some contradictory findings on the impact of prenatal cannabinoid exposure on offspring. In children and adolescents, behavioural effects which have been reported include a variety of cognitive changes (reviewed in *McLemore and Richardson*, 2016). Using a mouse model developed to capture pre- and peri-gestational THC and CBD exposure, we examined the effects on a range of behaviours in adult offspring. Beginning 7 days prior to mating and throughout gestation until E17.5, pregnant mice were subcutaneously injected once daily with either 5mg/kg THC, 60mg/kg CBD, or vehicle (1:4:15 95% EtOH:Tween80:saline). Starting at 9 weeks of age, male and female mice underwent a battery of behavioural tests which included accelerod (motor), elevated plus maze (anxiety), and open field (locomotion and anxiety). We found no significant differences in the behaviour of THC or CBD-exposed mice in the open field test compared to vehicle controls. Conversely, THC-exposed offspring spent more time in the open arms of the elevated plus maze compared to controls ($p<0.05$, ANOVA with Post-Hoc Tukey's multiple comparisons test). We found no differences in performance on the accelerod test between groups across all three experimental trials. These data suggest that the exposure of CBD in this model had no effect on the behaviours evaluated in adulthood. In the elevated plus maze specifically, offspring of THC-treated dams showed behaviour consistent with a reduction in anxiety. However, this was not recapitulated in the open field test. To investigate the effect of this gestational drug exposure on cognition, a novel object recognition assay is currently underway, with final results pending.

Disclosures: M. Drupals: None. **J. Hicks:** None. **B.E. Steinberg:** None. **M.W. Salter:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.06

Topic: G.09. Drugs of Abuse and Addiction

Support: R01DA043535

Title: Normalization of Goal-Directed Behaviors Associated with Chronic Cannabis Use in Bipolar Disorder

Authors: *A. MIRANDA, A. MINASSIAN, J. W. YOUNG, W. PERRY;
Psychiatry, UCSD, San Diego, CA

Abstract: Cannabis use is highly prevalent in people with bipolar disorder (BD), with many reporting using cannabis to ameliorate symptoms. These symptoms include deficits in goal-directed behaviors (i.e., decision-making and hyper-motivation) and cognitive function (i.e., attention and learning). However, chronic cannabis use is also associated with cognitive impairment, thus it is unclear to what degree cannabis is useful in ameliorating symptoms of BD. Here, we determined the effects of chronic cannabis use on goal-directed behavior and cognition

that are impaired in people with BD. We recruited BD+ and BD- participants that were either cannabis users (C+) or non-users (C-). We performed a 2X2 ANOVA on interim data using BD and cannabis use as between-subjects factors on the 4 diagnostic groups: BD-/C- (n=25), BD-/C+(n-21), BD+/C- (n=8) and BD+/C+ (n=12). Participants were tested with a cognitive battery measuring risky decision-making (Iowa Gambling Task; IGT), motivation (Progressive Ratio Breakpoint Ratio Task; PRBT), reward learning (Probabilistic Learning Task; PLT) and sustained attention (5-C CPT). Overall, cannabis users were younger than non-users. Using age as a covariate, we observed BD x cannabis interaction effects on the IGT and PRBT. BD+/C+ participants showed less risk-prone behaviors on the IGT (F (1,63), p=.015, ES=.09) and normalized motivation on the PRBT (F (1,61), p=.045, ES=.065). We observed moderate effects of cannabis on punishment sensitivity (F (1,63), p=0.059, ES=0.055) and sustained attention (F (1,48), p=0.056, ES=0.074). Chronic cannabis use was associated with a modest improvement in some cognitive functions. Cannabis use was also associated with a normalization of risky decision making and effortful motivation in people with BD, but not healthy participants. Thus, chronic cannabis use may have uniquely beneficial effects in people with BD. Previous studies suggest that some people with BD have increased dopaminergic activity due to a reduced dopamine transporter expression. Chronic cannabis use has been shown to reduce dopamine release, thus chronic cannabis use may result in a return to dopamine homeostasis in people with BD and consequently normalizing their deficits in goal directed behaviors. We are engaged in additional studies that explore this potential dopaminergic/endocannabinoid mechanism.

Disclosures: **A. Miranda:** None. **A. Minassian:** None. **J.W. Young:** None. **W. Perry:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.07

Topic: G.09. Drugs of Abuse and Addiction

Support: DP1DA042232
DP1DA042232

Title: Chronic adolescent exposure to cannabis in mice leads to sex-specific transcriptional changes across different brain regions and links to genetic susceptibility to psychiatric disorders

Authors: *Y. ZUO[1], A. IEMOLO[1], P. MONTILLA-PEREZ[1], H.-R. LI[1], X. YANG[2], F. TELESE[1];
[1]Med., UCSD, La Jolla, CA; [2]Integrative Biol. and Physiol., UCLA, Los Angeles, CA

Abstract: During adolescence, frequent and heavy cannabis use can lead to serious adverse health effects and have been linked with an increased risk of psychiatric disorders including cannabis use disorders (CUD) and schizophrenia (SCZ). However, the underlying molecular mechanisms are largely unknown. Here, we used a mouse model of adolescent exposure to the

main psychoactive component of cannabis, delta-9-tetrahydrocannabinol (THC), which mimics the behavioral alterations observed in adolescent users. We treated female and male C57BL6/N mice with high doses of THC during early adolescence, assessed their memory and social behaviors in late adolescence, and then profiled the transcriptome of five brain regions involved in cognition and addiction. We performed analyses on the gene, pathway, and coexpression network levels and identified gene coexpression modules that simultaneously correlated with THC treatment and memory phenotypes reduced by THC, termed cognitive modules. The cognitive modules were related to endocannabinoid signaling in the female dorsal medial striatum, inflammation in the female ventral tegmental area, and synaptic transmission in the male nucleus accumbens. We further constructed cross-brain region module-module interaction networks and uncovered intra- and inter-region molecular circuitries influenced by THC. Moreover, we integrated genetic data from human CUD and SCZ with mouse THC-induced transcriptional changes and a data-informed, brain-specific Bayesian network to identify gene networks and their key driver (KD) genes in each sex and brain region. Among these KDs, two genes (Hapln4 and Elavl2) were shared between male and female nucleus accumbens, and orchestrated transcriptional networks correlated with THC exposure and genetic susceptibility of CUD and SCZ. Hapln4 and Elavl2 regulate transcriptional subnetworks implicated in synaptic transmission, addiction processes, brain development, and circadian entrainment. Our study reveals sex- and brain region-specific transcriptional responses to chronic adolescent cannabis exposure, sheds light on molecular mechanisms of THC-induced cognitive alterations, and provides novel insights into shared mechanisms of adolescent exposure to THC and CUD/SCZ genetic susceptibility, opening up avenues for further investigation.

Disclosures: **Y. Zuo:** None. **A. Iemolo:** None. **P. Montilla-Perez:** None. **H. Li:** None. **X. Yang:** None. **F. Telese:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.08

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH NIDA R21 Grant DA051689-01

Title: Identifying behavioral predictors of cannabis vapor self-administration in female and male rats

Authors: *G. I. PARK[1], A. N. MALENA[1], M. A. BARRETT[1], M. L. SPENCER[1], A. E. THOMPSON[1], N. C. GLODOSKY[2], C. CUTTLER[2], R. J. MCLAUGHLIN[1];
[1]Integrative Physiol. and Neurosci., [2]Psychology, Washington State Univ., Pullman, WA

Abstract: Approximately 9% of first-time cannabis users will become dependent, yet there are no FDA-approved pharmacotherapies for managing cannabis use disorder (CUD). This is in part

due to flawed diagnostic nosology resulting in a lack of understanding of the mechanisms that give rise to CUD, as well as a lack of translationally relevant animal models of cannabis use. To address these gaps, we have developed and validated a novel model of cannabis self-administration that delivers vaporized cannabis extracts in a response-contingent manner via the pulmonary route of administration. We used this model in this study to identify behavioral predictors of high vs. low rates of cannabis-seeking behavior. To accomplish this goal, we conducted a battery of behavioral assays in male and female Long Evans rats prior to vapor self-administration training. Specifically, we characterized the phenotype of rats using preclinical assays that match the 5 behavioral dimensions of the NIMH Research Domain Criteria (RDoC) (i.e., positive and negative valence, social communication, cognition, and arousal/regulatory systems) and then determined whether performance in these tasks was correlated with the number of cannabis vapor deliveries earned during a progressive ratio test. Preliminary results indicate that the number of trials required to meet criterion in the shift component of the attentional set-shifting task was strongly correlated with the number of cannabis vapor deliveries earned during a progressive ratio challenge in male and female rats (r=.407, p=.048). Additionally, time spent immobile in the forced swim test showed a moderate-sized correlation with the number of cannabis vapor deliveries earned (r=.39, p=.058). Notably, when sexes were analyzed separately, anogenital sniffing (r=.576, p=.05) and body sniffing (r=.556, p=.06) of a novel conspecific during a social play session were highly correlated with the number of cannabis vapor deliveries earned in male, but not female, rats. Overall, these data indicate that poor cognitive flexibility and more reliance on passive stress coping strategies are the strongest predictors of cannabis-seeking behavior. Moreover, measures of social investigation in male (but not female) adolescent rats are particularly strong predictors of motivation to self-administer cannabis. Given that alterations in endocannabinoid (ECB) degradation have been associated with an exaggerated response to cannabis and increased problematic drug use, future studies will examine whether cannabis self-administration causes alterations in ECB hydrolysis in brain regions involved in cognitive flexibility and stress coping.

Disclosures: **G.I. Park:** None. **A.N. Malena:** None. **M.A. Barrett:** None. **M.L. Spencer:** None. **A.E. Thompson:** None. **N.C. Glodosky:** None. **C. Cuttler:** None. **R.J. McLaughlin:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.09

Topic: G.09. Drugs of Abuse and Addiction

Support: This work was supported by the DICBR of the NIAAA

Title: Preclinical modeling of spontaneous THC withdrawal symptoms in mice: dopamine, sleep, and behavioral maladaptations

Authors: *A. J. KESNER[1], Y. MATEO[2], K. ABRAHAO[3], S. RAMOS-MACIEL[1], M. PAVA[2], A. L. GRACIAS[2], R. T. PAULSEN[2], H. B. CALSON[1], D. M. LOVINGER[2];
[1]Unit on Motivation and Arousal, [2]Lab. for Integrative Neurosci., NIAAA, Rockville, MD;
[3]Univ. Federal de Sao Paulo, Univ. Federal de Sao Paulo, Sao Paulo, Brazil

Abstract: Withdrawal symptoms are observed upon cessation of cannabis use in humans. Symptoms include irritability, decreased appetite or weight loss, restlessness, depressed mood and hedonics, and more, and are thought to be driven by physiological changes that correspond with building tolerance to delta-9-tetrahydrocannabinol (THC) during prolonged cannabis use. Spontaneous THC withdrawal symptoms are thought to be difficult to observe in laboratory animal, and as such, back-translational studies on the physiological mechanism of cannabis/THC withdrawal are lacking. We filled this knowledge gap by implementing mouse behavioral paradigms that target specific clinical withdrawal symptoms. THC tolerance was induced via 10 intraperitoneal injections of 10 mg/kg THC over 6 days. Behavioral measurements from pretreatment, and early and late THC abstinence epochs were obtained. *Sleep.* We observed profound alterations in sleep and vigilance-state architecture in male mice during early THC withdrawal that normalized in late withdrawal. The metrics mimic clinical observations. Conversely, female mice appear resilient to effects of THC withdrawal on sleep. *Reward seeking.* Mice performing an operant-cue discrimination task tended to earn fewer rewards, and disrupted response to reward predictive cues were more prominent in male mice, suggesting motivation and attention is altered in mice during THC abstinence. Conversely, we found no change in sucrose preference, indicating hedonics are not affected by THC withdrawal. *Irritability.* Using the bottle brush test we found specific alterations of irritability-like behaviors in male mice only. *Consumption and Locomotion.* During twenty four-hour home cage food and water intake and locomotion monitoring we observed minimal alterations to intake and locomotion during THC withdrawal. *Plasma corticosterone.* Changes in these behavioral indices do not appear to be mediated by stress per se, as circulating plasma corticosterone was only modestly increased in male but not female mice. *Neurochemistry.* The neurotransmitter dopamine plays a known role in manifestation of withdrawal symptoms, so using ex-vivo fast-scan cyclic voltammetry we found dopamine levels within the striatum male and female mice differed in their spatial and temporal profiles. These data open the door for further pre-clinical research efforts to determine the neurobiological bases of, and potentially treat, primary withdrawal symptoms of cannabis use disorder.

Disclosures: A.J. Kesner: None. **Y. Mateo:** None. **K. Abrahao:** None. **S. Ramos-Maciel:** None. **M. Pava:** None. **A.L. Gracias:** None. **R.T. Paulsen:** None. **H.B. Calson:** None. **D.M. Lovinger:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.10

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant U54 DA-16511-19S1

Title: Stress-reactivity and cognition in older adults with cannabis use disorder

Authors: R. HICKEY, O. HORN, S. FOUNTAIN-ZARAGOZA, L. NUNN, A. MCRAE-CLARK, *A. BENITEZ;
Med. Univ. of South Carolina, Charleston, SC

Abstract: OBJECTIVE: Cannabis use is increasing in prevalence among adults over age 50 but little is known about its impact on brain health. This abstract describes an ongoing pilot study that investigates the associations between cannabis use and stress, cognition, measures of brain health via MRI, and plasma Alzheimer's disease and related dementia (ADRD) biomarkers. The primary hypotheses of the study are that cannabis use will be associated with poorer cognition, and that older women will be more likely to demonstrate worse cognitive impairment due to peri-/post-menopausal loss of progesterone and greater stress driving cannabis use. METHODS: We are conducting a pilot study of participants 50-80 years old with cannabis use disorder (CUD). At baseline participants completed a blood draw, brain MRI, an ADRD-focused neuropsychological test battery, and the NIH Toolbox Cognition Battery. For the next 7 days, participants used a smartphone to complete craving and stress questionnaires before and after ecological stress induction. Saliva samples were collected from female participants to quantify daily progesterone levels. On Day 8, participants completed the Trier Social Stress Test (TSST), during which mood, craving, and salivary progesterone and cortisol were measured. We present interim analysis of data from 5 participants (4 males/1 female, ages 54-64), reporting measures of experimental stress induction, and demographically-adjusted composite z-scores for memory, language, attention, speed/executive function, visuospatial, and global cognitive abilities. RESULTS: In 4/5 participants, the TSST induced increased anxiety from low to moderate/high anxiety as determined by The State-Trait Anxiety Inventory, along with an increase in overall and emotionality craving scores at the first post-task timepoint using the Marijuana Craving Questionnaire. All participants had impaired scores on global cognitive ability (z-score median=-2.11, range=-4.67 to -1.59). Cognitive composite scores were not statistically significantly associated with total years of use, but the correlations were in the hypothesized directions and highest for the speed/executive function composite ($r = 0.78$, $p=0.12$). DISCUSSION: These interim results demonstrate successful stress induction and support our hypothesis that CUD in aging is associated with normatively worse cognition, which we hope to further test when we reach the target recruitment of N=40. We will update these interim results during presentation and provide preliminary tests of study hypotheses regarding progesterone and sex differences with greater participant accrual.

Disclosures: R. Hickey: None. **O. Horn:** None. **S. Fountain-Zaragoza:** None. **L. Nunn:** None. **A. McRae-Clark:** None. **A. Benitez:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.11

Topic: G.09. Drugs of Abuse and Addiction

Support: F32DA051160
R01NS042595
R34NS126036
Diabetes Research Center Pilot Project
Hope Center Pilot Project
Whitehall Foundation Grant
Rita Allen Scholar Award in Pain

Title: Evaluation of co-administration of oxycodone and Δ^9-Tetrahydrocannabinol on measures of antinociception, dependence and reward.

Authors: *R. SLIVICKI[1], J. WANG[3], V. NHAT[4], M. CREED[2], A. V. KRAVITZ[5], R. W. GEREAU, IV[6];
[2]Dept. of Pharmacol., [1]Washington Univ. In St. Louis, Saint Louis, MO; [3]Washington Univ., St. Louis, MO; [4]Washington Univ. in St. Louis, St. Louis, MO; [5]Washington Univ. In St Louis, Saint Louis, MO; [6]Anesthesiol., Washington Univ. Sch. of Med., St. Louis, MO

Abstract: Oxycodone is one of the most commonly prescribed analgesics for moderate to severe pain disorders. While effective in the short term, long-term use can result in several unwanted side-effects including tolerance, physical dependence and the development of opioid use disorder. Δ^9-Tetrahydrocannabinol (Δ^9-THC), is a partial cannabinoid receptor agonist which has been reported in to enhance opioid analgesia in various animal models. However, it remains unclear if Δ^9-THC can modulate other aspects of repeated oxycodone intake such as tolerance and dependence. Thus, this study sought to evaluate the impact of Δ^9-THC administration on measures of oxycodone-induced antinociception, dependence and reward in mice.
To evaluate for thermal antinociception, dose-response curves were conducted for both oxycodone and Δ^9-THC using the hotplate test. ED_{50}'s were calculated on day 1 30 min post injection. Animals were treated twice daily with vehicle, oxycodone, Δ^9-THC or combination of oxycodone+ Δ^9-THC for 5 days. On the 6th day, naloxone was administered to precipitate somatic withdrawal. Animals were video recorded and classic measures of opioid withdrawal (e.g. jumps) were evaluated using open-source pose estimation software (Deeplabcut). In combinatorial conditions, cages were outfitted with passive infrared sensors to evaluate for alterations in circadian activity. Δ^9-THC and oxycodone alone and in combination were evaluated in the conditioned place preference assay to evaluate reward and/or aversion using doses derived from hotplate ED_{50} calculations. Oxycodone and Δ^9-THC produced clear antinociceptive effects in the hotplate assay.
Oxycodone produced a robust conditioned place preference, while Δ^9-THC did not produce preference or aversion. Oxycodone alone produced alterations in circadian activity while Δ^9-THC did not. Evaluations of oxycodone and Δ^9-THC combination groups are currently ongoing. The results of these findings will be informative regarding the potential implementation of Δ^9-THC with oxycodone in a therapeutic setting.

Disclosures: R. Slivicki: None. **J. Wang:** None. **V. Nhat:** None. **M. Creed:** None. **A.V. Kravitz:** None. **R.W. Gereau:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.12

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH NIDA Grant R01DA049470

Title: Evaluation of chronic combination cannabidiol and oxycodone treatment on pain- and reward-related behavior

Authors: *A. C. BRICE-TUTT[1], W. MALPHURS[1], A. SHARMA[2], A. W. BRUIJNZEEL[3], M. FEBO[3], B. SETLOW[3], R. M. CAUDLE[4], N. P. MURPHY[1], J. K. NEUBERT[1];
[1]Orthodontics, [2]Pharmaceutics, [3]Psychiatry, [4]Oral and Maxillofacial Surgery, Univ. of Florida, Gainesville, FL

Abstract: The objective of this study was to evaluate the effects of chronic treatment with cannabidiol and oxycodone, alone or in combination, on pain- and reward-related behavior in male and female rats (N = 9-10/treatment/sex). Using the orofacial pain assessment device (OPAD), rats were trained to consume a positive reinforcer consisting of a sweetened condensed milk solution under both thermal nociceptive (44.5°C) and non-nociceptive (37°C) conditions. Rats were then treated daily for 14 days with oxycodone (0.56 mg/kg, i.p.), cannabidiol (3.2 and 10 mg/kg, i.p.), cannabidiol + oxycodone combinations, or vehicle, and operant responding for the reinforcer at the different temperatures was evaluated. To observe potential development of locomotor sensitization over the 14 days of treatment, rearing behavior was also recorded. We found that oxycodone increased operant responding under both nociceptive and non-nociceptive conditions. Neither dose of cannabidiol alone altered responding but when combined with oxycodone, cannabidiol dose-dependently increased responding beyond that produced by oxycodone alone. This effect was more efficacious at the higher temperature. During testing at 44.5°C, but not 37°C, there was a significant interaction between cannabidiol and oxycodone on the number of licks on the reward bottle made per nociceptive stimulus contact ($F_{(2,103)} = 5.84$, $p < 0.05$, two-way ANOVA with Tukey's *post hoc* test). Oxycodone also produced locomotor sensitization (increased rearing), an effect that was not influenced by cannabidiol. These results suggest that while being devoid of any inherent activity at the doses tested, cannabidiol may potentiate the analgesic effect of oxycodone in a manner that is specific to nociceptive conditions. As such, cannabidiol may be useful as an opioid-sparing approach to treating pain. Future work will include analysis of manganese enhanced resonance imaging data to determine potential changes in calcium dependent synaptic activity produced by chronic oxycodone and cannabidiol treatment.

Disclosures: A.C. Brice-Tutt: None. **W. Malphurs:** None. **A. Sharma:** None. **A.W. Bruijnzeel:** None. **M. Febo:** None. **B. Setlow:** None. **R.M. Caudle:** None. **N.P. Murphy:** None. **J.K. Neubert:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.13

Topic: G.09. Drugs of Abuse and Addiction

Title: Effects of Cannabinoid Withdrawal Using Marble Burying in Long Evans Rats

Authors: *C. FELTER, A. BREWER, S. SPENCER;
Pharmacol., Univ. of Minnesota, Minneapolis, MN

Abstract: Cannabinoid withdrawal plays a role in maintaining the use of a drug, thus influencing a substance use disorder (SUD). The symptoms associated with withdrawal in humans, including anxiety and cravings, interfere with efforts to achieve cessation of drug use. We will characterize the behavioral profile of cannabinoid withdrawal in rats using models that measure multiple endophenotypes of cannabinoid dependence that may contribute to withdrawal and craving. Initial studies will be performed using adult male and female Long Evans rats receiving twice daily infusions of the synthetic cannabinoid agonist WIN55,212-2 or vehicle via jugular catheter starting at 0.2 mg/kg and increasing to 0.8 mg/kg over 4.5 days. In our hands, this model produces dependence such that somatic signs of spontaneous withdrawal are observed at 6 hours following the final infusion. Here we used marble burying to evaluate repetitive and anxiety-like behavior following both precipitated and spontaneous withdrawal. Since precipitated withdrawal is induced with the cannabinoid receptor type 1 (CB1R) inverse agonist rimonabant (10mg/kg), we have performed pilot experiments for marble burying in cannabinoid naïve male and female rats using rimonabant to verify the pharmacological effects in controls. The marble burying test was performed using a rectangular box (20in x 16in x 8in) with a 3cm corn bedding covered floor. 24 colored glass marbles were spread evenly across the floor. One hour prior to testing the animals habituated to the experiment room. During experimentation, the animal was placed in the center of the marble box and behavior was measured for 30 minutes. Criteria for buried marbles included those that were at least 60% buried under the corn bedding. Behavior observed during the 30 minutes included rears, writhes, hind leg scratches, and fecal boli. Adult male (n=4) and female (n=2) rats showed a decrease in marbles buried when treated with rimonabant. This replicates prior studies that have been performed in mice. Rimonabant also increased fecal boli in male and female rats. These data are consistent with rimonabant producing an anxiogenic effect in untreated rats which we predict will be exacerbated in our WIN55,212-2 treated rats. We are now extending this pilot experiment and examining marble burying following rimonabant precipitated withdrawal in our final WIN55,212-2 dependence model. Withdrawal is precipitated with 10mg/kg of rimonabant 4 hours after the final WIN55,212-2 infusion. In the future, we plan to evaluate behavior following spontaneous withdrawal either 1 week or 2 weeks after the final drug infusion and extend these studies to additional behavioral screens.

Disclosures: C. Felter: None. **A. Brewer:** None. **S. Spencer:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.14

Topic: G.09. Drugs of Abuse and Addiction

Title: Dissecting patterns of motor behavior across withdrawal observations during precipitated and spontaneous cannabinoid withdrawal

Authors: *A. L. BREWER[1], C. E. FELTER[2], S. M. SPENCER[1];
[1]Pharmacol., [2]Univ. of Minnesota, Minneapolis, MN

Abstract: Cannabinoid withdrawal likely plays an important in role in maintaining use and preventing cessation of cannabis use. In humans cannabis withdrawal is often associated with anxiety, restlessness, and mood disturbances, while in the rodent it is most common to perform quantitative scoring of somatic behaviors associated with withdrawal. While the somatic component of cannabinoid withdrawal is important, this behavioral scoring does not completely encompass cannabinoid withdrawal. As cannabinoid withdrawal is observed in freely moving rats it seemed logical to simultaneously assess the effect of cannabinoid withdrawal on locomotor behavior during observation periods in male and female rats as a first pass to additionally probe whether withdrawal changed locomotion patterns or exploratory ambulation. Adult male and female Long Evans rats received twice daily infusions of WIN55,212-2 or vehicle through an indwelling catheter implanted into the jugular vein starting at 0.2 mg/kg and increasing to 0.8 mg/kg over 4.5 day. Withdrawal was either precipitated with 10 mg/kg of rimonabant 4 hours after the final infusion or observed at regular intervals for 30 min periods at 6, 14, 24, 48, 72, and 96 hours after the final infusion. A Med Associates open field activity chamber with monitoring software was outfitted with a two-chamber preference insert. One side of the two chamber insert was used during behavioral observations to collect locomotor data from freely moving rats during withdrawal observation periods. Precipitated withdrawal has no effect on locomotor activity in female rats. In male rat's cannabinoid withdrawal increases locomotor activity compared to vehicle. Similarly, males undergoing spontaneous withdrawal showed increased locomotor activity out to 72 hours after the final infusion. However, females undergoing withdrawal engaged in less locomotor activity then vehicle females out to 72 hours after the final infusion. Analysis of potential sex differences is ongoing. We are also analyzing the data to determine estrous stage effects on withdrawal-related locomotor behavior. These results show that cannabinoid withdrawal significantly changed motor behavior in the rat in a sex and time dependent manner. Analyzing the subtle changes in motor behaviors across withdrawal observations provide important insights into sex differences in cannabinoid withdrawal.

Disclosures: A.L. Brewer: None. **C.E. Felter:** None. **S.M. Spencer:** None.

Poster

562. Cannabinoid Pharmacology and Abuse

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 562.15

Title: WITHDRAWN

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.01

Topic: G.03. Motivation

Support: NIH Grant 5 T32 MH 19938-27

Title: Establishing high potency opioid self administration and behavioral economic demand analysis in mice

Authors: A. OSTERMAN[1], A. SHANK[1], J. TUCCIARONE[1], M. B. POMRENZE[1], D. F. CARDOZO PINTO[1], Z. ZHANG[1], G. TOUPONSE[2], N. ESHEL[2], R. C. MALENKA[1]; [1]Nancy Pritzker Laboratory, Dept. of Psychiatry and Behavioral Sci., [2]Psychiatry and Behavioral Sci., Stanford Univ., Stanford, CA

Abstract: The opioid crisis persists and we need a better understanding of the biological, psychological and social underpinnings of opioid use disorder, its remission, and relapse. From a biological perspective, rodent models are an important entry point into understanding evolutionary conserved circuits that drive motivated behavior and addiction. In particular, self-administration paradigms closely model components of human drug dependence. Behavioral economic analysis of demand as a function of cost provides a rigorous, mathematical framework to analyze motivated behaviors. Through the extraction of demand curves from behavioral data, one can quantify how much reward a mouse consumes in the absence of cost (Qo) and the amount of work the mouse is willing to perform to achieve that reward (α). Combining economic demand analysis with advances in mouse genetics could allow for high precision in dissecting brain networks, circuits, and cell types underlying drug seeking behavior. As a first step in this effort, we established a time-efficient and reliable protocol that combines high-potency opioid IV self-administration with behavioral economics demand analysis. This pairing has allowed us to mathematically model motivated behavior in drug-seeking laboratory mice. In an escalating fixed ratio (FR) learning schedule (FR1-FR3-FR5), 76% of our animals (n=17) learned to press an active lever for the fentanyl derivative remifentanil (0.1mg/kg/infusion), taking an average of 8 days to complete. Once threshold criteria for acquisition were reached, animals transitioned to the behavioral economics (BE) task. In BE, animals are trained to press an active lever for drug

reward during five 10-minute 'bins' of varying fixed ratios (FR1, FR5, FR10, FR21, FR46). Six out of seven mice met criteria and reached an average stabilized α of 0.00164 (+/- 0.0002) and average Qo of XYZ in 4.5 days. During extinction sessions, we removed all drug-associated cues and did not allow the mice to obtain the drug. This led to an 84% (+/- 12%) reduction in lever presses across an average of 15 days. Reintroducing drug-associated cues in a cue-induced reinstatement task then led to a three-fold increase in total lever presses, despite the continued absence of the drug. In sum, we have established a task in mice that allows for the analysis of economic demand, extinction, and reinstatement. Future directions for this project include manipulating and monitoring specific cell populations implicated in opioid reward and withdrawal-related aversion.

Disclosures: **A. Osterman:** None. **A. Shank:** None. **J. Tucciarone:** None. **M.B. Pomrenze:** None. **D.F. Cardozo Pinto:** None. **Z. Zhang:** None. **G. Touponse:** None. **N. Eshel:** F. Consulting Fees (e.g., advisory boards); Boehringer Ingelheim. **R.C. Malenka:** F. Consulting Fees (e.g., advisory boards); MapLight Therapeutics, Bright Minds Biosciences, Mindmed, Cyclerion, AZ Therapies.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.02

Topic: G.03. Motivation

Support: DA011289
Stanford NeuroChoice
Stanford Dean's Fellowship

Title: Dynorphin release acting at presynaptic kappa opioid receptors in the VTA modulates GABAergic LTP

Authors: *V. MARTINEZ DAMONTE[1,2], J. A. KAUER[1,2];
[1]Stanford Univ., Stanford, CA; [2]Dept. of Psychiatry and Behavioral Sci., Nancy Pritzker Lab., Stanford, CA

Abstract: The VTA is a brain region necessary for drug reinforcement. Plasticity at GABAergic synapses controlling dopamine neuron excitability is a target of drugs of abuse and acute stress. We previously reported that nitric oxide-induced potentiation of inhibitory postsynaptic currents (LTP_{GABA}) onto VTA dopamine cells is lost after a single acute exposure to cold-water swim stress. Stress block of LTP_{GABA} relies on a persistent activation of kappa opioid receptors (kORs). NorBNI, a long-lasting kOR inverse agonist, given either *in vivo* prior to forced-swim stress or bath applied after stress during the electrophysiological recordings rescues LTP_{GABA}. These experiments indicate that kOR activation is sufficient for stress to block $LTP_{GABA,}$ and hence deleting kORs from the relevant cell type should prevent this. Importantly, we have

previously shown that this stressor also induces reinstatement of cocaine seeking, and that this is also dependent on kORs.Here we began to identify the kappa receptor and dynorphin circuit elements responsible for the block of LTP_{GABA} to promote relapse behaviors. Using a conditional knock-out approach, we found that kORs in the postsynaptic dopamine cells are not required for stress-induced loss of LTP_{GABA} (stress: 89.5 ± 18.5 % of baseline IPSC amplitude, n=10 p=0.065 Wilcoxon test; stress+norBNI: 152.1 ± 35.7 % of baseline IPSC amplitude, n=10 p=0.013 Wilcoxon test) suggesting that instead the relevant kORs are on presynaptic GABAergic terminals. We found that GABAergic afferents from nucleus accumbens (NAc) undergo LTP_{GABA} (118.6 ± 7.5 % of baseline IPSC amplitude, n=24 p=0.045 Wilcoxon test), and that selectively deleting kORs from these terminals prevents stress-induced block of LTP_{GABA} (in progress, 145.6 ± 28.9 % of baseline IPSC amplitude, n=5 p=0.062 Wilcoxon test).
The VTA receives dynorphin afferents from different brain regions such as the BNST, amygdala, NAc and the lateral hypothalamus. Here we will identify specific dynorphin pathways that are critical to stress blocking LTP, by optogenetically driving individual sets of dynorphin afferents. With this approach in brain slices we will induce dynorphin release optogenetically and test for the presence of LTP_{GABA}. Defining the stress-sensitive circuit involved in this form of reinstatement will allow selective control of this pathway and suggest specific therapeutic targets for treatment of stress-triggered relapse.

Disclosures: V. Martinez Damonte: None. **J.A. Kauer:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.03

Topic: D.02. Somatosensation – Pain

Support: NIH R01DA054583
NIH T32NS007431
New York Stem Cell Foundation - Robertson Investigator

Title: A comprehensive dissection of cell type, circuit, and molecular mechanisms of action of opioids in the cerebral cortex

Authors: *N. OCHANDARENA[1], J. NIEHAUS[2], R. PICKEN[2], Z. YAO[3], D.-W. KIM[3], H. ZENG[4], G. SCHERRER[5];
[1]UNC Neurosci. Curriculum, [2]UNC Neurosci. Ctr., UNC Chapel Hill, Chapel Hill, NC; [3]Allen Inst. for Brain Sci., Seattle, WA; [4]Allen Inst. For Brain Sci., Seattle, WA; [5]UNC Neurosci. Center, Dept of Cell Biol. and Physiol., The Univ. of North Carolina at Chapel Hill, Chapel Hill, NC

Abstract: Opioids are indispensable analgesics that can also produce harmful side effects including tolerance, addiction, and death through respiratory depression. Understanding the

organization and function of neural circuits underlying these different effects may reveal strategies to disassociate opioid-induced analgesia from unwanted side effects. Opioids elicit their effects through the μ, κ, and δ opioid receptors (MOPr, KOPr, and DOPr), which are expressed in neurons throughout the nervous system. Opioids act on different regions of the cerebral cortex to alter sensorimotor function, motivation, and affect, but the molecular identity, connectivity, and functional contributions of the neurons involved are unclear. Specifically, although cortical regions such as the infralimbic, prelimbic, anterior cingulate, insular, somatosensory, and motor cortices have been studied for their contribution to the sensory, affective and cognitive dimensions of pain experience, how opioids modulate cortical activity remains poorly understood. Because several of these regions also contain circuits that contribute to opioid addiction, delineating analgesia-and addiction-promoting circuits is challenging. To address this issue, here we use single-cell RNA-sequencing (scRNA-seq), circuit tracing, recording and manipulation of neural activity in freely behaving mice to establish the mechanisms of action of opioids in the cerebral cortex. Although scRNA-seq has been used to investigate the cellular heterogeneity of the cortex, the distribution and molecular characteristics of OPr-expressing neurons across different cortical regions and layers remains unclear. Furthermore, the low sequencing depth of droplet-based high-throughput scRNA-seq approaches limits reliable detection of lowly expressed transcripts, like the OPr-encoding genes *Oprm1*, *Oprk1*, and *Oprd1*. We analyzed scRNA-seq data from cortical samples prepared by SMART-seq technology, which yields full length transcriptomes with exceptional sensitivity for sparse transcripts. We assessed ~80,000 neurons sorted from 15 cortical regions to comprehensively characterize the molecular identity of neurons expressing each OPr gene across neocortical areas. Using marker genes identified from these data, we applied intersectional genetic approaches to determine the spatial distribution of different OPr-expressing subpopulations across cortical regions and resolve region- and layer-specific cell types. Together, these experiments resolve the molecular and spatial configuration of opioid-sensitive cortical neurons for dissecting their contributions to opioid-induced behaviors.

Disclosures: **N. Ochandarena:** None. **J. Niehaus:** None. **R. Picken:** None. **Z. Yao:** None. **D. Kim:** None. **H. Zeng:** None. **G. Scherrer:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Epiodyne, US patent application 20210220489. F. Consulting Fees (e.g., advisory boards); NIH Preclinical Screening Platform for Pain External Consulting Board.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.04

Topic: G.03. Motivation

Support: NIH Grant K12GM093854
NIH Grant K99DA045765
NIH Grant R01DA006214

Title: The dynorphin/kappa opioid system drives aspects of negative affect during acute abstinence from oxycodone.

Authors: *K. NEWMAN[1], J. A. AVILA[2], J. E. FRAGALE[3], V. A. STIRITZ[3], M. H. JAMES[4], G. S. ASTON-JONES[2];
[1]Rutgers Univ. Grad. Program In Neurosci., Piscataway, NJ; [2]Brain Hlth. Inst., [4]Psychiatry, [3]Rutgers Univ., Piscataway, NJ

Abstract: The current opioid crisis stems in part from increased abuse of prescription opioids. One facet of opioid addiction is the development of negative affect in withdrawal, which may drive drug seeking via negative reinforcement. The hypothalamic neuropeptide orexin has been implicated in opioid seeking and withdrawal. Recently, our lab found that orexin signaling contributes to negative affective behaviors seen in acute abstinence from oxycodone. Notably, orexin neurons also release dynorphin, which acts at kappa opioid receptors (KORs) to regulate dysphoric responses, and thus might contribute to aspects of opioid-induced negative affect. We first characterized changes in the dynorphin/KOR system following chronic oxycodone treatment. Adult male Sprague Dawley rats were given 21d of twice-daily injections of saline or oxycodone (3 mg/kg; ip) and were tested for signs of negative affect during acute abstinence. Orexin and dynorphin expression were assessed with immunohistochemistry 24h following the final injection. Compared to saline-treated rats (n=6), oxycodone-treated rats (n=5) showed increased numbers of neurons that co-expressed orexin and dynorphin (independent samples t-test; $p<0.05$). A separate cohort of rats (n=8) was given the KOR antagonist norbinaltorphimine (norBNI; 30mg/kg; ip) or vehicle (n=8) 1 wk prior to 21d of oxycodone treatment, and negative affect was assessed in acute abstinence by measuring body weight, Von Frey thresholds (hyperalgesia), saccharin preference (anhedonia), elevated plus maze (anxiety-like behavior), and forced swim behavior (despair-like behavior). Compared to vehicle controls, norBNI normalized body weight gain and Von Frey thresholds (mixed-design ANOVAs; $p<0.001$ and $p<0.05$, respectively), as well as forced swim mobility (independent samples t-test; $p=0.0759$), but not anhedonic or anxiety-like behaviors. Together, these experiments support the hypothesis that increased release of both orexin and dynorphin contributes to aspects of negative affect emerging in opioid withdrawal.

Disclosures: K. Newman: None. **J.A. Avila:** None. **J.E. Fragale:** None. **V.A. Stiritz:** None. **M.H. James:** None. **G.S. Aston-Jones:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.05

Topic: G.03. Motivation

Support: NIDA R00 045765

Title: The orexin (hypocretin) system is necessary for the development and expression of 'food addiction' phenotypes in obese female rats

Authors: *J. MEHR[1], A. ARMANIOUS[1], C. HUR[1], U. GYAWALI[1], M. S. PALADINO[1], G. S. ASTON-JONES[2], N. T. BELLO[3], M. H. JAMES[1];
[1]Robert Wood Johnson Med. Sch. Dept. of Psychiatry, [2]Brain Hlth. Inst., Rutgers Univ., Piscataway, NJ; [3]Animal Sciences, SEBS, Rutgers Univ., New Brunswick, NJ

Abstract: Introduction: Binge eating has been argued to reflect an 'addictive-like' response to some foods. In rodents, the orexin (hypocretin) system mediates hedonic consumption of highly palatable foods and motivated drug seeking. However, the significance of the orexin system in the development and expression of 'food addiction' phenotypes has not been examined. **Methods**: Female orexin-Cre Long-Evans rats were maintained on a high fat diet (HFD; 45% fat, ad libitum) for 8 weeks and were then split into two groups: Group 1 rats received bilateral intra-hypothalamic injections of viruses containing either a Cre-dependent inwardly rectifying potassium channel (AAV9-DIO-Kir2.1-mCherry; n=8) or a control construct (AAV9-DIO-EYFP; n=7); Group 2 rats received bilateral intra-hypothalamic injections of viruses containing either a Cre-dependent hM4Di DREADD (AAV9-hSyn-DIO-hM4Di-mCherry; n=7) or a control construct (AAV9-DIO-GFP; n=7). To induce a 'food addiction' phenotype, rats were given restricted access (30 mins, twice/week) to a sweetened fat solution (90% Crisco 10% sucrose), which promotes binge-like eating. Rats were then tested for several 'food addiction' endophenotypes, including responding for sucrose on progressive ratio and behavioral economics schedules, punished (foot shock) responding for sucrose reward, and cued reinstatement of sucrose seeking. Rats in Group 2 were treated with the DREADD ligand JHU37160 (J60) 15 minutes prior to behavioral testing. **Results**: Restricted access to sweetened fat promoted heightened 'addiction' behaviors on all tests. The development of these behaviors was blocked by chronic inhibition of orexin neurons (Group 1), and their expression was suppressed by acute chemogenetic inhibition of orexin neurons (Group 2). **Conclusions**: These data indicate that the development and expression of 'food addiction' behaviors following binge-like eating is dependent on orexin signaling, highlighting this system as a potential target for therapeutics designed to manage food overconsumption.

Disclosures: J. Mehr: None. **A. Armanious:** None. **C. Hur:** None. **U. Gyawali:** None. **M.S. Paladino:** None. **G.S. Aston-Jones:** None. **N.T. Bello:** None. **M.H. James:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.06

Topic: G.03. Motivation

Support: NIDA Grant R00045765
NJHF Grant PC98-22

Title: A novel highly selective sigma 1 receptor antagonist reduces binge-like eating and motivated responding for palatable food in obese female rats

Authors: *A. J. ARMANIOUS[1,3], Y. PENG[4], W. J. WELSH[4,2], M. H. JAMES[1,3];
[1]Dept. of Psychiatry, [2]Dept. of Pharmacol., Robert Wood Johnson Med. School, Rutgers Univ., Piscataway, NJ; [3]Brain Hlth. Institute, Rutgers Univ., Piscataway, NJ; [4]Rutgers-Cancer Inst. of New Jersey, Rutgers Univ., New Brunswick, NJ

Abstract: Introduction: The sigma-1 receptor (S1R) is a highly promising target in novel therapeutics to treat binge eating disorder (BED). However, the development of S1R antagonists for clinical uses has remained elusive. X-ray crystallography recently revealed a ligand-binding pocket of antagonists of the S1R, which allowed us to design and validate a lead compound (PW507) that binds S1R at nanomolar concentrations, has excellent blood-brain permeability, and acceptable oral bioavailability. Here, we tested the efficacy of PW507 in reducing binge-like eating and motivated responding for palatable food in rats. **Methods**: Female Long Evans rats (n=16) were maintained on a high fat diet (45% fat, ad libitum) for 8 weeks before being given intermittent access (30 min, twice/week, 4w) to sweetened fat (vegetable shortening/10% sucrose). Rats were then trained to lever press for sucrose pellets on a low (fixed ratio [FR] 1) or high effort (FR5) schedule of reinforcement. Rats were treated with PW507 (0, 5, 10, 15, 20mg/kg; i.p.) 15mins prior to binge and operant test sessions using a within-subjects design. We also tested the effects of PW507 on locomotor activity in an open field apparatus. **Results**: PW507 dose-dependently decreased binge-like intake of sweetened fat and high effort (FR5) responding for sucrose pellets. At the lowest effective dose (10mg/kg), PW507 had no effect on low effort (FR1) responding for sucrose or general locomotor activity. **Conclusions**: These data indicate that PW507 selectively reduces binge-like eating and motivated responding for palatable foods, supporting its potential utility as a novel compound for the management of BED and other disorders associated with food overconsumption.

Disclosures: A.J. Armanious: None. **Y. Peng:** None. **W.J. Welsh:** None. **M.H. James:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.07

Topic: G.03. Motivation

Support: NIDA R00 045765
Rutgers CEED NEIHS P50 Pilot Grant

Title: Peri-pubertal bisphenol-A exposure alters orexin expression in early adulthood: consequences for binge-like eating and mood outcomes

Authors: *M. M. BILOTTI[1], V. ISSKANDAR[2], S. Z. ANTHONY[1], T. A. ROEPKE[3], N. T. BELLO[3], M. H. JAMES[4];

[1]Joint Grad. Program in Toxicology, [2]Cell Biol. and Neurosci., [3]Animal Sci., [4]Psychiatry, Rutgers Univ., Piscataway, NJ

Abstract: Introduction: Bisphenol A (BPA) was a widely used chemical in the manufacturing of plastics and has noted estrogenic actions. Although the use of BPA has been limited, the long-term consequences of BPA exposure are unclear. Specifically, consequences of BPA exposure during adolescence, a period of heightened steroid sex hormone involvement and heightened risk of psychiatric disorder onset, has not been characterized. Thus, here we examined whether BPA exposure during the peri-pubertal period altered the risk for binge-like eating and mood dysregulation in early adulthood. Methods: Male (n=8/group) and female (n=12/group) Long-Evans rats were exposed to BPA (0, 25, 250µg/kg/day) via their drinking water between post-natal days (PND) 29-56. Female rats were monitored for vaginal opening (VO) and cytology. BPA was discontinued on PND 57, after which all rats were given intermittent limited access to sweetened fat (vegetable shortening with 10% sucrose; 30mins, twice/week for four weeks). Rats were also tested for anxiety-like behavior on the Open Field Test (OFT). On PND 97, brains were collected for immunohistochemistry and qPCR analyses. Results: In females, both BPA doses were associated with irregular cycling, earlier VO, reduced binge-like intake, and enhanced anxiety-like behavior; this was associated with decreased gene expression of orexin (Hcrt), and increased expression of orexin receptor 1 (Hcrtr1), in lateral hypothalamus. In males, BPA exposure was associated with increased binge-like eating; data on male orexin expression are forthcoming. Conclusions: Peri-pubertal BPA exposure has sex-dependent effects on binge-like eating in adulthood. Reduced binge-like eating in females following peri-pubertal BPA exposure is associated with reduced orexin gene expression pointing to the orexin system as a potential mechanistic target underlying these effects.

Disclosures: M.M. Bilotti: None. **V. Isskandar:** None. **S.Z. Anthony:** None. **T.A. Roepke:** None. **N.T. Bello:** None. **M.H. James:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.08

Topic: G.03. Motivation

Support: NIH grant 5R01DA006214-31

Title: Binge sucrose and saccharin-induced neuroadaptations: a focus on the orexin-hypocretin system in female rats

Authors: D. DE SA NOGUEIRA*, Y. RAKHOLIA, M. GREENEN, G. ASTON-JONES; Rutgers- Brain Hlth. Inst., Piscataway, NJ

Abstract: Binge eating disorder is the most common eating disorder. Animals, like humans, selectively binge on highly palatable food suggesting that this behavior is driven by hedonic,

rather than metabolic signals but the neuronal mechanisms involved in this maladaptive behavior are not well known. Behavioral and molecular adaptations induced by eating disorders share commonalities with those involved in addiction. Given that orexin/hypocretin signaling is linked to both reward processing and food intake, we examined the contribution of this system to binge-like eating in female rats. Separate groups were given intermittent (12h) or continuous (24h) access to 10% sucrose or 0.4% saccharin and food over 28 days. Only the intermittent access to sucrose group displayed excessive sucrose intake within an hour (i.e., binge eating). All the sucrose and saccharin groups exhibited an increase in the number of orexin-A peptide neurons within LH compared to the group with limited access to food only. The orexin 1 receptor antagonist, SB334867 (20 or 30mg/kg), reduced binge-like intake in groups with limited access to sucrose or saccharin but not in rats with continuous access to sucrose. We are currently assessing whether binge-like sucrose or saccharin intake alters expression of prepro-orexin RNA, or affects economic demand for cocaine. Altogether, our findings indicate that sucrose or saccharin bingeing alters the orexin system in LH. Our results broaden the understanding of neural alterations associated with binge-eating, and point towards addictive-like properties of palatable food.

Disclosures: D. De Sa Nogueira*: None. **Y. Rakholia:** None. **M. Greenen:** None. **G. Aston-Jones:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.09

Title: WITHDRAWN

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.10

Topic: G.03. Motivation

Support: NIH R00DA045765

Title: The orexin (hypocretin) system as a common mediator of dysregulated sleep and relapse in cocaine conditioned rats

Authors: *U. GYAWALI, M. S. PALADINO, K. SUCHOJAD, M. H. JAMES;
Rutgers Univ., Piscataway, NJ

Abstract: Substance use disorders are frequently associated with sleep disturbances, especially during acute withdrawal. Rats with a history of cocaine self-administration display significant sleep fragmentation, along with a decreased amount of nonrapid-eye-movement (NREM) and rapid-eye-movement (REM) sleep. Further, normalization of sleep during abstinence reduces drug craving, indicating a potential causal link between these two processes. The orexin (hypocretin) neuropeptide system represents a highly promising therapeutic target to normalize cravings associated with sleep dysregulation, as orexins simultaneously mediate the activity of brain circuits involved in both reward and arousal. However, direct evidence linking orexin signaling with dysregulated sleep and relapse-like behavior is lacking. In Experiment 1, we used conditioned place preference (CPP) to establish a contextual association with acute bolus injections of cocaine (10mg/kg). Rats then underwent 5d of drug abstinence followed by a place preference test. During abstinence, rats were treated daily with the dual orexin receptor antagonist suvorexant (0, 30 mg/kg, p.o.) immediately prior to the onset of the inactive period. In Experiment 2, rats were trained to self-administer cocaine on an intermittent access schedule before undergoing 2w home cage abstinence. During abstinence, rats were treated daily with suvorexant (p.o, 0, 30 mg/kg) immediately prior to the onset of the inactive period. On days 1 and 15 of abstinence, rats were tested for lever responding for drug-associated cues. In both experiments, electroencephalogram (EEG) and electromyogram (EMG) activity was monitored throughout the experiment in a subset of rats. In Experiment 1, we observed sleep disturbances on cocaine conditioning days, characterized by sleep fragmentation (more frequent stage transitions). These changes persisted into abstinence but were abrogated by suvorexant treatment. In Experiment 2, rats exhibited an incubation of craving across abstinence; data relating to the effects of suvorexant on incubation is forthcoming. Together, these data indicate that blocking orexin receptor signaling might be an effective strategy for normalizing sleep dysregulation and reducing craving during cocaine abstinence.

Disclosures: U. Gyawali: None. **M.S. Paladino:** None. **K. Suchojad:** None. **M.H. James:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.11

Topic: G.09. Drugs of Abuse and Addiction

Support: National Institute on Drug Abuse DA048336
National Institute on Alcohol Abuse and Alcoholism T32: AA007583

Title: The effects of chronic adolescent, and acute adult, nicotine exposure on the motivational efficacy of opioids: Behavioral economic analyses of polysubstance use

Authors: *S. C. HONEYCUTT[1], D. D. LICHTE[1], E. A. GILLES-THOMAS[1], S. L. MCSAIN[2], A. MUKHERJEE[1], G. C. LONEY[1];
[1]Psychology, [2]Biol. Sci., Univ. at Buffalo, Buffalo, NY

Abstract: Nicotine and opioid use disorder (OUD) are highly comorbid and nicotine increases liability for misusing prescription opioids. We have shown previously that former adolescent nicotine exposure (ANE) and ongoing acute nicotine administration in adulthood (ANA) significantly increase compulsive-like consumption of opioids in rats. Specifically, relative to controls, ANA and ANE rats self-administer more total opioids, have higher breakpoints in progressive ratio tests, and remain more motivated to consume opioids despite contingent foot-shock punishment. While it is established that nicotine enhances opioid intake regardless of age of initial exposure, it is unknown if ANE and ANA affect the essential value of, and demand for, opioids identically. To that end, we employed behavioral economics (BE) analyses to generate demand curves for remifentanil (RMF) in intravenous self-administration (IVSA) paradigms. These procedures compare various behavioral indices of opioid consumption as a function of cost between nicotine-treated and control rats. Briefly, male and female Long-Evans rats underwent ANE procedures in which they were treated with nicotine (0.4 mg/kg) or saline twice a day for 10 days (PND34-43). Upon reaching PND 75, rats were trained to self-administer RMF (3.2 µg/kg/inf) in operant chambers in 2-hr sessions on an FR-1 schedule for 10 days. Following training, rats underwent BE demand curve tests in which every 10 min the dose of delivered RMF was systematically decreased (3.2, 2.0, 1.0, 0.6, 0.3, 0.2, 0.1, 0.06, 0.03, 0.02, 0.01, 0.006 µg/kg) across the 2-hr session. Curve-fits generated motivational indices of RMF consumption at low price (Q0), normalized elasticity (α), and maximal responding (OMax). Rats in the ANA condition were given acute injections of nicotine 15 min prior to behavioral testing. Analyses revealed that both ANE and ANA significantly increased RMF consumption ($F(1, 38) = 4.18$, $p < 0.05$). Preliminary analyses of the BE factors revealed that only demand (Q0) tended to differ between ANE and control rats while Q0 and OMax where significantly higher, and elasticity (α) significantly lower, in ANA rats relative to controls. Therefore, despite both forms of nicotine administration serving to significantly enhance opioid consumption, it appears that ANE and ANA differentially affect the motivational properties and essential value of opioids. As such, differential neurobehavioral mechanisms may contribute to the augmentation of opioid consumption as a function of form of nicotine administration. Future studies are planned to increase statistical power and to examine how prior punishment affects the demand for opioids.

Disclosures: S.C. Honeycutt: None. **D.D. Lichte:** None. **E.A. Gilles-Thomas:** None. **S.L. McSain:** None. **A. Mukherjee:** None. **G.C. Loney:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.12

Topic: G.03. Motivation

Support: NIH Grant NS120628

Title: Input-specific regulation of discrete populations of lateral habenula neurons by kappa opioid receptors

Authors: *S. SIMMONS, S. GOUTY, W. FLERLADGE, B. COX, F. LISCHKA, J. SMYTH, F. NUGENT;
Uniformed Services Univ. of the Hlth. Sci., Bethesda, MD

Abstract: The lateral habenula (LHb) is an epithalamic brain region associated with value-based decision making and stress evasion through its modulation of dopamine (DA)-mediated reward circuitry. Increased activity of the LHb is associated with drug addiction and stress-related mood disorders. Additionally, dynorphin (DYN)/Kappa opioid receptor (KOR) signaling is an endogenous mediator of stress response in reward circuitry. Previously, we have shown a novel functional role of KOR signaling in LHb of rats which is altered by severe early life stress, 24h maternal deprivation in rats. In non-stressed rats, KOR activation has bidirectional effect on LHb neuronal excitability, which is altered in maternal deprived rats. Here we used several methods to elucidate the neural circuitry and synaptic integration of DYN/KOR signaling in LHb in both rats and mice. First, we used an unbiased high-throughput approach of in-vitro GCAMP calcium signaling combined with electrophysiology in the LHb of mice to identify effects of KOR agonist (U50,488) on LHb spontaneous neuronal activity. Consistent with our previous findings from rats, we found a bidirectional modulation of GCaMP signals by KORs within different subpopulations of LHb neurons that were reflective of changes in bursting and high frequency tonic activity. To identify KOR-expressing or DYN-expressing projections to the LHb we used viral retrograde tracing in KOR-Cre and DYN-Cre mice and identified entopenduncular nucleus (EP) as a major KOR-expressing input to the LHb. EP has been implicated in stress-induced mood disorders and may contribute to aberrant LHb excitability in depressive-like phenotypes. KOR activation significantly reduced LHb action potential generation in a subset of LHb neurons in response to optical stimulation of EP inputs. This suggests input and cell-type specific KOR regulation of LHb neurons. In maternal deprivation model, we have also found a shift in excitation to inhibition balance (E/I) towards excitation at EP-LHb inputs. Furthermore, we have piloted study using limited bedding and nesting (LBN) model of early life adversity which uncovered sex-specific differences in basal LHb activity as well as increased LHb bursting in both sexes in LBN mice. In the future we will explore the effects of both early life stress models on KOR modulation of LHb activity in projection and input-specific manner.

Disclosures: S. Simmons: None. **S. Gouty:** None. **W. Flerladge:** None. **B. Cox:** None. **F. Lischka:** None. **J. Smyth:** None. **F. Nugent:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.13

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA/NIH DP1DA051550

Title: Heroin sex-specifically regulates orbitofrontal circular RNAs

Authors: *G. FLORIS[1], S. SILLIVAN[2];
[1]Temple Univ., [2]Temple Univ., Philadelphia, PA

Abstract: Opioid use disorder (OUD) and dependence have become one of the largest public health crises in the US, with devastating socio-economic repercussions. To curb this epidemic, the identification of new targets and the development of newer and better strategies is necessary for OUD prevention and therapy. One potential strategy is represented by circular RNAs (circRNAs), a class of long noncoding RNAs (lncRNAs) characterized by the presence of covalently linked ends produced in a noncanonical splicing event called back-splicing. CircRNA regulatory roles pass through all steps of gene regulation, ranging from mRNA transcription and splicing to RNA decoy and translation (Floris et al., 2017). Nevertheless, only few circRNAs have been associated with a clear function and while studies on their regulatory roles in psychiatric disorders are emerging, studies on OUD are still lacking. We have established in our lab an animal model of heroin self-administration in which rats are trained to stably self-administer heroin. After 10 self-administration sessions, we profile circRNAs in the orbitofrontal cortex (OFC) of heroin-exposed rats by RNAseq. The OFC, a brain area previously implicated in drug seeking behavior and relapse, is also one of the brain regions with the highest abundance of circRNAs. Our circRNA profile has revealed a total of 42 differentially expressed circRNAs in males (23 up regulated 19 down regulated) and 34 in females (21 up regulated and 13 down regulated). We noted the only two circRNAs were found to be common between males and females, but their expression was in opposite direction suggesting sex-specific circRNA regulation by heroin. Next, we apply an integrated bioinformatics analysis to sex-specific regulated circRNAs to identify microRNAs and associated pathways predicted to bind heroin-regulated circRNAs. We found a total of 59 and 52 miRNA that were associated with up regulated circRNAs in males and females respectively. Strikingly, we found that 24 of these circRNA-associated miRNAs were common in both sexes. MiRNA-associated pathway analysis of common miRNAs has revealed 16 significantly enriched terms including morphine addiction but also cocaine and amphetamine addiction, consistent with a potential role of circRNAs in miRNAs regulation following heroin self-administration. It appears that despite sex-specific regulation of heroin-associated circRNA, there is convergence on the regulation of common miRNAs that can be important for regulating heroin-associated pathways and therefore contribute to the cellular adaptations that arise from chronic heroin administration.

Disclosures: G. Floris: None. **S. Sillivan:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.14

Topic: G.09. Drugs of Abuse and Addiction

Support: ReBUILDetroit Bridge Award

Title: A translational rodent model of opioid exposure during pregnancy: Effects of morphine compared to buprenorphine on maternal behavior, brain, and the microbiome

Authors: *L. RICHARDSON[1], A. MYERS[1], C. M. WALLIN[1], S. NEOLE[1], N. KULAGLIC[1], S. A. PERRINE[2], C. J. DAVIDSON[2], M. ANGOA-PEREZ[2], D. M. KUHN[2], S. E. BOWEN[1], S. BRUMMELTE[1];
[1]Psychology, [2]Psychiatry and Behavioral Neurosciences, Wayne State Univ., Detroit, MI

Abstract: The recent opioid epidemic has resulted in many opioid-dependent pregnant women receiving opioid maintenance therapies. Buprenorphine (BUP) or methadone are pharmacological treatments used to reduce negative effects of misused opioids on the mother and developing fetus. Clinically, BUP produces preferable outcomes for exposed infants as compared to methadone or opioid misuse. However, a dearth of knowledge remains for BUP's effects on neural networks that are critical during the transition to motherhood and for maternal caregiving behaviors. BUP's mechanism of action (partial mu-agonist/kappa antagonist) varies significantly from morphine's (MS; full mu-agonist), which may result in a different impact on the maternal brain during a critical neuroplasticity period. In the current study, we used a translational rodent model to mimic chronic opioid (mis)use (MS exposure, 3-6mg/kg/day, b.i.d.) or opioid maintenance drug (BUP exposure, 1mg/kg/day) to investigate the behavioral and neurochemical consequences of gestational opioid exposure on dams and their offspring. Opioid or saline administration to female rats (N=50) via subcutaneous injections began 7 days prior to mating and continued daily throughout pregnancy until postpartum day 2 (PD2) or was discontinued on gestational day 19 to allow for drug clearance before parturition. Dams' maternal behaviors were monitored through detailed observations of pup-directed and non-pup directed behaviors. Dams were also evaluated with several behavioral tests, including a pup retrieval test, a hunting task, and two-chamber pup-odor preference test. Our preliminary findings indicate that continued and discontinued BUP exposure resulted in more maternal care deficits, increased postpartum pup mortality, and maternal deficiencies in the pup retrieval and pup-odor preference test, but not in the hunting task, as compared to our control group. Conversely, MS may have resulted in fewer pregnancies (despite obvious sperm in vaginal lavage samples), but care behavior and survival rates of the MS groups varied little from controls. Interestingly, each opioid also resulted in a unique change in the microbiome profile of the gut. Using high performance liquid chromatography, we will further analyze neurotransmitter levels and their metabolites in the maternal brain (collected on PD2) to investigate potential neurological effects of these opioids on the maternal brain network. More research is critical to elucidate how BUP mechanistically interacts with the neural network during the transition to motherhood to help alleviate possible negative consequences.

Disclosures: L. Richardson: None. **A. Myers:** None. **C.M. Wallin:** None. **S. Neole:** None. **N. Kulaglic:** None. **S.A. Perrine:** None. **C.J. Davidson:** None. **M. Angoa-Perez:** None. **D.M. Kuhn:** None. **S.E. Bowen:** None. **S. Brummelte:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.15

Topic: G.09. Drugs of Abuse and Addiction

Title: The Microbiomes Influence on Opioid Addiction

Authors: *S. C. IRIAH[1], N. RODRIGUEZ[2], M. FEBO[3], P. KULKARNI[4], C. F. FERRIS[5]; [1]Northeastern Univ., Northeastern Univ., Boston, MA; [2]Univ. of Florida, Gainsville, FL; [3]Univ. of Florida, Univ. of Florida Dept. of Neurosci., Gainesville, FL; [4]Northeastern Univ., Northeastern Univ. Dept. of Psychology, Boston, MA; [5]Northeastern Univ. Program In Behavioral Neurosci., Northeastern Univ. Program In Behavioral Neurosci., Boston, MA

Abstract: Opioids are the strongest and most effective analgesics available for acute pain management. With no viable alternative for treating chronic or post operative pain, it is not surprising that over 10 million people misuse opioids and 75,673 Americans died from opioid overdose in 2021. To our knowledge this is the first study to explore the developmental influence of the gut microbiota on resistance to addictive behavior and functional connectivity. Germ free (GF) mice which are microbiologically sterile, show a significant reduction in conditioned place preference (CPP) after oxycodone (OXY) opioid exposure when compared to wild type mice (WT). Furthermore, once GF mice are conventionalized i.e., take on the microbiota of WT controls, CPP reward behavior mirrors WT mice. There were no significant differences in brain volumes between germ-free and wild-type before or after conventionalization using a 3D MRI mouse atlas providing site-specific analysis on over 140 different brain areas. However functional connectivity data showed significant differences across several brain regions e.g. midbrain, hippocampus, prefrontal ctx between germ free and wild type before and after conventionalization. Fecal analysis before conventionalization showed complete absence of bacteria with the exception of Corynebacterium psudogenitalium-tuberculostearicum, Staphylococcus haemolyticus, Paenibacillus amylolyticus-barcinonesis-oceanisediminis and Turicibacter sanguinis. Following conventionalization these opportunistic bacteria disappeared and were replaced with a spectrum of bacteria similar across experimental groups. The implications of these results suggest the contents of the microbiome have a direct impact on the development of reward seeking behavior. With the widespread number of opioid receptors found in the gut, studying the interaction between the microbiota and substance use disorder may lead to a better understanding of the mechanism that leads to addiction development as well as potential treatments.

Disclosures: S.C. Iriah: None. **N. Rodriguez:** None. **M. Febo:** None. **P. Kulkarni:** None. **C.F. Ferris:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.16

Topic: G.09. Drugs of Abuse and Addiction

Support: AA020919
DA035958

Title: Mechanoreceptor activation ameliorates symptoms of opiate withdrawal in rats by modulating mesolimbic GABA and dopamine neurons

Authors: *G. C. JONES[1], C. A. SMALL[2], A. A. MOEDL[2], D. Z. OTTESON[3], S. C. STEFFENSEN[4], K. B. BILLS[5];
[2]Psychology, [3]Biomed. Sci., [4]Dept of Psych and Neuro, [1]Brigham Young Univ., Provo, UT; [5]Dept. of Biomed. Sci., Noorda Col. of Osteo. Med., Springville, UT

Abstract: Previous research suggests that applied mechanical stimulation in the form of whole-body vibration (MStim) modulates substrates which activate the mesolimbic dopaminergic (DA) pathway between the ventral tegmental area (VTA) and nucleus accumbens (NAc). While this treatment has been explored in rats with alcohol dependency, it has not previously been studied in the context of opioid withdrawal. We aimed to determine whether MStim applied via vibration plate at 80 Hz would ameliorate the effects of morphine withdrawal as manifested behaviorally and electrophysiologically in 52 Wistar rats (n=26 with MStim treatment). We utilized an assay of behavioral tests, in vivo single unit recordings, and in-vivo measures of DA release. We also analyzed physiological alterations via the use of whole-cell patch clamp. Rats in morphine withdrawal that underwent MStim treatment spent on average 24% less time (±16%, $p < 0.05$) in closed areas of the Elevated Plus maze as compared to naive rats. In naive rats, an acute dose of morphine causes an inhibition of VTA GABA neurons (8%baseline; 1mg/kg). Rats in withdrawal from morphine exhibit a tolerance to an acute reinstatement dose (91%baseline; 1mg/kg). These effects are mitigated with MStim treatment one hour before the reinstatement dose (34%baseline; 1mg/kg). Baseline firing rate elevation of VTA GABA neurons, typical in withdrawal, was also reduced in MStim treated rats. The behavioral assay and electrophysiological measurements indicate a significant anxiolytic effect of MStim treatment on opioid withdrawal. Studies are ongoing that will examine the effect of MStim on subsequent dopamine release in the NAc as well as the effect of MStim on ultrasonic vocalizations, analgesia, and locomotor activity.

Disclosures: G.C. Jones: None. **C.A. Small:** None. **A.A. Moedl:** None. **D.Z. Otteson:** None. **S.C. Steffensen:** None. **K.B. Bills:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.17

Topic: G.09. Drugs of Abuse and Addiction

Support: NSF GRFP
OHSU startup funds

Title: Cell-type specific upregulation of calcium-permeable AMPA receptors in the rat nucleus accumbens during incubation of oxycodone craving

Authors: *K. ENGELN[1], A. MOUTIER[1], E.-K. HWANG[2], M. E. WOLF[3];
[1]Oregon Hlth. and Sci. Univ. Sch. of Med., Portland, OR; [2]Oregon Hlth. & Sci. Univ. Behavioral Neurosci., Portland, OR; [3]Behavioral Neurosci., Oregon Hlth. & Sci. Univ., Portland, OR

Abstract: The prescription opioid oxycodone (Oxy) is a widely prescribed and highly effective treatment for pain, but its addicting properties and severe withdrawal symptoms have contributed significantly to the opioid epidemic. Avoiding relapse to Oxy during abstinence is a major challenge for recovering Oxy users because cues associated with previous Oxy intake can elicit cravings that often lead to relapse even after prolonged periods of abstinence. Cue-induced opioid cravings that persist throughout abstinence can be studied in rats using the incubation of drug craving model, in which cue-induced drug seeking progressively increases during withdrawal from drug self-administration.
Studies of incubation of stimulant craving (cocaine, meth) have demonstrated that calcium-permeable AMPA receptors (CP-AMPAR) upregulate on medium spiny neurons (MSN) in the nucleus accumbens (NAc) during incubation and their activation is required for its expression in late withdrawal. Recently, work from our lab has shown that CP-AMPAR are also upregulated on MSN following incubation of Oxy craving. However, drug seeking behavior is differentially regulated by MSN expressing the dopamine D1 receptor (D1R) or the dopamine D2 receptor (D2R). The purpose of this study was to determine whether upregulation of CP-AMPAR occurs on D1 and/or D2 MSN during incubation of Oxy craving.
We used adult male and female D1-cre or A2a-Cre rats (A2aR is a marker for the D2R) crossed with a TdTomato reporter line so that D1 and A2a MSN could be visualized. Rats underwent 10 days of extended access Oxy or saline self-administration (SA) (6 h/day for 10 days; 0.1 mg/kg/infusion). Following SA, rats were tested for cue-induced drug seeking on withdrawal day (WD1) and WD15 to assess incubation. Two to twelve days after the WD15 seeking test, whole cell recordings of NAc core MSN were performed from optically identified D1 or A2a MSN. The contribution of CP-AMPAR to excitatory postsynaptic currents was assessed using the rectification index and naspm sensitivity in Oxy and saline groups.
Compared to cells from saline-treated animals, CP-AMPAR were upregulated on D1 MSN similar to what we have observed for stimulants. However, unlike stimulants, CP-AMPAR were also detected on A2a MSN. These results suggest that CP-AMPAR upregulation is a form of plasticity common to protracted withdrawal from stimulants and the opioid oxycodone, but strengthening of excitatory synaptic transmission on A2a MSN may regulate unique aspects of opioid withdrawal.

Disclosures: K. Engeln: None. **A. Moutier:** None. **E. Hwang:** None. **M.E. Wolf:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.18

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA Grant DA048336
NIAAA T32 Grant AA007583

Title: Acute nicotine administration affects the hedonic properties of opioids without interfering with the ability to discriminate the opioid-interoceptive state

Authors: *A. MUKHERJEE[1], M. PALADINO[1], S. MCSAIN[2], R. CAMADINE[1], Z. GIANDOMENICO[3], K. SONTATE[1], S. HONEYCUTT[1], G. LONEY[1];
[1]Dept. of Psychology, [2]Dept. of Biol. Sci., [3]Dept. of Biotechnical and Clin. Lab. Sci., Univ. at Buffalo, Buffalo, NY

Abstract: Nearly 90% of those undergoing treatment for opioid use disorder report comorbid nicotine use, as such neurobehavioral effects of nicotine and opioid co-use need further investigation. Previous work shows that systemic and intra-insular administration of nicotine reduce the strength of a conditioned taste aversion to morphine (5.0-20 mg/kg) and enhance conditioned place preference to higher doses of morphine that control rats find less reinforcing (10-20 mg/kg). One interpretation is that nicotine specifically reduces the malaise-inducing effects of morphine while another is that it generally reduces sensitivity to the interoceptive effects of morphine. Here we used an occasion-setting paradigm where the morphine drug state serves as a positive feature for establishing a predictive relationship between a discrete cue and delivery of a food pellet. Adult male Wistar rats were assigned to continuous pretreatment with either nicotine (0.4mg/kg, s.c.) or saline (0.1ml/kg, s.c.) and subsequently treated with either morphine (3mg/kg, s.c) or saline on CS+ or CS- days, respectively. Testing order occurred in a pseudorandom pattern. On CS+ days, morphine administration was indicative that a stimulus light predicted response-independent delivery of banana-flavored sucrose pellets. On CS- days, a saline injection was given and cue light illumination was absent of any consequences. Ability to discriminate between CS+ and CS- interoceptive states was assessed using discrimination scores, calculated as the difference in number of head entries that occurred during the pre-cue period from those that occurred during cue illumination. We analyzed both the first head entry discrimination score which occurred in the absence of any feedback, as well as the total discrimination score for that day. Conditioning lasted for a total of 18 days. Analysis of the First Discrimination Score revealed significant main effects of Stimulus and Day ($F_{(1,16)}$: 46.48, p<0.05; $F_{(17,242)}$: 3.28, p<0.05 respectively), whereas the Total Discrimination Score had a significant Stimulus x Day interaction ($F_{(17,272)}$: 6.99, p<0.05) indicating that rats could effectively discriminate between the morphine and saline states as conditioning continued. There were no significant effects of nicotine on the First Discrimination Score (ps=0.47-0.98) or Total Discrimination Score (ps=0.77-0.84), indicating that nicotine does not affect the ability to

discriminate the interoceptive effects of morphine from saline in this paradigm. As such, these data suggest that nicotine may specifically increase the reinforcing effects of opioids, possibly by obfuscating its aversive effects.

Disclosures: A. Mukherjee: None. **M. Paladino:** None. **S. McSain:** None. **R. Camadine:** None. **Z. Giandomenico:** None. **K. Sontate:** None. **S. Honeycutt:** None. **G. Loney:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.19

Topic: G.09. Drugs of Abuse and Addiction

Title: Preclinical effects of a specific NOP receptor agonist on heroin addiction-like behaviors

Authors: *A. MARTINEZ[1], C. CROOK[1], S. DIRIK[1], N. T. ZAVERI[2], M. KALLUPI[1], G. DE GUGLIELMO[1];
[1]Psychiatry, UC San Diego, La Jolla, CA; [2]Astraea Therapeutics, LLC, Mountain View, CA

Abstract: Over the past 15 years opioid-related overdose deaths have quadrupled, but the neurobiological mechanisms that are responsible for the escalation of heroin use are poorly known. Extensive work has found that the activation of mu-opioid receptors is critical for the initial euphoric and acute positive reinforcing effects of opioids. In our recent work we found that the dysregulation of another opioid receptor, the nociceptin/orphaninFQ (N/OFQ) receptor, may be even more important after chronic exposure to opioids. Here tested a novel N/OFQ receptor-selective non peptide agonist (AT-403), on different measures of OUD (motivation to seek heroin, withdrawal-induced hyperalgesia and opioid-induced respiratory depression). Male and female Wistar rats were trained to self-administer heroin on 12h daily sessions for 3 weeks. After establishment of escalation of intake, the rats were tested for the effects of AT403 on their motivation for heroin(progressive ratio schedule) and on withdrawal-induced hyperalgesia (vonFrey test). The rats then underwent extinction, were treated with AT-403 and tested for its effect on cue-induced reinstatement of heroin seeking in a counterbalanced order. Finally, in a separate cohort of animals, we tested the effects of AT403 on the heroin-induced respiratory depression. Rats were injected IP in a counterbalanced order with AT403 or its vehicle and 2h later with 2.5mg/kg IV of heroin. Respiration was measured using a MouseOX® Plus - pulseoximeter in freely moving rats using a sensor via a neck collar (obtains vital signs from the carotid artery) connected to a monitor. Oxygen saturation (SpO2) and respiratory rate were recorded for 5 min before the heroin injection (baseline) and for 10 min after the heroin injection.The results showed that AT403treatment significantly reduced the motivation for heroin, as reflected by the lower breakpoint reached in the PR session in the treated rats. Treatment with AT403 dose-dependently reduced hyperalgesia during withdrawal, as demonstrated by the increased paw withdrawal threshold int he treated rats. In the reinstatement study, the pretreatment with AT-403 prevented the cue-induced reinstatement in a dose-

dependent manner. Finally, in the respiratory depression study, heroin induced significant SpO2 drop (from 95% to50%) in all the animals, but the ones treated with AT403 showed a faster recovery as demonstrated by the significantly higher SpO2 in the last 2minutes of recording.These results suggest that the development and repurposing of small molecules that target the N/OFQ system may have therapeutic efficacy in the treatment of opioid use disorder.

Disclosures: **A. Martinez:** None. **C. Crook:** None. **S. Dirik:** None. **N.T. Zaveri:** None. **M. Kallupi:** None. **G. De Guglielmo:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.20

Topic: G.09. Drugs of Abuse and Addiction

Support: ERC grant 802885

Title: Stress-enhanced opioid self-administration in healthy men

Authors: *M. EIKEMO[1], G. E. LØSETH[1], G. ERNST[2,1], M. CARLYLE[1], C. PAZMANDI[1], M. THOMPSON[1], C. VEZZANI[1], I. MEIER[3], M. TRØSTHEIM[3], T. JOHNSTONE[4], M. A. HEILIG[5], G. BIELE[1,6], S. LEKNES[1,3];
[1]Dept. of Psychology, Univ. of Oslo, Oslo, Norway; [2]Kongsberg Hosp., Kongsberg, Norway; [3]Oslo Univ. Hosp., Oslo, Norway; [4]Sch. of Hlth. Sci., Swinburne Univ. of Technol., Hawthorn, Australia; [5]Ctr. for Social and Affective Neurosci. (CSAN), Linköping Univ., Linköping, Sweden; [6]Norwegian Inst. of Publ. Hlth., Oslo, Norway

Abstract: Most people will receive an opioid drug during their lifetime. The rate of addiction following prescription opioid use is alarming. Non-human animal research links addiction with the powerful relief opioids can offer to animals in distress. In humans, epidemiological and clinical studies converge upon stress as a key risk factor for addiction. Despite this, the mechanisms through which stress alters opioid abuse liability in humans remain poorly understood. Here, we used a pre-drug social stress induction before opioid infusion in healthy men and women. The study was preregistered (OSF.io/bcxv8). Healthy volunteers participated in a double-blind placebo-controlled pseudo-randomized repeated-measures study; 63 completed all four 3-hour sessions (32 women, mean age 30 years, BMI 24). Two potent stress tasks and two non-stressful control tasks were performed immediately before dose 1 of oxycodone (i.v. 3mg/70kg) or saline. The dose was piloted to yield noticeable subjective effects, with few adverse effects. 15 minutes later, participants performed an effort-based self-administration task in which they could work to obtain 0-125% of the first dose effect. Dose 2 was given ~45 minutes later. Subjective state ratings, heart rate and blood samples were collected repeatedly. *Self-administration* data were analyzed using hierarchical Bayesian ordinal logistic regression. Overall, pre-drug stress increased oxycodone self-administration by 5% (OR = 2.2 [1.03, 4.6]).

The increase was driven by the men, who on average self-administered 14% more drug after stress (OR = 5.15 [1.6, 16.4] per 5%). Men exhibited significantly larger stress-enhanced abuse liability than the women (-3%, OR = 0.9 [0.4, 2.2] per 5%), despite lower subjective and physiological stress indices. We found no direct stress relief from oxycodone in men or women. This study reveals a mechanism through which acute stress can alter opioid abuse liability. We find that men may preferentially turn to opioid drugs in response to stress.

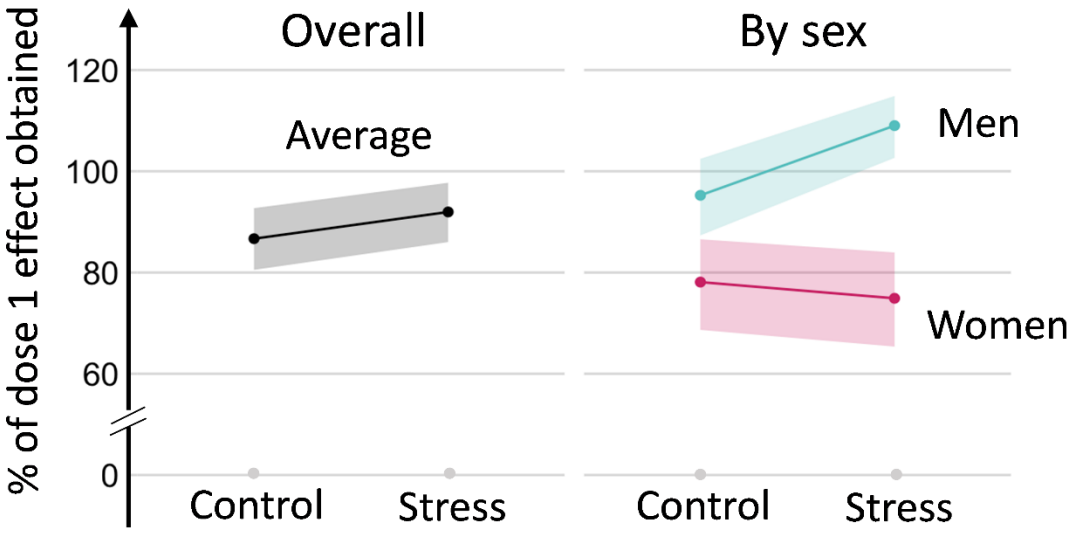


Figure 1. *Average dose self-administered oxycodone immediately following the stress and control inductions (estimated means). Ribbons represent the 95% highest density interval (HDI).*

Disclosures: **M. Eikemo:** None. **G.E. Løseth:** None. **G. Ernst:** None. **M. Carlyle:** None. **C. Pazmandi:** None. **M. Thompson:** None. **C. Vezzani:** None. **I. Meier:** None. **M. Trøstheim:** None. **T. Johnstone:** None. **M.A. Heilig:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Janssen, BrainsWay. F. Consulting Fees (e.g., advisory boards); Aelis, Indivior, Camurus, Pfizer, Novo Nordisk, Endosane. **G. Biele:** None. **S. Leknes:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.21

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA grant R01DA039100
Commonwealth Universal Research Enhancement Program

Title: The effects of dual hypocretin/orexin receptor blockade on oxycodone seeking and dopamine neurotransmission in the nucleus accumbens

Authors: *K. R. SAMSON, R. A. ESPAÑA;
Neurobio. and Anat., Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: A major barrier in successful treatment of opioid use disorder is persistent drug craving during periods of abstinence, which in turn, greatly influences relapse rates. While opioid-based medications have been used for treatment of opioid use disorder for decades, there remains a critical need for novel pharmacotherapeutic targets to treat opioid use disorder. The hypocretin/orexin (hypocretin) system is an attractive candidate for treating opioid use disorder due to substantial evidence that hypocretins influence motivation for drugs of abuse and dopamine neurotransmission. In the current studies, we examined the effects of the FDA approved dual hypocretin receptor antagonist suvorexant on oxycodone seeking and dopamine transmission during abstinence in female and male rats. Rats self-administered oxycodone under an intermittent access schedule for 10 days and were then subjected to 14 days of forced abstinence in their home cage. Rats were treated with either vehicle or 30 mg/kg suvorexant 24 hr prior to a cue-induced seeking test on abstinence day 14. On abstinence day 15, we performed *ex vivo* fast-scan cyclic voltammetry in the nucleus accumbens core to measure dopamine release and uptake. Preliminary results indicate that treatment with suvorexant reduces drug seeking behavior, but these effects may not be associated with changes in dopamine release or uptake rate. These data suggests that hypocretin based treatments may be useful in reducing opioid craving, although the extent to which these actions involve dopamine in the nucleus accumbens remains unclear.

Disclosures: K.R. Samson: None. **R.A. España:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.22

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA/NIH Grant DA045836
NIDA/NIH Grant DA048974

Title: Infralimbic cortex interneurons encode the incentive salience of heroin cues in a rodent model of opioid use disorder

Authors: *R. VAREED[1], G. GIANNOTTI[2], J. PETERS[2];
[1]Univ. of Colorado, Anschutz Med. Neurosci. Grad. Training Program, Univ. of Colorado, Anschutz Med. Neurosci. Grad. Training Program, Aurora, CO; [2]Univ. of Colorado Anschutz Med. Campus, Univ. of Colorado Anschutz Med. Campus, Aurora, CO

Abstract: Infralimbic (IL) cortex exerts top-down inhibitory control over drug seeking in preclinical rodent models of substance use disorders. Interneurons in the IL are the predominant cell type expressing the mu-opioid receptor (MOR), though they comprise the minority of the IL neuronal population (~5%). We used a rodent model of opioid use disorder (OUD) combined with in vivo fiber photometry and the fluorescent calcium sensor RCaMP to examine the activity of IL interneurons during different phases of heroin self-administration, extinction, and reinstatement. A dual virus strategy was employed to restrict RCaMP to IL interneurons by injecting a cocktail of DLX-Cre and DIO-RCaMP1b viruses into the IL cortex of male and female rats. Heroin self-administration training occurred over a period of 13 daily sessions, beginning on an FR1 schedule of reinforcement. Thereafter, the response requirement increased to a VR5 and then VR15. Heroin infusions were paired with a tone and light cue that was subsequently used to trigger reinstatement, or relapse. After heroin self-administration, heroin and heroin cues were withheld during operant extinction training, and drug seeking extinguished over 7 daily sessions. On the reinstatement test, the heroin cues were re-introduced (on a VR15), but heroin was not. We observed an increase in interneuron calcium activity (ΔF/F (z-score) + AUC analysis) in response to rewarded lever presses by the end of FR1 self-administration training. This activity was maintained over VR5 and VR15 training, but only for rewarded presses. During extinction training, interneuron activity was not associated with heroin seeking. During the cued reinstatement test, this pattern of increased IL interneuron activity returned, but only in response to lever presses which delivered the heroin cue (e.g. in-ratio presses), suggesting that IL interneurons are tracking delivery of heroin cues. Consistent with this, non-contingent presentations of the heroin cue (but not heroin alone) were associated with a similar pattern of IL interneuron activation. After the initial cued reinstatement test, rats continued to undergo daily cue tests for an additional 5 days, in order to extinguish heroin seeking in the presence of heroin cues. IL interneuron activity extinguished in parallel with behavioral extinction. Collectively, these data indicate that IL interneurons encode the incentive salience of heroin cues, suggesting that strategies targeting this small neuronal subpopulation may yield viable new treatments for OUD.

Disclosures: R. Vareed: None. **G. Giannotti:** None. **J. Peters:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.23

Topic: G.09. Drugs of Abuse and Addiction

Support: DA048974 to GG
DA045836 to JP

Title: The paraventricular thalamus to nucleus accumbens pathway mediates aversive states during withdrawal from heroin self-administration

Authors: *G. GIANNOTTI, J. PETERS;
Anesthesiol., Univ. of Colorado Anschutz Med. Campus, Aurora, CO

Abstract: Clinical studies indicate that the aversive somatic states experienced during opioid withdrawal correlate with drug craving and can increase the risk of relapse. We recently found the paraventricular thalamus (PVT) to the Nucleus accumbens (NAc) pathway to be necessary for heroin relapse after abstinence, but not after extinction training. Here we used chemogenetics to investigate the contribution of the PVT→NAc pathway to heroin withdrawal-induced hyperalgesia and aversive somatic states after extinction training or abstinence from heroin self-administration. A dual virus strategy employing an AAVrg-Cre injection into the NAc and a DIO-Gi-DREADD into the PVT was used to chemogenetically inhibit activity in the PVT→NAc pathway. After 12 days of heroin self-administration and 14 d of withdrawal (7d of extinction or 14 d of abstinence), mechanical hyperalgesia and aversive somatic signs were assessed 15 min after J60 or vehicle pretreatment. We found that heroin withdrawal-mediated hyperalgesia and somatic states were maximal during early (24 h) withdrawal from heroin self-administration. Interestingly, the extinction training procedure reduced aversion during protracted (14 d) withdrawal compared to acute (24h) withdrawal, and chemogenetic inhibition of the PVT→NAc pathway after extinction did not further reduce aversive withdrawal states. In the absence of extinction training, aversive states remained high even after protracted abstinence, and PVT→NAc pathway inhibition significantly reduced aversion metrics to levels seen after extinction training. To emulate the effects of extinction training, in a separate cohort of male and female rats, we applied an vivo LTD protocol (1 Hz -15 min) 45 min before a cued relapse test. In vivo fiber-photometry validated that this protocol effectively induces LTD, evidenced by reduced optogenetically evoked calcium transients in the NAc after induction. However, the data set is currently underpowered to detect a decrease in heroin seeking during the relapse test (in this same cohort). Finally, using the withdrawal-related aversion metrics to train a regression machine learning model, I was able to accurately predict the individual propensity to relapse (i.e., active lever presses) during the cued relapse test. Overall, these data suggest that the PVT→NAc pathway is necessary for the aversive experience of heroin withdrawal, and that extinction training may reduce aversion during withdrawal by diminishing activity in this pathway.

Disclosures: G. Giannotti: None. **J. Peters:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.24

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA R21 DA052706

Title: Time-dependent and sex-specific alterations in neural circuit control over remifentanil self-administration.

Authors: *A. OUIMET, R. PUPP, A. CZYZ, M. HEARING;
Marquette Univ., Milwaukee, WI

Abstract: Opioids are mainstays for pain management despite risk of dependence and abuse even when used as prescribed. Canonical thinking in the addiction field posits that early drug use is controlled and mediated by goal-directed circuits such as the prefrontal cortex (PFC). With extended use, due in part to hypofunction of the PFC, drug taking is thought to become more reliant on cortico-striatal habit-associated circuits, however, this has not been empirically demonstrated with opioids. We recently found that self-administration of the opioid, remifentanil, promotes a progressive hypoactive state in the prelimbic cortical region of the PFC (PrLC) that underlies impaired decision-making, develops faster in females, and aligns with an escalation of drug intake and drug motivation. The present study sought to examine whether 1) the PrLC becomes less involved in drug seeking with increased exposure, 2) if this phenomenon occurs faster in female mice, and 3) if effects of PrLC inhibition are selective for drug rewards. To do so, we virally expressed the inhibitory hM4Di DREADD or an mcherry control in pyramidal neurons of the PrLC (AAV8-CaMKII-hM4Di) of male and female C57bl6/j mice (PXX-XX). Mice intravenously self-administered (SA) remifentanil (5ug/kg/infusion) or orally administer a liquid reward (50% Ensure) for up to 28 days (3h/day/session). Habituating saline injections were given (i.p) on day 13 and 27 of SA with clozapine-n-oxide administered to all mice (2.0 mg/kg; i.p.) on day 14 and 28, with drug available during all sessions. Data show that CNO reduces active lever pressing and remifentanil infusions in both male ($n= 14$; $p<0.001$) and female ($n=11$; $p<0.01$) on day 14 versus the prior saline day. Preliminary findings ($n=3$) in females indicate that unlike early SA, CNO is no longer effective at reducing drug intake on day 28 of SA. Alternatively, inhibition of the PrLC reduced active lever pressing and consumption of Ensure in males ($n=10$; $p<0.001$) but not females ($n=0.8$) on day 14, but no longer alters behavior in males on day 28 ($n=3$; $p=0.7$). This data highlights a time-dependent and sex-specific role for the PrLC in driving opioid and non-drug reward taking. Ongoing studies are further examining effects of PrLC inhibition on remifentanil and Ensure SA at the more protracted 28 day timepoint and using DREADD approaches to target circuits associated with habit, including the anterior dorsal striatum. As clinical literature indicate that females tend to transition towards out-of-control drug use more rapidly than men, this data indicates that this may be due in part to a more rapid hypofunction of PrL-PFC and regulatory control over behavior.

Disclosures: **A. Ouimet:** None. **R. Pupp:** None. **A. Czyz:** None. **M. Hearing:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.25

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA R21 DA052706

Title: Motivational shifts during protracted opioid withdrawal: a sex- and dopamine subcircuit-specific effect

Authors: *E. BRETTINGEN[1], D. GOMEZ[2], M. C. HEARING[3];
[2]Biomed. Sci., [1]Marquette Univ., Milwaukee, WI; [3]Dept. of Biomed. Sci., Marquette Univ., MILWAUKEE, WI

Abstract: According to recent CDC data, opioid overdose deaths inthe US have increased 38% from 2019 to 2020. Withdrawal symptoms, both physical and psychological, are believed to contribute to addictive behaviors in opioid-use disorder patients. Among these psychological symptoms, opioid withdrawal has been associated with motivational shifts favoring increased drug intake, paralleled by decreased motivation for non-drug reward. The mesolimbic dopamine system is believed to be acritical regulator of motivated behavior, including that associated with addiction. However, studies of this system have traditionally failed to include periods of protracted withdrawal and have failed to compare possible differences indiscrete mesolimbic subcircuits. To address the lack of data during protracted withdrawal, we find that morphine dependent male and female mice subjected toa14-day period of forced drug abstinence display increased morphine intake and motivation for morphine compared to previously non-dependent mice (males: Mann-Whitney p=.0029, n=10,14; female: Mann-Whitney p=.0404, n=12,12). A sex-dependent motivational shift away from sucrose towards morphine was also exhibited in morphine dependent mice (male decrease in sucrose breakpoint values: Wilcox test p=0.0132, n=23;male increase inmorphine breakpoint values: Mann-Whitney p=0.0029, n=10,14). Using retrograde labeling and whole-cell slice recordings, we find increased rheobase (t-test p=.0167) along with increased inhibitory postsynaptic potentials(t-test p=.0199) in lateral ventral tegmental area(VTA) dopamine neurons projecting to the lateral nucleus accumbens (NAc) shell that is not observed in the medial VTA-NAc shell subcircuit. Further, selective chemogenetic inhibition of the lateral VTA-NAc shell subcircuit (2mg/kg CNO) decreases motivation for sucrose reward in drug-naive mice (Wilcox test p=.0313,n=6), an effect lacking in medial VTA-NAc shell subcircuit targeted mice and viral controls (Wilcox test p=.1250, n=6). This data suggests that motivational shifts away from non-drug rewards during protracted withdrawal is associated with latVTA-NAc hypoactivity that is driven by increased GABAergic inhibition of dopamine cells.

Disclosures: E. Brettingen: None. **M.C. Hearing:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.26

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH DA050908
Start up fund from University of Kansas

Title: Prefrontal cortex to ventral tegmental area projection regulates early social isolation stress-potentiated heroin seeking

Authors: *Y. WANG, S. YUE, G. LI, A. SINGH, Z. WANG;
Pharmacol. and Toxicology, The Univ. of Kansas, Lawrence, KS

Abstract: Prefrontal cortex to ventral tegmental area projection regulates early social isolation stress-potentiated heroin seeking
Yunwanbin Wang, Shuwen Yue, Guohui Li and Zi-Jun WangDepartment of Pharmacology & Toxicology, University of Kansas, Lawrence, Kansas, USA.
Abstract: Projections from prefrontal cortex (PFC) to ventral tegmental area (VTA) is an important pathway in regulating motivational behaviors. Nevertheless, the role of PFC to VTA projections in drug addiction-related behaviors is not clear. Our recent study shows that early social isolation (ESI) stress enhances heroin addictive-like behaviors and reduces the neuronal activity in both PFC and VTA. However, whether this circuit is casually linked to ESI-induced addiction vulnerability to heroin is unclear. By integrating chemogenetic tools, translating ribosome affinity purification (TRAP), behavioral, electrophysiological, and bioinformatic strategies, we evaluated how PFC-VTA projection contributes to the cue-induced heroin seeking in control and ESI mice. We found that the frequency of spontaneous action potential (sAP) of prefrontal cortical pyramidal neurons projecting to VTA was decreased during heroin relapse. Moreover, ESI stress lowered the sAP of prefrontal cortical pyramidal neurons on the projection compared to group-housed (GH) mice. We also showed that chemogenetic activation of the PFC-VTA circuitry with DREADD (designed receptor exclusively activated by designed drugs) tools attenuated cue-induced heroin-seeking behavior in ESI mice, whereas inhibition of this projection upregulated heroin-seeking behavior in GH mice. In the meantime, the activation of the circuitry recovered heroin-reduced sAP frequency caused by ESI stress. We also profiled the transcriptional changes that potentially contribute to ESI-potentiated heroin seeking on the PFC-VTA projecting neurons using the TRAP approach. Our data showed that transcriptional changes in PFC-VTA projecting neurons caused by ESI stress and heroin relapse are clustered in signaling pathways related to oxidative stress and mitochondrial damage. In summary, these results indicate that PFC-VTA projection is involved in the ESI-potentiated heroin-seeking. Our work will provide novel insight into the understanding of neurobiology underlying OUD.

Disclosures: Y. Wang: None. **S. Yue:** None. **G. Li:** None. **A. Singh:** None. **Z. Wang:** None.

Poster

563. Opioids: Reward and Reinforcement II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 563.27

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH-R01MH084894

Title: Sex-specific role for the 5-HT_{2A}receptor in the effects of psychedelics on opioid place preference in mice

Authors: *A. M. JASTER[1,2], J. GONZALEZ-MAESO[3,1];
[2]Pharmacol. and Toxicology, [3]Physiol. and Biophysics, [1]Virginia Commonwealth Univ., Richmond, VA

Abstract: Opioid use disorder affects over two million people in the United States with more than 120,000 deaths worldwide annually attributed to opioids. Psychedelic serotonergic agonists, such as lysergic acid diethylamide (LSD) and psilocybin, are being increasingly studied for their potential therapeutic effects in treatment of a variety of psychiatric conditions, including substance use disorders (SUD). Psychedelics produce profound alterations in human perception and sensory processing through activation of the serotonin 2A receptor (5-HT_{2A}R), but they do not cause dependence, and there is no known risk of lethal overdose. Activation of serotonin receptors has modulatory effects on the mesolimbic pathway, which is implicated in the neurobiology of addiction. Administration of psychedelics activates 5-HT_{2A}Rs in frontal cortex pyramidal neurons, which also project to subcortical regions involved in dopaminergic pathways, such as the nucleus accumbens. Our recent findings suggest differences across sexes on head-twitch behavior - a physiological proxy of psychedelic action - in C57BL/6J mice. Here, we aimed to assess the effect of psilocybin, a tryptamine psychedelic, on oxycodone-induced conditioned place preference (CPP) as a model of drug-craving in male and female C57BL/6J mice. Male and female animals displayed oxycodone preference as compared to vehicle. Importantly, male mice showed a decrease in expression of oxycodone-induced CPP following post-acute psilocybin administration, a phenotype that was not observed in female mice. The potential role of 5-HT_{2A}R-dependent signaling mechanisms in these SUD-related phenotypes was tested in knockout mice and controls. Our results support the notion that psychedelics alter sex-specific behaviors associated with preclinical models of SUDs potentially via 5-HT_{2A}R, and suggest that this class of psychoactive drugs may induce long-lasting therapeutic effects relevant to addiction.

Disclosures: A.M. Jaster: None. **J. Gonzalez-Maeso:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.01

Topic: H.03. Decision Making

Support: Department of Defense VBFF
NIH Grant 5U19NS107609-03

Title: Rapid learning with highly localized synaptic plasticity

Authors: *M. C. ROSEN[1], N. Y. MASSE[1,3], D. J. FREEDMAN[1,2];
[1]Dept. of Neurobio., [2]Inst. for Neurosci., Univ. of Chicago, Chicago, IL; [3]Meta Platforms, Inc., New York, NY

Abstract: The brains of all animals are plastic, a feature critical for forming new memories, adapting to new environments, and learning new tasks. However, plasticity, particularly of synaptic connections, is energetically expensive and can overwrite or interfere with prior knowledge. These costs can be mitigated by limiting the scope of synaptic plasticity within a neural network, as in reservoir computing, which employs large, fixed recurrent networks with plastic readout synapses. But reservoir approaches struggle to learn and flexibly switch between complex tasks. We are thus interested in understanding the tradeoffs between rigidity and plasticity in neural networks, and how compartmentalized plasticity can promote context-dependent learning and behavior. Here, we use biologically-inspired recurrent neural network (RNN) models obeying Dale's principle to show that sparse, localized plasticity-at ~0.5% of a network's synapses-can support rapid multitask learning (~6,000 trials apiece for each of 8 tasks). This learning requires specific combinations of network properties, such as topology, normalization, and reciprocal connection strength: for networks with 2,500 units, 26 of the hyperparameter combinations we tested (N=~2x105) reached 85% accuracy across all 8 tasks (chance = 12.5%). Analysis of network activity dynamics suggests that this learning occurs through a mechanism of error-driven subspace capture. Initially, network activity across all tasks occupies a common, overlapping subspace, precluding context-dependent behaviors. As learning proceeds, activity during each task is progressively sequestered into its own subspace of network activity that emphasizes the information relevant for that task, diminishing interference between tasks. Simultaneously, learning aligns readout weights specifically to those dimensions of activity that discriminate between decisions for each task, even if these are not the dominant variance-carrying dimensions of activity during that task. Together, these mechanisms-adjustment of top-down weights to drive network activity to task-specific subspaces that enact task-appropriate computations, and alignment of readouts to catch decision-related variation within these subspaces-facilitate reliable, context-dependent readout of different decisions about a single set of stimuli. Importantly, the arrangement of task subspaces with respect to one another reflects the similarity between the computations that different tasks require. This work demonstrates how rapid learning may be accomplished using localized, highly-constrained synaptic plasticity.

Disclosures: M.C. Rosen: None. **N.Y. Masse:** None. **D.J. Freedman:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.02

Topic: H.03. Decision Making

Support: NIH R01 DC018650
NIH R00 DC015014
NSF CAREER 2145247
BBRF NARSAD

Title: Dynamic and structured action bias masks learned task contingencies early in learning

Authors: *Z. ZHU[1], C. DRIEU[2], K. KUCHIBHOTLA[2];
[1]Neurosci., Johns Hopkins Univ. Sch. of Med., Baltimore, MD; [2]Psychology and Brain Sci., Johns Hopkins Univ., Baltimore, MD

Abstract: Learning is not only the acquisition of knowledge, but also the ability to express that knowledge when needed. We tested the acquisition of task knowledge using non-reinforced probe trials while mice were trained to perform a balanced, wheel-based auditory two-alternative forced choice (2AFC) task. During probe trials, animals exhibited surprisingly higher accuracy and lower directional bias early in learning, as compared to reinforced trials, suggesting that they have already acquired unbiased knowledge of stimulus-action contingency, but expressed this knowledge much slower under reinforcement. Why do animals exhibit this gap in accuracy and directional bias between acquisition and expression, despite already acquiring the stimulus-action associations? Animals may (1) exhibit motor biases that they slowly learn to suppress, (2) continue to explore different choice alternatives, or (3) base decisions on recent trial history, including choice and reward, rather than current stimuli. To test between these and other potential drivers, we first used a generalized linear model to separate different contributors to animals' choice during learning, including stimulus identity, trial history effects, and a continuous but slowly evolving directional preference (not influenced by stimulus or history factors), which we term action bias. Action bias, but not trial history, was the most important contributor to choice besides stimulus identity, and partially bridged the gap between acquisition and expression. We then asked if the structure of this action bias is static, reflecting a motor bias, or dynamic, reflecting changing behavioral strategies. Individual animals showed distinct states with left or right bias in blocks of tens to hundreds of trials and transitioned between both directions and un-biased states throughout learning, suggesting a dynamic bias structure. As learning progressed, animals exhibited less extreme bias, but continued to transition in and out of low biased states even at expert level performance. Taken together, behavioral expression may reflect an action bias driven exploratory process that is uncoupled from acquisition, evolves during learning, and persists to a lower degree at expert level to potentially maintain flexibility.

Disclosures: Z. Zhu: None. **C. Drieu:** None. **K. Kuchibhotla:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.03

Topic: H.03. Decision Making

Title: Effects of Threat Imminence on Learning Under Uncertainty

Authors: *M. SATTI[1,2,3], R. M. CICHY[1], N. W. SCHUCK[3,4], P. DAYAN[5,2,6], R. BRUCKNER[1,3];
[1]Dept. of Educ. and Psychology, Freie Univ. Berlin, Berlin, Germany; [2]Max Planck Sch. of Cognition, Leipzig, Germany; [3]Max Planck Res. Group NeuroCode, Max Planck Inst. For Human Develop., Berlin, Germany; [4]Max Planck UCL Ctr. for Computat. Psychiatry and Ageing Res., Berlin, Germany; [5]Max Planck institute for Biol. Cybernetcs, Tuebingen, Germany; [6]Univ. of Tuebingen, Tuebingen, Germany

Abstract: Survival in non-stationary aversive environments is critically dependent on adapting to their inherent uncertainties. Uncertainties appear in different forms: irreducible variability (expected uncertainty) and systematic but unpredictable environmental changes (unexpected uncertainty). An optimal learner dynamically regulates learning according to both forms by weighing prediction errors using a varying learning rate (LR; which should be higher under unexpected uncertainty). An inability to adjust the LR flexibly can lead to learning biases, one source of which is internalizing disorders like anxiety disorder and depression. An important distinction is made in theoretical models in the anxiety literature (the Threat Imminence Continuum) between anxiety evoked by distal threats and fear evoked by immediate proximal ones. Anxiety and fear might be associated with different degrees of learning. In particular, we hypothesize that the often intentional vagaries of the behaviour of proximal threats eliciting fear demand a high LR, even when other aspects of the environment are stable. This idea has yet to permeate the learning literature, and so we explore adaptive learning under varying levels of threat and uncertainty. We designed a new online game-based behavioral experiment in which human participants save themselves by placing a flaming torch in the path of an attacking predator, but get more points the longer they wait before moving the torch. To succeed, they must therefore learn the arrival location of the predator under expected and unexpected uncertainty. To manipulate threat imminence, three predators attack at different times, corresponding to distal (anxiety), medium, and proximal threat (fear). Data were analyzed using regression and a normative computational model. We first investigated whether participants responded differently to different predators and found that the alacrity with which they moved the torch to protect themselves was modulated by the type of threat encountered. Participants consistently reacted faster to proximal threats; this increased their chances of survival, suggesting that they adaptively respond to threats (Mann-Whitney $U = 0.0$, $p = 0.0002$, two-sided). In line with the hypothesis sketched above, we found an effect of threat type on the LR, with elevated LRs after encountering proximal threats (fear) compared to distal threats (anxiety) ($U = 26$, $p = 0.037$). To conclude, fear induced by immediate threats might raise the subjective unpredictability of threat. Therefore, learning biases might be particularly prominent for proximal compared to more distal threats.

Disclosures: M. Satti: None. **R.M. Cichy:** None. **N.W. Schuck:** None. **P. Dayan:** None. **R. Bruckner:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.04

Topic: H.03. Decision Making

Support: IBS R015-D1
NRF-2019M3E5D2A01060299
NRF-2019R1A2C1085566

Title: Understanding spatial regularity of reward enhances foraging outcome.

Authors: *J. JEON[1,2], S. M. YOO[1,3], W. SHIM[1,3,2];
[1]Ctr. for Neurosci. Imaging Res., Inst. for Basic Sci. (IBS), Suwon, Korea, Republic of; [2]Dept. of Intelligent Precision Healthcare Convergence, [3]Dept. of Biomed. Engin., Sungkyunkwan Univ., Suwon, Korea, Republic of

Abstract: Foraging, a successive action of seeking reward that yields maximum long-term benefit, requires cognitive processes with high ethological relevance. Understanding the spatial regularity of the environment and transferring it into efficient foraging behavior through navigation are fundamental to forming an efficient foraging route. However, classic laboratory studies that abstractly imitate foraging behavior, such as patch-leaving, did not consider spatial information. Here, using an interactive Minecraft-based platform, we developed a 3D foraging task where recognizing spatial regularity is a crucial component of the harvest rate. The task consisted of two stages: 1) regularity detection phase and 2) reward collection phase. In the first phase, subjects navigated the open field with two types of rewards and decided whether rewards of the same type are spatially clustered (structured environment) or different types of rewards are randomly distributed (random environment). In the second phase, subjects either accepted or rejected affordable offers while continuing to navigate the environment and maximizing the final reward based on their previously formed beliefs about spatial regularity. We hypothesized that 1) understanding spatial regularity would require a vector spatial representation of reward distribution rather than a scalar representation of the environment (e.g., ratio of experienced reward type), and 2) once subjects recognize environmental regularity, the frequency of rejecting the suboptimal offers would increase and the foraging route would become closer to optimal. Using the drift-diffusion model that takes the degree of reward clusterness as a piece of evidence, we found that the subjects' decision about environmental regularity was more accurately predicted when reward clusterness incorporates spatial information. We also observed larger variance in the harvest rate across subjects under a structured environment compared to a random one, indicating that accurate beliefs about environmental regularities may encourage a strategic rejection for efficient foraging. To examine whether recognizing spatial regularity increases the rejection of a myopic reward that would harm the long-term harvest rate, we develop agent-based models with different types of constraints. We predict that a systematic rejection pattern would emerge when spatial input is allowed and rejection is used as one of the actions, and that

this adaptive behavior would result in a higher harvest rate than other agents. In sum, this study suggests that integrating environmental regularities into foraging behavior is key to optimal spatial foraging.

Disclosures: **J. Jeon:** None. **S.M. Yoo:** None. **W. Shim:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.05

Topic: H.03. Decision Making

Support: European Research Council Consolidator Grant IndDecision – 865474,
Irish Research Council Government of Ireland Postgraduate Scholarship Programme: GOIPG/2019/808

Title: Neurally informed insights into perceptual learning in decision making tasks

Authors: *C. JUDD[1,2], E. A. CORBETT[1,3], R. G. O'CONNELL[1,2];
[1]Trinity Col. Inst. of Neurosci., [2]Sch. of Psychology, Trinity Col. Dublin, Dublin, Ireland; [3]Sch. of Engin., Univ. Col. Dublin, Dublin, Ireland

Abstract: Objectives and Research Question
Research in psychophysics has indicated that behavioural improvements arising from training on perceptual tasks are attributable to reductions in internal noise. In parallel, mathematical modelling studies within the decision-theoretic framework have tended to point to alternative accounts including increased drift rates and reduced non-decision times and boundary separation. However, these models fix within-trial noise as their scaling parameter, thus preventing it from accounting for any learning effects. EEG derived markers of decision making may be useful in enabling us to inform and constrain decision models and examine changes in within-trial noise over learning.
Materials and Methods
Sixteen participants completed an Equivalent Noise Estimation of internal noise and five blocks of a two-forced choice random-dot motion task across three sessions while 128-channel EEG was recorded.
Results and Conclusions
Participants showed significant increases in accuracy by session, but not within a session, while reaction time reduced over the course of a session, but did not lower significantly across blocks in one day. A significant reduction in psychophysical estimates of internal noise was also seen across sessions. Analysis of the contingent negative variation and beta as markers of urgency suggested significantly more urgency in earlier sessions. Pre-evidence beta adjustments and behavioural estimates of within-trial noise were used to constrain the model. These constraints matched unconstrained model predictions. Constrained models gave good fits but did not

outperform an unconstrained model where noise was allowed to vary by session. Collectively the modelling efforts indicate a reduction in internal perceptual noise alongside more consistent response styles through adapted urgency dynamics as the drivers of the observed learning effects. The methods highlight a novel and potentially impactful approach to the study of perceptual decision making.

Disclosures: C. Judd: None. **E.A. Corbett:** None. **R.G. O'Connell:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.06

Topic: H.03. Decision Making

Support: NSF Award 2043637

Title: Decision-making while walking in risky environments: using insights from behavioral economics

Authors: *S. JAIN, N. SCHWEIGHOFER, J. FINLEY;
USC, Los Angeles, CA

Abstract: Fall-related injuries are a primary health concern, and several risk factors have been targeted to reduce fall risk. While clinical measures can be used to identify the presence of some physiological and psychological risk factors, these measures do not assess behavioral fall risk. Behavioral risk is determined by the decisions that people make when walking in risky environments. These decisions can have devastating consequences, particularly if they lead to risky behaviors. Here, we apply computational models from behavioral economics to people's walking decisions as they walk under the risk of balance perturbations. We first investigated perceptual sensitivity to the direction of perturbations while walking. Participants experienced one trip-like perturbation and one slip-like perturbation, and then chose the one that they would be willing to repeat. While keeping the magnitude of the trip-like perturbations constant, we used an adaptive staircase algorithm to determine the slip-like perturbation that was equally preferred. This was repeated for five magnitudes, 0.2-0.6 m/s about the self-selected walking speed. On average, participants preferred trip-like perturbations that were 0.14 ± 0.04 m/s faster than a given slip-like perturbation. Next, we manipulated the distribution of perturbation magnitudes as participants walked with trip-like perturbations. They walked for two bouts while experiencing perturbations of varying magnitude and then chose the bout that they would be willing to repeat. One bout ("FIXED") had low uncertainty as all the perturbations in it were of equal magnitude, and the other bout ("VARIABLE") had high variability as the perturbations in it could vary in magnitude. We compared three models of decision making in their ability to predict these choices. The Mean-Variance model, which assumes that the subjective value of each walking bout is computed as the mean of the perturbations discounted by the variance in amplitude,

predicted the highest proportion of choices accurately (0.83 ± 0.08). This was followed by Prospect Theory (0.81 ± 0.07), according to which the value of each perturbation is weighted by its perceived probability, and the recency heuristic (0.78 ± 0.08), which proposes that perturbations experienced closer in time to the decision are weighted more heavily than those occurring earlier. Together, these studies will provide insight into how people evaluate and decide between risky walking options. Examining this decision making process and understanding how it is affected by age-related factors will allow us to quantify and potentially mitigate behavioral risk factors in individuals who are at risk of falls.

Disclosures: S. Jain: None. **N. Schweighofer:** None. **J. Finley:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.07

Topic: H.03. Decision Making

Title: The computational mechanisms supporting adaptive decision making during real-world threat

Authors: *J. SIEGEL[1], A. XUE[2], M. N. SHADLEN[4], A. BAKKOUR[5], D. SHOHAMY[3]; [1]Columbia Univ., Brooklyn, NY; [2]Columbia Univ., New York City, NY; [3]Zuckerman Inst., Columbia Univ., New York, NY; [4]Columbia University/HHMI, New York, NY; [5]Dept. of Psychology, Univ. of Chicago, Chicago, IL

Abstract: Efficient and effective decision-making during threatening situations is crucial for navigating environmental challenges, such as a new pandemic. Threatening events trigger autonomic responses that change how we perceive and respond to our environment to prepare us for defensive behavior. However, since autonomic states are often not considered in models of decision making, it's unclear how the mobilization of the systems evolved for navigating threats spill over to influence the computations of decisions in everyday life. Here, we use behavioral experiments and computational modeling to unveil the cognitive computations employed when deciding during real-world environmental threats. At a time when wildfires and hurricanes were at large, we recruited 890 participants to complete a value-based decision-making task and a perceptual decision-making task. Self-report questionnaires measured participants' level of anxiety about being harmed by wildfires and hurricanes and were used to compute a threat-induced anxiety index. Across both tasks, we found that individuals with heightened threat-induced anxiety exhibited more stochastic choices and were significantly faster to make decisions that were especially ambiguous. Crucially, threat-induced anxiety did not impact the efficiency of decisions, suggesting that choice accuracy was not sacrificed by choice speed, as additional time is unlikely to improve accuracy for highly ambiguous decisions. To decompose the cognitive processes that unfold during decision making, we modeled participants' choice and response time distributions using a multi-attribute extension of the drift-diffusion model (DDM).

The model revealed that individuals with heightened anxiety were slower to accumulate evidence about the choice options when decisions were easy, indicating they require more evidence to make unambiguous decisions. Additionally, threat-induced anxiety was associated with lower caution in decision-making as indicated by a lower threshold to reach a decision. Results were consistent across value-based and perceptual decisions and provide initial evidence that the effect of autonomic states on decision making may not be restricted to computations of value. The model and data provide insights into how threat-induced arousal enables efficient decision-making by adapting the rate at which information is integrated and amount of caution exerted depending on choice ambiguity. These mechanisms may have evolved to preserve cognitive resources for threat-related defense behavior, and their breakdown may play a role in the development of maladaptive responses observed in pathological anxieties.

Disclosures: **J. Siegel:** None. **A. Xue:** None. **M.N. Shadlen:** None. **A. Bakkour:** None. **D. Shohamy:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.08

Topic: H.03. Decision Making

Support: 2019M3E5D2A01060293
2021M3E5D2A01019542

Title: Monkey shows an environment-specific choice stochasticity for optimal decision

Authors: *S.-H. KIM[1], Y. CHO[2], H. PARK[3], G. KIM[2], J.-W. SOHN[4];
[1]Intelligent Precision Healthcare Convergence, Sungkwunkwan Univ., Suwon, Korea, Republic of; [2]SungKyunKwan Univ., Suwon, Korea, Republic of; [3]Catholic Kwandong Univ., Gangneung, Korea, Republic of; [4]Med. Sci., Catholic Kwandong Univ., Yeonsu-gu, Korea, Republic of

Abstract: Human and animal often faces problem of exploration and exploitation trade-off (EETO) for decision making in non-stationary environment. Modeling subjects' strategies for EETO has been widely studied. However, simple metric to measure optimal EETO is scarce. The aim of study is to propose a metric to measure EETO and to validate with behavioral data. For this, we trained a rhesus monkey (male, 7.2 kg) with two-alternative forced choice (2AFC) task using joystick in reward foraging environment. One session was composed of five blocks (100 trials per each block) during which reward contingency to given stimuli (red versus blue circular target) was fixed. The reward probability of each target in a given block was pseudo randomly chosen from 0 to 1 in 0.25 steps. The sum of reward probability of each target was one. We collected the behavioral data of 27 sessions. The overall success rate of capturing the target was 88%. Average value of matching slope was 0.793 with linear regression between reward fraction

and choice fraction of the monkey's choices. A slope closer to 1 means that the animal exploitatively chooses stimulus based on past experience. So, we considered the matching slope as an indication of exploitation. Next, as a proxy to exploration degree, we calculated the fraction of win-shift-lose-stay trials to target capture trials. The average fraction was 0.333. The matching slope and the fraction were negatively correlated ($r^2 = 0.614$, $p < 0.01$). This implies that the animal showed EETO. For the further analysis, we defined the multiplication of these two values as a EETO metric. Then, we fitted the animal's behavior with Rescorla-Wagner (RW) model combined with logistic function. The value of each stimulus was estimated in RW model, and the choice of the animal was predicted with a logistic function based on the value. The likelihood of model for the animal's choice was maximized. Three free parameters were used, which were learning rates for positively and negatively reinforcement in RW and the inverse temperature (β) in the logistic function. The average value of β was 5.237. We further investigated relationship between β with the EETO metric by simulation. We simulated animal's choices with the model by changing β from 0 to 100 step 0.1 with 100 repetitions at each β points. We calculated inverse temperature (β_{max}) values that maximized the EETO metric. The β_{max} ranged from 1.8 to 9 in different session. However, there was positive relationship between the β_{max} and β from the animal's actual choices ($r^2 = 0.396$, $p < 0.01$). This result shows that the animal's exploration-exploitation trade-off behavior can be modeled by an environment-specific choice stochasticity.

Disclosures: **S. Kim:** None. **Y. Cho:** None. **H. Park:** None. **G. Kim:** None. **J. Sohn:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.09

Topic: H.03. Decision Making

Support: Wellcome Trust WT101092MA
Medical Research Council MR/K005480/1

Title: Modelling behavioural choices during a wisconsin card sorting task analogue using reinforcement learning

Authors: *M. AINSWORTH[1], J. M. GALEAZZI[2], M. J. BUCKLEY[1];
[2]Exptl. Psychology, [1]Univ. of Oxford, Oxford, United Kingdom

Abstract: Reinforcement learning (RL) models have become an increasingly popular tool to characterise learning during decision-making tasks. Crucially such models allow researchers to quantify behaviourally relevant parameters, (for example cue value) on a trial-by-trial basis, and examine how these values change in response to feedback. Here we use RL models to characterise the behaviour of macaque monkeys whilst they performed a Wisconsin Card Sorting Task Analogue (WCST). On each trial of this task the animals were required to match either the colour or shape of a cue stimuli with one of three subsequently presented targets. The correct

abstract rule changed on a block-by-block basis, and the animals received no cue as to the correct rule. Therefore on each block animals learnt the correct rule by exploring both before determining which to exploit.We compared the behavioural choices predicted by three RL models (RL_{chosen}, $RL_{+random\ noise}$, $RL_{chosen + other}$) with the animals actual choices and show that patterns of explorative errors made by a sub-population of animals are best captured by modelling the value of the chosen and unchosen abstract rule simultaneously. However, these errors were not captured by models in which values decayed during the block or were influenced by the addition of random noise during the block. Finally, we show that the RL model derived learning rate predicted some, but not all of the explorative and perseverative errors made by animals. Our findings confirm the importance of using RL models to compute values on a trial-by-trial basis, and suggest two different behavioural approaches used by sub-populations of animals to perform the WCST. The first group performed the task in an ideal fashion: on each block exploring until determining the correct rule, then exploiting the rule until making an error, at which point they repeated the process. By contrast, a second group appeared to use an alternative strategy in which they used the value of both rules to make behavioural choices.

Disclosures: **M. Ainsworth:** None. **J.M. Galeazzi:** None. **M.J. Buckley:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.10

Topic: H.03. Decision Making

Support: Gatsby Charitable Foundation
Wellcome Trust

Title: Behavioural markers of decision making and uncertainty in continuous memory recall tasks across sensory modalities

Authors: P. VINCENT[1], M. SAHANI[2], *A. AKRAMI[3];
[1]Sainsbury Wellcome Ctr., [2]Gatsby Computat. Neurosci. Unit, Univ. Col. London, London, United Kingdom; [3]Sainsbury Wellcome Ctr., UCL, London, United Kingdom

Abstract: The perception of sensory stimuli is frequently variable. Previous studies have shown that observers account for uncertainty arising from internal variability when they combine sensory cues, integrate sensory input with prior expectation, or select actions under externally imposed cost functions. But how does the distribution of internal uncertainty shape free perceptual report, and are these mechanisms consistent across sensory modalities and statistics? Behavioural models have assumed that unitary percepts may reflect means, modes or samples of internal belief distributions. Here, we show that observers' reconstructions of the remembered orientation of a visual grating correspond to means of the variability-induced likelihood, not the mode or a random sample of their distributional belief. This behaviour remains robust as either

the distribution of stimuli, or the degree of internal variability, change. Furthermore, responses in two similar working memory reconstruction tasks in the auditory domain follow the same pattern, again corresponding to the means of internal belief distributions. However, in the case of sounds, these beliefs are more strongly shaped by the distributions of stimuli used during the experiment.These results suggest that the general rules of decision making under uncertainty are consistent across modalities, but that patterns of adaptation to sensory statistics may differ.

Disclosures: P. Vincent: None. **M. Sahani:** None. **A. Akrami:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.11

Topic: H.03. Decision Making

Support: NRF Grant 2021M3A9E4080784
NRF Grant 2020M3E5D9079910
KBRI Grant 21-BR-03-01
NRF Grant 2019M3E5D2A0106626721
IITP Grant 2019-0-01371

Title: Fronto-striatal decoupling during arbitration between goal-directed and habitual decision-making in obsessive-compulsive disorder

Authors: *T. KIM[1], S. LEE[1], S. LHO[2], S.-Y. MOON[2], M. KIM[2], J. KWON[3];
[1]KAIST, Daejeon, Korea, Republic of; [2]Seoul Natl. Univ. Hosp., Seoul, Korea, Republic of; [3]Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: A theory that excessive habits serve as a building block of compulsions has drawn attention to identifying a brain model underlying habit-bias in obsessive-compulsive disorder (OCD) to bridge the cognitive model and neurotherapeutics. To extend the current understanding that an imbalance in goal-directed versus habitual neural systems underlies habit-bias,[1] it needs to clarify how the imbalance occurs during an arbitration process between the two strategies. When goal-directed learning is deemed to be dominant, the ventrolateral prefrontal cortex (vlPFC) tracking reliability (i.e., probability of successful outcome predictions) of the two strategies downregulates the putamen engaged in habitual learning.[2] We aimed to determine whether this neurocircuit is aberrantly attenuated in OCD during the arbitration. In this study, thirty patients with OCD (age 26.9 ± 6.2 years, 12 females) and thirty healthy controls (age 25.0 ± 4.7 years, 17 females) underwent fMRI scans while simultaneously performing the sequential two-step decision task, designed to observe model-based (goal-directed) and model-free (habitual) learning.[2] We employed a computational model devised to account for the arbitration process, which makes inferences about the reliability of each learning based on the history of its prediction errors and determines that the control is given to a more reliable strategy. We

estimated brain activity encoding variables of the computational model (reliability and action value of each learning) and analyzed psychophysiological interaction effects of the model-choice preference on the vlPFC-putamen coupling. Compared to healthy controls, patients with OCD exhibited a stronger habit-bias (P = .006), attributed to less reliable predictions in goal-directed than habitual learning. When behaviors should be model-based, patients exhibited a weaker strength of the fronto-striatal negative coupling than healthy controls (t = 4.43, P_{FWER} = 0.001). This hypoconnectivity was correlated with more severe compulsivity (r = -0.56, P = 0.001). We suggest that the attenuated top-down control of the habit controller by the prefrontal arbitrator underlies habit-bias in OCD. Enhancing the fronto-striatal connectivity may be a potential neurotherapeutics for compulsivity. **References [1]** Voon V, Derbyshire K, Ruck C, *et al*. Disorders of compulsivity: a common bias towards learning habits. *Mol Psychiatry* 2015;20(3):345-352. **[2]** Lee SW, Shimojo S, O'Doherty JP. Neural computations underlying arbitration between model-based and model-free learning. *Neuron* 2014;81(3):687-699.

Disclosures: **T. Kim:** None. **S. Lee:** None. **S. Lho:** None. **S. Moon:** None. **M. Kim:** None. **J. Kwon:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.12

Topic: H.03. Decision Making

Support: National Institutes of Health-National Institute on Aging R01AG058817
National Institutes of Health-National Institute on Aging R01AG022983
Larry L. Hillblom Foundation (2018-A-006- NET)

Title: Age effects on information processing speed: a drift diffusion model analysis

Authors: *C. SANCHES, C. YOUNG, H. ROMERO-KORNBLUM, W. CHIONG;
Univ. of California San Francisco, Memory and Aging Ctr., San Francisco, CA

Abstract: Declines in cognitive function are one of the most debilitating facets of growing old, and cognitive processing speed (the time it takes to perform a cognitive task) has been shown to decline with age even in healthy older adults. It has been suggested that the information manipulation and decision-making components are the most specific measures of cognitive processing speed. The drift diffusion model has proved useful in distinguishing between different processes involved in task performance, since it allows for the statistical separation of different components of binary decisions, such as decision thresholds to reach a response (*boundary separation*), sensory encoding and motor execution (*non-decision time*) and evidence accumulation and decision-making (*drift rate*). Whereas *boundary separation* and *non-decision time* have been consistently shown to increase with age, results regarding age effects on *drift rate* are inconsistent. We fit this model to the outcomes of three tasks that tap different cognitive

abilities and evaluate the effects of age on these three parameters of the model in each task, focusing on the *drift rate* parameter. Given the hypothesis that processing speed slowing with age is associated with the age-related loss of white matter integrity, we will also explore the relation between age, white matter integrity and *drift rate.* Neurologically healthy older adults (age range 54-95yo, mean = 77yo) were recruited from a cohort that includes in-person behavioral and neuroimaging measurements, and completed different online instruments, including a delay discounting (n = 167), an emotion recognition (n = 195), and a word memory task (n = 173). We will compute the bivariate correlations with age for the diffusion model parameters, separately for each task and establish mediation models with age as the primary predictor variable, *drift rate* as the outcome variable and white matter integrity as the mediator. Our preliminary results suggest that time needed to perform these tasks increases across adulthood. Reaction times in all three tasks were positively associated with age ($r(165) = .32$; $r(193) = .47$; $r(171) = .38$; all $p < 0.001$). Understanding how different components of cognitive processing speed evolve and interact and if they decline generally or specifically in certain domains, will contribute to our understanding of age-related cognitive decline, and might have an impact on possible environmental changes benefiting an optimal performance of everyday tasks by older adults, helping them maintaining their independence and higher quality of life.

Disclosures: C. Sanches: None. **C. Young:** None. **H. Romero-Kornblum:** None. **W. Chiong:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.13

Topic: H.03. Decision Making

Support: Fondation Pour la Recherche Médicale

Title: How incidental moods bias economic decisions

Authors: *R. HEEREMA, P. CARRILLO, F. VINCKIER, M. PESSIGLIONE;
Paris Brain Inst., Paris, France

Abstract: The moods that we experience in everyday life have an impact on our behavior, even when we are doing tasks that should not depend on our affective state. Moods build up over time such that their valence may spill over to proximal decisions we are making. For example, incidental joyful moods may enhance risk-taking, as observed with people buying more lottery tickets when the weather is nicer than expected (Otto, Fleming, & Glimcher 2016). Here, we report a systematic and replicated investigation in 2 lab studies (total *n* = 121 human participants) of mood effects on economic decisions that involve tradeoffs between a monetary reward and one of 4 kinds of costs: a delay, risk, physical effort, or mental effort. Using an established mood induction paradigm where participants play a quiz game, affective states were

successfully manipulated, as shown by subjective ratings, between episodes providing high rates of positive and negative feedbacks about the responses to the quiz questions (Vinckier et al. 2018). Accordingly, preferences differentially shifted throughout these episodes, such that costly but more rewarding options were more often chosen in the positive condition, and less often chosen in the negative condition. When comparing computational models of choice behavior, this effect was best explained as the consequence of an additive decision bias proportional to mood rating, favoring the pursuit of larger rewards, irrespective of the cost. We then used simulations of foraging behavior to identify the environments in which this mood bias would be adaptive. In these simulations, mood is construed as a global estimate of the current cost/benefit tradeoff that the foraging agent learns through a delta update rule. Thus, a high-reward / low-cost action would improve mood, which in turn would favor foraging in the next decision process. We found that, in environments where costs and benefits are auto-correlated, it is optimal to have decisions biased by mood to a certain extent. In real life, the impact of mood on decisions may therefore be particularly adaptive in environments that vary a lot across seasons, with foraging costs being high during winter, and various sources of rewards being abundant during summer (Eldar et al. 2015).

Disclosures: **R. Heerema:** None. **P. Carrillo:** None. **F. Vinckier:** None. **M. Pessiglione:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.14

Topic: H.03. Decision Making

Support: DAAD Graduate School Scholarship Program 2020

Title: Uncertainty-driven integration of visual and reward information for economic decision making and reward learning

Authors: ***P. GANESH**[1,2,3], N. SCHUCK[3], R. M. CICHY[1], C. FINKE[4], R. BRUCKNER[1,3]; [1]Dept. of Educ. and Psychology, Freie universität berlin, Berlin, Germany; [2]Berlin Sch. of Mind and Brain, Humboldt-Universität zu Berlin, Berlin, Germany; [3]Max Planck Res. Group Neural and Computat. Basis of Learning, Memory and Decision Making, Max Planck Inst. for Human Develop., Berlin, Germany; [4]Charite - Universtätsmedizin Berlin, Charite Universtätsmedizin Berlin, Berlin, Germany

Abstract: When making choices, humans consider both reward and visual properties of the available options. For example, imagine picking a restaurant, which could be governed by the experienced quality of the dishes (reward-guided choice) and the décor of the restaurant (visually-guided choice). Here we investigate what drives this integration of visual and reward information during economic decision making and learning. We hypothesize that uncertainty is

one pivotal factor that determines the extent to which reward and visual properties drive adaptive behavior. In particular, (H1) under low reward uncertainty conditions (i.e., low uncertainty about which option yields the most reward, such as picking a restaurant in a familiar neighborhood), choices might primarily be driven by the options' expected reward values. However, under high reward uncertainty conditions (i.e., uncertainty about which option leads to better outcomes, such as picking a restaurant in a new city), the visual features of an option (like salience) may influence one's choice. Furthermore, (H2) when learning from reward feedback, the speed of learning might be driven by uncertainty about the options' perceptual features. If options are perceptually distinct, learning might be faster than under higher perceptual uncertainty when choice options cannot reliably be distinguished. To test these hypotheses, we combined computational modeling and a perceptual uncertainty-augmented bandit task in which human participants make economic choices and learn the reward values of the available choice options. Across two behavioral experiments (total N &=199) and in line with our hypotheses, we show that (regarding H1) choice behavior is particularly biased towards visually salient options when reward uncertainty is high, as compared to choices under lower reward uncertainty. This bias might be ecologically beneficial when visual salience acts as a proxy for an option's expected reward, thus aiding decision-making under reward uncertainty. Moreover, (regarding H2), we show that higher perceptual uncertainty attenuates reward learning. This could help avoid incorrect assignment of received rewards to options that cannot be clearly identified due to perceptual uncertainty. Taken together, our study provides new insights into how humans dynamically combine perceptual and reward information. Our results suggest that humans adaptively utilize visual salience in the service of economic decision making and learning and highlight the potential ecological and normative role of using visual salience in conjunction with reward values for adaptive behavior.

Disclosures: **P. Ganesh:** None. **N. Schuck:** None. **R.M. Cichy:** None. **C. Finke:** None. **R. Bruckner:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.15

Topic: H.03. Decision Making

Title: Synaptic depression with short time constant enable adaptation in primary auditory cortex with long recovery time constant

Authors: *S. QIU[1], S. KOERNER[2], T. TEICHERT[2], C. HUANG[1];
[1]Siwei Qiu, Univ. of Pittsburgh Dept. of Neurosci., Pittsburgh, PA; [2]Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Across the auditory pathway neural responses are significantly attenuated if the same stimulus was presented less than a few seconds ago. The time constant at which firing rates in

primary auditory cortex (PAC) recover back to baseline is approximately two seconds. This response attenuation has often been linked to short-term synaptic depression. However, synaptic time constants are typically well below 1 second. It is thus unclear if and how the synaptic time-constants could give rise to the much longer time-constants of firing rates. To address this question, we combine recordings from monkey primary auditory cortex with a theoretical approach to investigate under which circumstances the firing rates of a neural network can recover with different time-constants than the underlying synapses. We first explored this idea by fitting a mechanistic Wilson-Cowan model with short-term synaptic plasticity to artificial data from a descriptive model that allowed us directly specify firing rate time-constants and the dependence of firing rate r on stimulus intensity I ($r=I^a$). For feedforward synaptic depression, we found that the rate recovery time constant can be longer (equal or shorter) than the synaptic time constant if the r-I relation is supralinear (linear or sublinear). For recurrent synaptic depression, rate recovery time constants are longer than synaptic time constants, except when the r-I relation is supralinear. We then defined a metric that quantified the convexity/concavity of the relationship between firing rate and the depression variable x (r-x relation) as the area between the unity line and r-x curve. This metric is positively correlated with the rate-to-synapse time constant ratio. Based on these theoretical results, we fit the Wilson-Cowan model to the multi-unit activity data of monkey. We found that the rate-to-synapse time constant ratio is constrained within a narrow distribution peak at 1.4, indicating that the firing rates in auditory cortex recover more slowly than synapses. Our findings show that at least some of the discrepancy between rate recovery and synaptic time-constants can be explained by the non-linear relation between firing rate and the depression variable x.

Disclosures: **S. Qiu:** None. **S. Koerner:** None. **T. Teichert:** None. **C. Huang:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.16

Topic: H.03. Decision Making

Support: European Union's Horizon 2020 Framework Programme for Research and Innovation 945539

Title: A theoretical formalization of consequential decision-making: Modelling complex brain dynamics

Authors: *G. CECCHINI[1], E. BASPINAR[2], M. DEPASS[1], A. DESTEXHE[3], R. MORENO-BOTE[4], I. COS[1];
[1]Dept. of Mathematics and Computer Sci., Univ. of Barcelona, Barcelona, Spain; [2]CNRS/NeuroPSI, Inst. des Neurosciences, Univ. Paris-Saclay, Paris, France; [3]CNRS, CNRS, Saclay, France; [4]Ctr. for Brain and Cognition, Univ. Pompeu Fabra, Barcelona, Spain

Abstract: How is the knowledge of consequence incorporated into decision-making? Despite the vast literature on reward driven decision-making, the neural computations underlying the assessment of option complexity and their potential consequence on decision-making processes remains to be well understood and properly formalized.
To study the role of consequence in a decision-making framework, we developed a novel task, in which consequence could be quantified and its effect measured on the decision-making policy. This task targets not only the notion of consequence between consecutive trials but allows the study of how consequence is learned over time. This task was performed by thirty-four human participants, instructed to perform sequences of binary decisions aiming at maximizing cumulative reward. In brief, at each trial the participants had to select between two visual stimuli each associated with a specific reward. The size of the stimuli depended on the participants' previous decisions (the consequence).
In a more theoretical framework, we investigated how the knowledge of consequence is incorporated into reward-driven decision-making model This model is based on an approximation of the neural dynamics of two populations and is capable of learning sequences of binary decisions adaptively to maximize cumulative reward. It incorporates complexity, visual stimuli discrimination and predicted consequence into its dynamics in a parsimonious fashion. Furthermore, it also implements an internal oversight mechanism of consequence, and of learning by reinforcement.
We fitted this model to the experimental data as part of a global validation process, and carefully investigated the role of each model parameter. We found that specific parameters needed to be fit with specific metrics from the data, such as reaction times and accuracies. Remarkably, our model was capable of faithfully predicting both the non-trivial inhibitory patterns of decision-making, as well as the sequences of decisions of each individual, regardless of their level of accuracy throughout the experimental session.
In conclusion, we offer a novel approach that explains how consequence could be incorporated into the neural dynamics of decision-making, and how adaptation may occur to this end. Furthermore, our theoretical characterization of this process was able to properly describe vastly different decision-making policies by varying a few parameters.

Disclosures: **G. Cecchini:** None. **E. Baspinar:** None. **M. DePass:** None. **A. Destexhe:** None. **R. Moreno-Bote:** None. **I. Cos:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.17

Topic: H.03. Decision Making

Title: Machine learning to discover the governing dynamical mathematical equations underlying perceptual decision-making

Authors: *B. LENFESTY, K. WONG-LIN, S. BHATTACHARYYA;
Univ. of Ulster, Ulster Univ., Derry, United Kingdom

Abstract: Most computational modelling studies on perceptual decision-making identify computational model parameters based on fitting choice behavioural data, and to a lesser extent, fitting neural dynamics. However, no study has made use of data-driven approaches to elucidate the underlying dynamical mathematical equations generating the observed neural dynamics, which can be nonlinear in nature. In this work, we make use of a data-driven algorithm called sparse identification of nonlinear dynamics (SINDy), originally developed to discover governing physical equations from measurement data, to discover the dynamical equations of various stochastic two-choice decision-making models based on simulated data. In particular, we collated simulated data from the standard drift-diffusion model (DDM), the linear leaky competing accumulator (LCA) model and its DDM-approximated version, and a nonlinear dynamical model endowed with working memory. Model parameters were estimated by applying the SINDy algorithm to the neural dynamics. To reduce noise effects, we proposed an averaging method in which model parameters were estimated across trials. After model parameters were estimated, neural dynamics and choice behaviour (accuracy and decision time) of the deduced model were simulated and compared with that of the original model. We found that SINDy could readily identify the linear LCA model parameters for across-trial and single-trial conditions, especially for higher signal-to-noise ratio. This was expected as SINDy was originally developed for deterministic dynamical systems. In contrast, for the rest of the models, as signal-to-noise ratio increased, the SINDy-derived models' behaviour became more dissimilar to that of the original respective models. To understand this, we determined the root-mean-square error for these models' parameters and found strong positive correlation with the model's choice behaviour discrepancy for the DDM and nonlinear model. However, this was not the case for the DDM-approximated LCA model; as signal-to-noise ratio increased within the reaction time task (a first-passage time framework), the dynamics generated by the original models ramp up too fast towards decision thresholds, thus revealing less data and SINDy becoming less accurate in elucidating the underlying governing dynamics. Taken together, we showed for the first time, the potential and limitation of SINDy applied to stochastic first-passage time problem for decision-making. Further investigations include applying to fuller neural dynamics with different decision task paradigm and exploring other averaging approaches.

Disclosures: B. Lenfesty: None. **K. Wong-Lin:** None. **S. Bhattacharyya:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.18

Topic: H.03. Decision Making

Support: NSFC Grant 31871101

Title: Real-time sequential decisions under temporal expectation

Authors: *M. WANG[1], J. LI[1,2], H. ZHANG[1,2,3];
[1]Sch. of Psychological and Cognitive Sci., [2]PKU-IDG/McGovern Inst. for Brain Res., [3]Peking-Tsinghua Ctr. for Life Sci., Peking Univ., Beijing, China

Abstract: Most laboratory studies on temporal expectation focus on how people can prioritize attention across time to prepare for specific future events. In such situations, the decision process that links temporal expectation to the final action is usually considered trivial. In real life, however, temporal expectation can involve more complicated decisions, where people must plan their actions based on the consequences of their past actions as well as the expectations of future events. Here we developed a real-time gambling task to simulate such decision situations and compared human performance to those of three computational models to understand the underlying decision process.
Methods: On each trial of the experiment, human participants (N = 31) were asked to press keys to make real-time predictions on the time of target onset. The closer their last key press was to the time of target onset, the higher the reward. Participants could press keys for multiple times, but every press incurred one unit of cost. Their net gain would be the reward minus the total cost. We manipulated the temporal distribution of target onsets in 4 different blocks. Apparently, participants should increase their key presses when the target is expected to be more likely to occur. Meanwhile, according to the rewarding rule, the marginal gain of a second press would be small or even negative when it is close to the first press. In other words, to maximize expected gain, participants should make decisions based on their own action history as well as their expectations of near future. To understand participants' decision process, we constructed a gain-maximizing ideal observer whose response probability at each moment takes into account both temporal expectation and action history. As alternative hypotheses, we also considered two heuristic models, whose response probability is simply determined by instantaneous temporal probability. We fit the three computational models to the timing of participants' key responses and compared the goodness-of-fit of models.
Results: (1) The timing pattern of participants' key responses varied with the temporal distribution of target onsets, implying the influence of temporal expectation. (2) The ideal observer model outperformed the two heuristic models in predicting the response patterns (the summed ΔAICc between the ideal observer model and the second-best model was -3925.8; according to the group-level Bayesian model selection, the protected exceedance probability > 99.9%). It suggests that participants were not just responding proportionally to the temporal probability at the moment, but integrated their past actions and future prospects to make decisions.

Disclosures: **M. Wang:** None. **J. Li:** None. **H. Zhang:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.19

Topic: H.03. Decision Making

Support: HBP SGA3, EU H2020 Research and Innovation Programme, Grant 945539

Title: Learning to infer transitively: ranking symbols on a mental line in premotor cortex

Authors: *S. RAGLIO[1,2], G. DI ANTONIO[1,4], E. BRUNAMONTI[3], S. FERRAINA[3], M. MATTIA[1];
[1]Natl. Ctr. for Radiation Protection and Computat. Physics, Inst. Superiore di Sanità, Rome, Italy; [2]PhD Program in Behavioral Neurosci., [3]Physiol. and Pharmacol., La Sapienza Univ. di Roma, Rome, Italy; [4]PhD Program in Applied Electronics, Roma Tre Univ., Rome, Italy

Abstract: Transitive inference (TI) is a form of deductive reasoning that allows to infer unknown relations among premises. It is believed that the task is cognitively solved resorting to a mental linear workspace, namely the mental line, in which the stimuli are mapped according to their arbitrary assigned rank. This means that, if one experiences that A>B and B>C, the relationship A>C can be transitively inferred as after learning the items A, B and C are properly located on adjacent positions along such linear workspace. An open question is whether this mental line is encoded somewhere in the brain and, if so, where and how. Here, we investigated the possible role of the dorsal premotor cortex (PMd) in representing the hypothesized mental line during the acquisition of the relations between items, eventually leading to successfully perform a TI task. Two rhesus monkeys were tested requiring selecting the higher ranked item, presented alternatively on the left or right position of a computer monitor while the neural activity of the PMd was recorded simultaneously by 96 probes. We analyzed the multi-unit neural activity (MUA) by relying to a mathematical framework in which it is possible to carry out the needed mental line as a linear combination of the representations of the stimuli/items in the neural state space. The applicability of this theoretical model relies on the hypothesis that both the identity and the spatial position of the stimuli are encoded in the probed network. As a first result, we found that PMd represents such information in its neural activity together with a correlate of the difficulty in motor decision. According to the model, we found that the PMd representations of the stimuli, once projected on the theoretical mental line (a linear neural subspace), are predictive of the motor decision.Finally, we found striking evidence that representations of both the stimuli and the motor plan are plastic, as they change in time according to the behavioral output. A realignment of the mental line thus results leading to an increasing overlap with the axis decoding the motor responses. Our results then provide evidence that a TI task can be solved as a linear transformation of the neural representations of arbitrarily ranked stimuli. PMd appears to have a leading role in manipulating such representations, efficiently transforming the ordinal knowledge of the stimuli relations into the motor output decision.

Disclosures: S. Raglio: None. **G. Di Antonio:** None. **E. Brunamonti:** None. **S. Ferraina:** None. **M. Mattia:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.20

Topic: H.03. Decision Making

Support: NIH R01NS113078
NSF 1707398
NSF 1707398
GAT3708

Title: Representational geometry correlates with behavioral differences across two monkeys

Authors: *V. FASCIANELLI[1], F. STEFANINI[1], S. TSUJIMOTO[2], A. GENOVESIO[3], S. FUSI[1];
[1]Zuckerman Institute, Columbia Univ., Columbia Univ., New York, NY; [2]Nielsen Neurofocus, Tokyo, Japan; [3]Dept. di Fisiologia e Farmacologia, Sapienza Univ., Roma, Italy

Abstract: Our ability to perform cognitive tasks critically depends on the geometry of the neural representations. High dimensional representations allow downstream neurons to implement a large number of input-output functions. In contrast, low dimensional representations, which typically require a process of abstraction, allow for better generalization. An interesting compromise between these two types of geometries would be to dynamically adjust the geometry depending on task demands. Using analytical tools recently developed to study abstraction, we observed that the geometry of the recorded neural representations correlates with behavior and changes rapidly during a trial.
We studied the correlation between representational geometry and monkeys' behavior in the dorsolateral prefrontal cortex(PFdl) of two monkeys performing a visually cued strategy task. Two different pairs of visual stimuli cued the repetition of the previous response (stay strategy) and the shift to the alternative response (shift strategy). The representational geometry of the relevant task variables differed between monkeys. Indeed, during the cue presentation, the visual cue was represented in abstract format in the first monkey, and the strategy in the second monkey. Interestingly, these different geometries correlated with different reaction times of the two monkeys. To better investigate the difference in behavior, we built a model to predict the reaction times on a single trial basis. We found that the visual cue has a stronger weight in predicting the reaction time of the monkey with the visual cue in abstract format than the other one. On the opposite, the strategy has a stronger weight in predicting the reaction time of the monkey with the strategy in abstract format that the other one. To investigate the underlying computations that could explain the differences in the neural representation, we trained an artificial neural network to perform the task. We observed a change of the representational geometry of the task variables along the training epochs, that matched the different neural representations across monkeys in two different ranges of training epochs. Our analysis reveals important computational properties and interesting differences in neuronal representations between two monkeys performing the same task with high performance. This difference is mirrored by the different geometry of the neural representation of two task variables: the visual cue and the strategy. Moreover, the geometry can modulate the reaction time of the monkeys and it can be highly dynamic to reflect cognitive demands.

Disclosures: **V. Fascianelli:** None. **F. Stefanini:** None. **S. Tsujimoto:** None. **A. Genovesio:** None. **S. Fusi:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.21

Topic: H.03. Decision Making

Support: NIH R01 NS091010
NIH R01 DC018545
NIH R03 MH120426
NIH R01 EY025349
NIH P30 EY02259
NSF 1940181
NIH F31 MH116613

Title: Diverse behavioral timescales encoded in retrosplenial cortex explain hyperbolic behavior

Authors: *B. P. DANSKIN[1], R. HATTORI[1], Y. E. ZHANG[1], M. C. AOI[1,2], T. KOMIYAMA[1,2,3];
[1]Univ. of California San Diego, Univ. of California San Diego, La Jolla, CA; [2]Halicioglu Data Sci. Inst., La Jolla, CA; [3]Ctr. for Neural Circuits and Behavior, La Jolla, CA

Abstract: Animals rely on their experience to guide their next choice. In foraging-type tasks guided by history-dependent value, these experiences are integrated such that the weights of past events initially decay quickly over time but show a longer tail than expected by exponential decay, which is better described by a hyperbolic function. Hyperbolic integration affords sensitivity to both recent environmental dynamics and long-term trends, however the mechanism by which the brain implements this hyperbolic integration is unknown. We trained mice on a history-dependent, value-based decision task and found that the mice indeed showed hyperbolic decay on their weighting of past experience. However, the activity of history-encoding cortical neurons showed weighting with exponential decay. In resolving this apparent mismatch, we observed that cortical neurons encode history information heterogeneously across a wide variety of time-constants, with the retrosplenial cortex (RSC) overrepresenting longer time-constants than other areas. A model that combines these diverse timescales can recreate the heavy-tailed, hyperbolic-like behavior. In particular, time-constants of RSC neurons best matched the behavior, and optogenetic inactivation of RSC uniquely reduced the use of history information. These results indicate that behavior-relevant history information is maintained in neurons across multiple timescales in parallel, and suggest RSC is a critical reservoir of this information guiding decision-making.

Disclosures: **B.P. Danskin:** None. **R. Hattori:** None. **Y.E. Zhang:** None. **M.C. Aoi:** None. **T. Komiyama:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.22

Topic: H.03. Decision Making

Support: NIH Grant R01DA048096 (KTK)
NIH Grant R01MH121099 (KTK)
NIH Grant R01MH124115 (KTK)

Title: Selective attention via reinforcement learning, optimal decision making, and dopamine?

Authors: *A. R. SHIPP[1], J. HUMPHRIES[2], J. TRATTNER[2], K. KISHIDA[3];
[1]Dept. of Physiol. and Pharmacol., [2]Dept. of Physiol. and Pharmacology, Neurosci. Grad. Program, [3]Dept. of Physiol. and Pharmacology, Neurosci. Grad. Program, Dept. of Neurosurg., Wake Forest Univ. Sch. of Med., Winston Salem, NC

Abstract: Traditionally, selective attention has been considered under the principal that limits in information processing bandwidth and related energetic constraints motivates the determination of environmental features saliency. While these and related "bottleneck" concerns are important, evolutionary processes may have discovered ways to efficiently allocate resources via optimal learning advantages. Dayan and colleagues (2000) proposed such an approach whereby considerations from optimal learning theory underlie efficient selective attention behavior. Their algorithm requires *a priori* knowledge about the statistical structure of reinforcers in the environment. In particular, an estimate of the variance of reinforcement must be known, which is used to augment 'reward prediction error' based updating of feature values. In contrast, more recent work has suggested that extracellular dopamine may encode the 'precision' of stimulis' ability to engender action, which the authors operationally define as 'salience' (Friston et al., 2015). This latter work considers dopamine's role from a descriptive Bayes-optimal behavioral framework, and in so doing proposes a theory apparently intended to contrast to the longstanding reinforcement learning and optimal decision-making theories that frame dopamine as a reinforcement prediction error signal.We propose a middle ground where dopamine's established role as a reward prediction error signal (in reinforcement learning and optimal decision-making theories) can be used by an organism as an estimate of the variance, which would be equivalent to the inverse precision in the Bayes-optimal behavioral framework. Our approach largely follows Dayan and colleagues' framework but uses a 'temporal difference reinforcement learning framework to estimate of the standard deviation (σ) of the expected value of stimuli that lead to reward. This error term is then used to provide an estimate of the precision ($1/\sigma 2$) which adjusts the learning rate and differentially weights stimuli that may anticipate reward.To investigate the efficay of our approach, we ran simulations of feature-based Pavlovian

conditioning comparing traditional TDRL versus our selective attention augmented TDRL (SA-TDRL). In SA-TDRL, value estimates and learning rates are dynamically augmented using feature-level precision estimates, which results in larger relative value weights for features more certain to lead to reward and changes in learning rate as a function of stimulus reliability. Future work will compare this algorithm to human data to test the hypotheses that human dopamine levels augment behavior in a manner consistent with SA-TDRL.

Disclosures: **A.R. Shipp:** None. **J. Humphries:** None. **J. Trattner:** None. **K. Kishida:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.23

Topic: H.03. Decision Making

Support: NRF Grant 2021M3E5D2A0102249311

Title: Medial prefrontal cortex activity mediates social stress-induced decrease in disadvantageous inequality aversion

Authors: *K. KIM[1], J.-H. LEE[2], W.-Y. AHN[2,3], H. KIM[1];
[1]Sch. of Psychology, Korea Univ., Seoul, Korea, Republic of; [2]Dept. of Psychology, [3]Dept. of Brain and Cognitive Sci., Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: It has been reported that social stress can have contradictory effects on social behavior, increasing prosocial or antisocial behaviors. Such findings may be attributed to the modulatory role of stress in context-dependent value computation because cortisol hormone affects a wide range of neural regions including the medial prefrontal cortex (MPFC). In this study, by incorporating stress treatment with a modified social discounting task, we investigated the neural mechanism underlying the association between the physiological responses to social stressors and inter-subject variability in money-sharing behavior. 41 male participants performed the Montreal imaging stress task (MIST), in which they were asked to solve arithmetic problems under time pressure and received negative social feedbacks. About 20 minutes after stress onset, samples of their saliva were collected to measure changes in cortisol concentration from the baseline. Then, participants performed a modified social discounting task, which was designed to measures money sharing with targets of varying social distance when money sharing results in inequal (i.e., disadvantageous inequal condition) or equal (i.e., equal condition) consequence. We used computational modeling and hierarchical Bayesian analysis to identify the three distinctive dimensions of motivation: 1) the sensitivity to target's social distance, 2) the sensitivity to reward for self, and 3) the degree of disadvantageous inequality aversion as measured by the tendency of avoiding an option that gives more money to a target than to self. Behavioral results showed a negative correlation between the degree of increase in cortisol concentration and the degree of disadvantageous inequality aversion. The degree of increase in cortisol concentration

was negatively correlated with the degree of the dorsomedial prefrontal cortex (DMPFC) activity in tracking the value of reward for self in the disadvantageous inequal condition. In turn, those with higher degree of disadvantageous inequality aversion exhibited higher degree of DMPFC activity and lower degree of VMPFC activity in tracking the value of reward for target in the disadvantageous inequal condition. Finally, a mediation analysis confirmed that the association between the degree of increase in cortisol level and the degree of disadvantageous inequality aversion was indirectly mediated by the DMPFC activity. These findings suggest that the DMPFC plays a key role in promoting disadvantageous inequality aversion, which can be interrupted by socially induced stress, resulting in self-protective prosocial motivation.

Disclosures: K. Kim: None. **J. Lee:** None. **W. Ahn:** None. **H. Kim:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.24

Topic: H.03. Decision Making

Support: NIH Grant 1R01NS118461-01

Title: Controlled Arousal Predicts Epochs of Optimal Sensory Discrimination

Authors: *D. HULSEY, K. ZUMWALT, L. MAZZUCATO, D. A. MCCORMICK, S. JARAMILLO;
Inst. of Neurosci., Univ. of Oregon, Eugene, OR

Abstract: Rapid fluctuations in neural dynamics correlate with measures of pupil size, facial (e.g. whisker) movements, and locomotion in mice. These measures also broadly correlate with task performance, but their relationships vary by study. Recent modeling work finds that mice employ distinct performance strategies within a training session, and spontaneously alternate between them across minutes. Based on these findings, we hypothesized that arousal related measures may form predictable relationships with task performance states, and covary with their transitions.
To investigate these questions we implemented head-fixed two alternative choice tasks requiring either auditory or visual stimulus categorization to receive a water reward. We fit hidden Markov models with states characterized by generalized linear models to task data. Using cross validated test sets we determined an ideal number of states and fit a model for each subject. Six stereotypic states emerged across models, with each model including an optimal (choices guided by stimulus) and disengaged (no response to any stimulus) state, and a variable number of left or right biased states. During task performance we measured pupil diameter, whisker pad motion energy, and locomotion speed. We found a clear relation between the probability of being in the optimal task performance state and each of these measures for both sensory modalities. Task performance had a consistent inverted-U relationship with pupil diameter across subjects, with

optimal performance state more likely at intermediate pupil diameters. The ideal pupil diameter was lower for subjects performing the auditory compared to the visual task. Movement measures had more influence on optimal occupancy for visual task subjects than those performing the auditory task. Next, we quantified changes in behavioral measures at transitions into and out of the optimal state. While no consistent changes were apparent in the raw behavioral measures, there was a marked decrease in trial to trial variability of measures upon entering the optimal state, and an increase in variability upon exiting it. Using a cross-validated classification analysis, we found that occurrence of optimal and suboptimal task performance states can be predicted by changes in pupil diameter and movements with high accuracy.
Based on these results we conclude that pupil size, facial movement, and locomotion measures can reliably indicate transitions in task engagement states. Decreased variability of these measures during optimal engagement states may further suggest active control of arousal to maintain optimal brain states for task performance.

Disclosures: D. Hulsey: None. **K. Zumwalt:** None. **L. Mazzucato:** None. **D.A. McCormick:** None. **S. Jaramillo:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.25

Topic: H.03. Decision Making

Support: R00MH111926
Alfred P. Sloan Research Fellowship, Klingenstein-Simons Fellowship Award in Neuroscience
5T90DA043219
2T32MH019524

Title: Distinct controllers for motivation and deliberation

Authors: *A. MAH[1], V. BOSSIO[3], C. M. CONSTANTINOPLE[2];
[2]Ctr. for Neural Sci., [1]New York Univ., New York, NY; [3]Columbia Univ., New York, NY

Abstract: A principle of modern decision theories is reference dependence, in which the subjective value of a reward is computed relative to an internal reference point that is thought to reflect expectations of future rewards. Reinforcement learning describes how agents can estimate reference points through experience and can be generally divided into two classes: “model-free” algorithms which repeat previously rewarded actions, and “model-based” algorithms which learn a world model and use it to select actions. Distinct neural circuits implement these algorithms, and a central question is how an agent can perform “meta-control” - how to decide which system to use and when. We used high-throughput training to collect well-powered datasets from hundreds of rats (n=211) performing a temporal wagering task with semi-observable states

(blocks of large or small rewards) distinguishable only by their latent reward statistics. Rats normatively adjust how quickly they initiated trials (motivation) and how long they waited for rewards (deliberation) in response to varying reward statistics. However, motivation and deliberation displayed distinct dynamics around block transitions, suggesting that these systems have distinct reference points that differentially respond to change in the environment. Statistical modeling revealed that motivation reflects a model-free algorithm, while deliberation reflects a model-based algorithm. These results provide a finer timescale resolution of meta-control and makes testable hypotheses about interactions among model-free and model-based neural circuits during complex behavior.

Disclosures: A. Mah: None. **V. Bossio:** None. **C.M. Constantinople:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.26

Topic: H.03. Decision Making

Support: NIH Grant RF1DA055666
Swartz Foundation
Alfred P. Sloan Foundation Research Fellowship

Title: Reconciling population dynamics- and neural circuits perspectives on the context-dependent decision-making.

Authors: *P. TOLMACHEV, C. LANGDON, T. A. ENGEL;
Engel's Lab., Cold Spring Harbor Lab., Cold Spring Harbor, NY

Abstract: The ability of animals to select alternative responses to the same stimulus depending on context is a hallmark of cognitive flexibility. A plausible mechanism for selection of the relevant stimuli relies on inhibition of the irrelevant sensory responses, as hypothesized in neural circuit models. This mechanism, however, is difficult to reconcile with neural responses observed in higher cortical areas during flexible behavior, where representations of the irrelevant stimuli are still present, posing an apparent contradiction with the inhibitory mechanism. Studies using recurrent neural networks (RNNs) suggested that heterogeneous neural responses reflect a nonlinear dynamical process unfolding at the population level, such that, within a given context, only the inputs aligned with a specific “selection vector” drive the activity along the line attractor. The selection via population dynamics yields a fundamentally distinct perspective on representations of irrelevant stimuli than neural circuit models based on inhibition. Yet, it is unknown what circuit structure gives rise to the dynamical selection mechanism at the population level. Therefore, it remains unclear whether the dynamical selection-vector mechanism is qualitatively different from the inhibitory circuit mechanism. In this work, we show that the inhibitory circuit mechanism can give rise to the selection of relevant stimuli via

population dynamics. We explicitly construct an interpretable neural circuit, which flexibly selects alternative stimulus-response associations by inhibiting the irrelevant stimuli. First, we show that the line attractors in the state space—the key components of the dynamical selection mechanism—are present in the engineered interpretable circuit. Second, we find that the circuit also relies on a dynamical selection vector mechanism. The selection vector has a concrete realization as an excitatory connection between two nodes in the circuit. Finally, we illustrate that the same picture is preserved if the small circuit is embedded in a higher dimensional RNN. The activity of the circuit nodes corresponds to the distributed activity patterns in the RNN. These results demonstrate that the population-level dynamical mechanism for context-dependent behavior is not fundamentally distinct from previously hypothesized circuit mechanisms based on inhibition. This work links the dynamical-systems approach to cognition with the underlying circuit structure, opening new possibilities for causal manipulations of the neural networks to validate these mechanisms in experiments.

Disclosures: P. Tolmachev: None. **C. Langdon:** None. **T.A. Engel:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.27

Topic: H.03. Decision Making

Support: NSFC Grant 31871101

Title: The alternation of human temporal beliefs between different possibilities

Authors: *S. CHEN[1], M. WANG[1], H. ZHANG[1,2,3];
[1]Sch. of Psychological and Cognitive Sci., [2]PKU-IDG/McGovern Inst. for Brain Res., [3]Peking-Tsinghua Ctr. for Life Sci., Peking Univ., Beijing, China

Abstract: Our perceptual and motor systems are smart planners for the future, allocating resources adaptively across time according to how likely an event will occur at different moments. However, it is largely unknown how the relevant temporal uncertainty is represented, whether it is an integrative representation reflecting the probabilities over different temporal possibilities (the integration hypothesis) or alternates from trial to trial with each trial focusing only on one temporal possibility (the alternation hypothesis). To differentiate between these two hypotheses, here we introduced a new behavioral task to manipulate human temporal beliefs and investigated its effects on simple response times (RTs). In two experiments (N = 20 and N = 16), human participants made speeded key response to the onset of a visual target. The duration before target onset (SOA) varied from trial to trial, which could be 2 s or 6 s for Experiment 1 and 1 s, 1.5 s, 2 s, 2.5 s, 3 s, 3.5 s, or 4s for Experiment 2. In half of the trials, before the start of the speeded response task, participants were also required to predict whether the SOA of the incoming target would be relatively short or long. Each participant completed multiple blocks

that differed in the odds of short- to long-SOA trials (1:3, 1:1, or 3:1 for Experiment 1 and 1:3 or 3:1 for Experiment 2). Participants were not instructed the temporal structures of the experiments but received feedbacks about their predictions as well as speeded responses. We examined how participants' prediction on a trial might influence the RT on the same trial and constructed four integration- or alternation-based computational models to explain these effects. Results: (1) Participants' probabilities to predict short and long in each block matched the corresponding true probabilities. (2) These cue-free, self-initiated predictions influenced the subsequent RTs in a way mimicking traditional cue-based temporal expectations, which provides evidence that people can spontaneously alternate between different temporal beliefs. (3) An alternation-based model with post-decision bias best fit participants' RT patterns (mean, variance, and skewness), which suggests that not only prediction-elicited beliefs but also the prior beliefs before predictions may alternate between the early and late temporal categories, further supporting the alternation hypothesis.

Disclosures: S. Chen: None. **M. Wang:** None. **H. Zhang:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.28

Topic: H.03. Decision Making

Support: European Research Council consolidator grant ERC-CoG-2014-646696

Title: Auditory perceptual and confidence decisions in humans and artificial neural networks

Authors: *L. FRANZEN[1,2], L.-M. SCHMITT[1,3], C. ANDREOU[1,2], J. OBLESER[1,2];
[2]Ctr. of Brain, Behavior and Metabolism, [1]Univ. of Lübeck, Lübeck, Germany; [3]Donders Inst. for Brain, Cognition and Behaviour, Radboud Univ., Nijmegen, Netherlands

Abstract: Our daily environment is inherently noisy, and we rarely have full certainty about the auditory percept we just heard. Perceptual decisions, such as whether I hear a human scream or wind (type I decision), come with an inherent metacognitive evaluation of the accuracy of this decision, which can be reported as confidence (type II decision). Both decisions are believed to be based on the accumulation of sensory evidence towards a decision boundary. This process may also result in frequent misperceptions, such as confidently reporting a human voice when no voice was present. Such hallucinations are a hallmark of schizophrenia and do also occur among the normal population. However, the underlying cognitive and neurobiological processes that affect our confidence in healthy and psychiatric populations remain elusive. We used a large-sample behavioural online experiment (N=216), a concordant laboratory study including recordings of EEG and pupillometry data (N=30), and multi-layered convolutional neural networks (CNNs) with different architectures. Both humans and machines were presented with sounds at low vs. high levels of noise and reported if and with how much confidence they

perceived a voice. In the online study, we found that trials presenting more sensory evidence resulted in higher false alarm rate, type I sensitivity (d'), confidence ratings, and metacognitive efficiency (meta-d'/d'). For all voice trials, while type I accuracy was lower, confidence was higher. Lastly, higher individual hallucination proneness was linked to faster confidence response times. What are the computational underpinnings explaining this behavioural bias towards more incorrect perceptual decisions of high confidence when presented with a voice? An initial CNN analysis showed that separately accumulating evidence for type I and II decisions benefits metacognitive efficiency, while shared evidence accumulation benefits perceptual sensitivity. This trade-off between perceptual and confidence decision-making serves as a basis for modelling observed behavioural effects more precisely by additionally introducing connections between type I and II routes at different hierarchical processing stages akin to human cortical architecture. In sum, our results demonstrate a conservative decision strategy in difficult environments, with fewer decisions towards perceiving a human voice. However, confidence appears decoupled from this auditory decision strategy, suggesting separate evidence accumulation routes.

Disclosures: **L. Franzen:** None. **L. Schmitt:** None. **C. Andreou:** None. **J. Obleser:** None.

Poster

564. Biological and Computational Models of Decision Making

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 564.29

Topic: H.03. Decision Making

Support: AFOSR Award FA9550-20-1-0106

Title: Identifying neurocomputational correlates of perceptual confidence in decision-making regions using fMRI in humans

Authors: *S. ABACHI, B. MANISCALCO, M. A. K. PETERS;
Cognitive Sci., Univ. of California, Irvine, Irvine, CA

Abstract: Perceptual decisions are typically accompanied by metacognitive evaluation. These confidence judgements usually covary with decisional accuracy, but sometimes this correspondence breaks down, e.g. in atypical environments or clinical populations. This observation raises an important question: what are the neural computations of perceptual metacognition if their output can diverge from perceptual decisions themselves? In a recent paper, we argued that tuned inhibition—i.e., the degree to which a neuron is inhibited by neighboring neurons with opposing tuning preferences, which differs from neuron to neuron—is a crucial part of the underlying mechanism[1]. Using fMRI in humans, here we sought to validate this model of tuned inhibition within decision-making areas at the voxel level. Participants completed a visual perception task, viewing random dot kinematograms with varying levels of dot motion energy and conflict (motion in opposing directions). We characterized the inhibition

tuning level of voxels within target areas based on BOLD responses to these stimuli, then compared voxels' inhibition levels to their predictive power for subjects' motion-direction choices and confidence ratings. Importantly, we compared these findings to simulated BOLD responses from an extended version of the previously-published computational model[1]. Results demonstrate that our paradigm provides a viable mechanism for identifying inhibition tuning in decision-making regions at the voxel level, and—critically—that inhibition tuning levels can explain the degree to which a voxel contributes to confidence precisely in accordance with model predictions. Our findings support our tuned inhibition model describing the neural computations underlying perceptual metacognition.

1. Maniscalco et al. (2021). PLoS Computational Biology.

Disclosures: S. Abachi: None. **B. Maniscalco:** None. **M.A.K. Peters:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.01

Topic: H.03. Decision Making

Support: Academy of Finland
Sigrid Juselius Foundation
University of Helsinki
MPI for Biological Cybernetics
Joachim Herz Foundation
Alexander von Humboldt-Stiftung

Title: Distinct anterior cingulate neurons drive changes-of-mind and monitor past performance

Authors: *D. VASILEV[1], O. VINOGRADOV[3], R. IWAI[4], A. LEVINA[3], N. K. TOTAH[2]; [2]Helsinki Inst. of Life Sci., [1]Univ. of Helsinki, Helsinki, Finland; [3]Univ. of Tübingen, Tübingen, Germany; [4]MPI for Biol. Cybernetics, Tuebingen, Germany

Abstract: When choosing between two actions, sensory evidence is accumulated toward one of the choices until a threshold is reached. Sensory neuron spike rate has been shown to track evidence accumulation. However, subjects will occasionally change their mind (switching from one action to the other), which is accompanied by a reversal in sensory neuron activity. It is unknown what neurons activate during a change-of-mind (CoM) potentially driving it. One candidate is anterior cingulate cortex (ACC), which has been shown in human EEG studies to respond around a CoM. Here, we for the first time directly report CoM in a rodent model. Head-fixed rats (N=43) were trained to discriminate stimuli by running on a treadmill past a distance threshold (Go) or remaining immobile (NoGo). On some NoGo stimulus trials, rats began running shortly after NoGo stimulus onset but made a CoM and chose to return to immobility before crossing the threshold. The time of CoM was defined by peak velocity of the treadmill,

when the rat stopped running and began slowing down toward immobility. We tested the hypothesis that reversal from a higher velocity Go response to the NoGo response (immobility) would be associated with a larger ACC single unit response during the CoM. We recorded 574 ACC single units. On CoM trials, we found that stimulus-evoked responses of units tuned to the Go stimulus instead responded to the NoGo stimulus, suggesting engagement of the incorrect stimulus-response mapping in ACC. During the subsequent CoM, 20% of units scaled their activity with movement reversal size; moreover, a demixed PCA analysis showed that movement reversal size explained a large amount of variance in the population activity. Separately, 13% of units responded similarly but at the end of the trial and apparently quantified the magnitude of conflict between competing actions occurring earlier in the trial. In addition, whole-cortex 32-electrode EEG translationally linked rat and human CoM by identifying a frontal response during CoM. Our results support current theories that the ACC monitors past conflict between competing actions and significantly broaden the role for ACC by showing that ACC neurons may drive CoM.

Disclosures: **D. Vasilev:** None. **O. Vinogradov:** None. **R. Iwai:** None. **A. Levina:** None. **N.K. Totah:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.02

Topic: H.03. Decision Making

Support: IBS-R002-A1

Title: Role of vasoactive intestinal polypeptide (VIP)-expressing neurons in the prefrontal cortex in probabilistic reversal learning

Authors: J. YI[1,2], ***Y. YOON**[3,1,2], S. CHOE[1], M. JUNG[3,1];
[1]Ctr. for Synaptic Brain Dysfunctions, Inst. for Basic Sci., Daejeon, Korea, Republic of; [2]These authors contributed equally, Daejeon, Korea, Republic of; [3]Dept. of Biol. Sci., Korea Advanced Inst. of Sci. and Technol., Daejeon, Korea, Republic of

Abstract: The prefrontal cortex (PFC) plays a crucial role in flexible control of behavior. Of various types of neurons in the PFC, VIP-expressing neurons are thought to exert powerful influences on PFC circuit operations by disinhibitory control of other inhibitory interneurons. Here, to obtain insights on the role of VIP-expressing neurons in flexible control of behavior, we investigated modulation effects and activity dynamics of VIP-expressing neurons in the medial PFC (mPFC) in adult male mice performing reversal learning under a probabilistic classical conditioning paradigm. Chemogenetic (n = 7) and optogenetic modulation (n = 5) of VIP-expressing neurons in the mPFC significantly impaired reversal learning. Calcium imaging (n = 10) revealed diverse patterns of activity dynamics of mPFC VIP-expressing neurons during

reversal. As a population, VIP-expressing neurons conveyed strong signals related to reward prediction error (RPE) during, but not before or after, reversal learning. These results suggest that VIP-expressing neurons may modulate mPFC neural activity in an RPE-dependent manner, thereby contributing to flexible control of behavior.

Disclosures: J. Yi: None. **Y. Yoon:** None. **S. Choe:** None. **M. Jung:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.03

Topic: H.03. Decision Making

Support: NIH Grant K08MH116125

Title: The activity in the anterior cingulate cortex is required for effort-based decision making

Authors: *A. Q. KASHAY, J. Y. CHEUNG, R. N. VAKNALLI, M. J. DELANEY, M. B. NAVARRO, Jr., C. JUNAIDI, M. E. NEUWIRTH, A. QI, F. VEENKER, M. UMAGUING, S. A. WILKE;
UCLA, Los Angeles, CA

Abstract: Effort-based decision making requires weighing predicted gains against effort costs and is disrupted in depression, schizophrenia, addiction and Parkinson's disease. The anterior cingulate cortex (ACC) is postulated to control effort-based action selection, but its precise functional role is poorly understood. In rodent studies, lesions of the ACC and ventral striatum (VS) produce characteristic deficits in spatial effort-based decision making tasks. However, temporally precise and cell-type specific methods of manipulating neural activity have rarely been applied to study effort-based decision making. To address this deficit, our lab developed and validated a mouse version of the barrier T-maze task, in which mice must choose between climbing a barrier for a large reward or taking a direct path to a small reward. We then used optogenetics to silence ACC excitatory neurons at specific times during this task. Bilateral inhibition of ACC, immediately prior to a choice, rapidly and reversibly impaired preference to exert greater effort for a larger reward when a less rewarded, low effort alternative was available. Silencing ACC-to-VS projection neurons also impaired the selectionof high effort choices, but only transiently when effort-reward tradeoffs had recently changed. These results establish a causal relationship between ACC activity and effortful action selection, and suggest the existence of functionally distinct ACC projection neuron subpopulations.

Disclosures: A.Q. Kashay: None. **J.Y. Cheung:** None. **R.N. Vaknalli:** None. **M.J. Delaney:** None. **M.B. Navarro:** None. **C. Junaidi:** None. **M.E. Neuwirth:** None. **A. Qi:** None. **F. Veenker:** None. **M. Umaguing:** None. **S.A. Wilke:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.04

Topic: H.03. Decision Making

Support: NIH Grant R01AG061888
NSF Grant IIS-1703340
Undergraduate Biology Research Program at the University of Arizona

Title: The effects of time horizon and guided choices on explore-exploit decisions in rodents

Authors: S. WANG[1], B. GERKEN[2], J. WIELAND[2], R. WILSON[2], ***J.-M. FELLOUS**[3];
[1]NIH, Natl. Inst. of Mental Hlth. (NIMH), North Bethesda, MD; [2]Psychology, [3]Psychology and Biomed. Engin., Univ. of Arizona, Tucson, AZ

Abstract: Humans and animals have to balance the need for exploring new options with exploiting known options that yield good outcomes. This tradeoff is known as the explore-exploit dilemma. One key factor in explore-exploit decisions is the time horizon, i.e., the number of known future choices remaining which can be influenced by the current decision. Horizon adaptation is thought to be a hallmark of effective exploration. Recent studies showed that humans were able to adapt the extent of their directed and random exploration with the time horizon. Yet apart from one early study in birds, very little work has investigated how animals explore under different time horizons.
To better understand the neural mechanisms underlying how humans and animals address the explore-exploit dilemma, a good animal behavioral model is critical. Most previous rodent explore-exploit studies used ethologically unrealistic operant boxes and reversal learning paradigms in which the decision to abandon a bad option is confounded by the need for exploring a novel option for information collection, making it difficult to separate different drives and heuristics for exploration. In this study, we investigated how rodents make explore-exploit decisions using a spatial navigation Horizon Task adapted to rats to address the above limitations. In this task, rats were asked to choose between two options that gave out different number of drops of sugar water. The reward size from one of the two options is known to the rat, whereas the reward size of the other option is unknown. We assess how rats "explore" the unknown option as a function of time horizon, i.e., the number of choices they have in a game. We compared the rats' performance to that of humans using identical measures. We used hierarchical Bayesian analysis to quantify directed exploration and random exploration for both humans and rats. We showed that like humans, rats use prior information to effectively guide exploration. In addition, like humans, rats use information-driven directed exploration, but the extent to which they explore has the opposite dependence on time horizon than humans. Moreover, we found that free choices and guided choices have fundamentally different influences on future exploration in rodents, a finding that has not yet been tested in humans. This study reveals that the explore-exploit spatial behavior of rats is more complex than previously thought.

Disclosures: **S. Wang:** None. **B. Gerken:** None. **J. Wieland:** None. **R. Wilson:** None. **J. Fellous:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.05

Topic: H.03. Decision Making

Support: NIMH R01-MH115030

Title: Distributed processing for value-based choice by prelimbic circuits targeting anterior-posterior dorsal striatal subregions

Authors: *K. CHOI[1], E. PIASINI[3], E. DÍAZ-HERNÁNDEZ[1], L. CIFUENTES-VARGAS[2], N. T. HENDERSON[1], E. N. HOLLY[1], M. SUBRAMANIYAN[1], C. R. GERFEN[4], M. V. FUCCILLO[1];
[1]Dept. of Neurosci., [2]Grad. group of Neurosci., Univ. of Pennsylvania, Philadelphia, PA; [3]Neural Computation Lab, Intl. Sch. for Advanced Studies (SISSA), Triesta, Italy; [4]Natl. Inst. of Mental Hlth. (NIMH), Bethesda, MD

Abstract: Fronto-striatal circuits have been extensively implicated in the cognitive control of behavioral output for both social and appetitive rewards. The functional diversity of prefrontal cortical populations is strongly dependent on their synaptic targets, with control of motor output in part mediated by connectivity to dorsal striatum. Despite evidence for functional diversity along the anterior-posterior axis of the dorsomedial striatum (DMS), it is unclear how distinct fronto-striatal sub-circuits support neural computations essential for value-based choice. Here we identify prefrontal populations targeting distinct DMS subregions and characterize their functional roles. We first performed neural circuit tracing to reveal segregated prefrontal populations defined by anterior/posterior dorsomedial striatal target. The parallel nature of these pathways was evident both from afferent input biases and unique local synaptic connectivity within striatum. We probed the functional relevance of these parallel circuits via *in vivo* calcium imaging and temporally precise causal manipulations during a feedback-based 2-alternative choice task. Single-photon imaging revealed circuit-specific representations of task-relevant information with prelimbic neurons targeting anterior DMS (PL::A-DMS) robustly modulated during choices and in response to negative outcomes, while prelimbic neurons targeting posterior DMS (PL::P-DMS) encoded internal representations of value and positive outcomes contingent on prior choice. Consistent with this distributed coding, optogenetic inhibition of PL::A-DMS circuits strongly impacted choice monitoring and responses to negative outcomes while perturbation of PL::P-DMS signals impaired task engagement and strategies following positive outcomes. Together our data uncover novel PL populations engaged in distributed processing for value-based choice.

Disclosures: **K. Choi:** None. **E. Piasini:** None. **E. Díaz-Hernández:** None. **L. Cifuentes-Vargas:** None. **N.T. Henderson:** None. **E.N. Holly:** None. **M. Subramaniyan:** None. **C.R. Gerfen:** None. **M.V. Fuccillo:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.06

Topic: H.03. Decision Making

Title: The infra-slow brain activity influences behavior in conditions of uncertainty

Authors: *A. DE DIEGO AJENJO, J. KAUR, R. BERG, M. MOLDOVAN, U. GETHER, A. SØRENSEN, D. WOLDBYE;
Copenhagen Univ., København N, Denmark

Abstract: The infra-slow brain activity influences behavior in conditions of uncertainty
Rhythmic neuronal activity at frequencies <0.1 Hz, referred to as infra-slow activity, has been observed across wide brain areas in humans and rodents. It has been suggested in humans that infra-slow activity can modulate neuronal activity to an extent that can affect behavior. The mouse prefrontal cortex plays an important role in functions associated with goal-directed behavior and cognition. We investigated in mice the relationship between infra-slow activity and behavior. For that purpose, we developed an imaging-behavioral system that could assess this relationship in mice performing the continuous performance test (CPT), where adult male mice learned to press a touchscreen during correct stimulus presentation (hits), followed by reward delivery. Then, a calcium indicator (GCaMP6f) was adenovirally expressed in either pyramidal neurons or interneurons from the medial prefrontal cortex (MPFC). We performed imaging recordings by fiber-optics, using confocal laser endomicroscopy, synchronized with CPT events. The interaction between the phase of the infra-slow activity and the amplitude of faster event-related activity at the time of these events, which depicted infra-slow activity modulation, was quantified. We compared the changes of this interaction in the MPFC during hits versus when touchscreen pressing occurred during distracter stimuli presentation (mistakes). To stimulate uncertainty in responses, reward contingencies were altered by inducing anosmia and/or introducing unrewarded hits in sequence or randomly. We found that infra-slow activity influenced event-related neuronal activity in the MPFC during the CPT. During baseline recordings, the infra-slow activity modulation was larger at the time of mistakes. Following conditions of uncertainty, the susceptibility to infra-slow activity modulation changed, being higher at the time of hits and lower at the time of mistakes. We suggested a model that could explain these complementary changes in infra-slow activity modulation under conditions of uncertainty. Our study proposed that infra-slow activity acts as a source of noise at the MPFC level, to an extent that can influence behavior in conditions of uncertainty.

Disclosures: **A. De Diego Ajenjo:** None. **J. Kaur:** None. **R. Berg:** None. **M. Moldovan:** None. **U. Gether:** None. **A. Sørensen:** None. **D. Woldbye:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.07

Topic: H.03. Decision Making

Support: NIH R01NS119813
R21MH125107

Title: Noradrenergic dynamics in the sensory and prefrontal cortices during a perceptual decision making task

Authors: *C. SLATER[1,2], K. YU[1], B. YUAN[1], M. KANN[1], T. LANTIN[1], Y. LIU[1], J. S. KIM[1], R. SALDANHA[1], J. FENG[3,4], G. LI[3,4], Y. LI[3,4], Q. WANG[1];
[1]Biomed. Engin., [2]Vagelos Col. of Physicians and Surgeons, Columbia Univ., New York, NY; [3]Sch. of Life Sci., Peking Univ., Beijing, China; [4]PKU-IDG/McGovern Inst. for Brain Res., Beijing, China

Abstract: In a perceptual decision making task, sensory information is accumulated over time in the central nervous system, eventually leading to a decision to choose between alternatives and the subsequent generation of motor commands that indicate the choice made. The perceptual decision making process involves both sensory and prefrontal cortices and is heavily influenced by neuromodulatory systems, such as the noradrenergic system. However, noradrenergic dynamics in the sensory and prefrontal cortices and their association with behavioral outcomes remain poorly understood. Using a genetically encoded norepinephrine (NE) fluorescent biosensor, we simultaneously measured NE dynamics in the primary somatosensory cortex (S1) and the prefrontal cortex (PFC) in mice performing a tactile decision making task. Pupil dynamics were also measured throughout the behavioral sessions. In the task, head-fixed mice were trained to make decisions within a window of opportunity to respond to whisker deflections resulting from a brief air puff with various pressures. In idle mice, we found that NE dynamics were tightly correlated with pupil fluctuations. Despite the relative synchronization between NE levels in the PFC and S1, there were slight differences in the ability of pupil size to index regional NE activity. The response probability of our behaving mice increased monotonically with the intensity of air puffs. Consistent with our previous findings, whisker stimulation induced pupil dilation, and pupil baseline differed across different behavioral outcomes. Relative changes in NE following stimulus presentation were larger and possessed a higher rate of increase between baseline and peak states in the PFC as compared with S1. Interestingly, NE dynamics differed little between stimuli of different strengths in both brain regions, though NE dynamics differed dramatically between trials with different behavioral outcomes. Taken together, our results suggest that the NE dynamics encode an animals' decision to action and to a

lesser extent the sensory stimuli, and this encoding is different between S1 and PFC.
Q.W. and C.S. designed the study. C.S., K.Y., and Y.Liu. performed experiments and analyzed data. B.Y., M.K., T.L., J.S.K, and R.S. performed experiments. J.F., G.L. and Y. Li produced the GRAB_NE biosensor for the study.

Disclosures: **C. Slater:** None. **K. Yu:** None. **B. Yuan:** None. **M. Kann:** None. **T. Lantin:** None. **Y. Liu:** None. **J.S. Kim:** None. **R. Saldanha:** None. **J. Feng:** None. **G. Li:** None. **Y. Li:** None. **Q. Wang:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Financial interest in Sharper Sense.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.08

Topic: H.03. Decision Making

Support: 22-CoE-BT-03
NRF-2018M3C7A1022310

Title: Modulation of auditory discrimination performance of autism spectrum disorder mice triggered by optogenetic activation of medial prefrontal cortex to primary auditory cortex circuit

Authors: *Y. KIM[1], J. KIM[1], J. LEE[2], H. CHOE[1];
[1]Dept. of Brain Sci., [2]Brain Sci. Res. Ctr., Daegu Gyeongbuk Inst. of Sci. and Technol. (DGIST), Daegu, Korea, Republic of

Abstract: Autism spectrum disorder (ASD) is a developmental disability characterized by social deficit and repetitive behavior. In addition, intellectual disability, common comorbidity to ASD core symptoms, aggravates the caregivers' burden by hampering cognitive therapies. To devise a neuromodulation procedure that can rescue learning deficits in the ASD, we first addressed the learning deficits in the ASD mouse model using a go/no-go based pure tone-discrimination task. Cntnap2 knockout mice, a mouse model of ASD, exhibited significant retardation in learning with a similar plateau learning curve to wildtype controls. The prefrontal cortex is known to be important in attention and decision making during the discrimination task while the sensory cortex also shows distinct activity during sensory discrimination learning. Fiber photometry analysis of top-down attention control mediated by the anterior cingulate cortex (ACC) to the primary auditory cortex (Au1) revealed that population calcium transient in the ASD model is abnormally regulated during tone discrimination. In wildtype, the manipulation of ACC to Au1 neurons by optogenetic stimulation bidirectionally enhances or decreases discrimination performance in wildtype mice. This observation inspired us to optogenetically stimulate the ACC-Au1 circuit of ASD during the learning. The optogenetic activation of ACC-Au1 projecting neurons indeed enhances learning efficiency to a level similar to non-stimulated

wildtype. In summary, we report that the manipulation of ACC-Au1 neurons bidirectionally modulates discrimination performance. Also found regulation of ACC-Au1 neurons decreases the learning deficit of ASD and possibly suggests its therapeutic potential for intellectual disabilities of ASD.

Disclosures: **Y. Kim:** None. **J. Kim:** None. **J. Lee:** None. **H. Choe:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.09

Topic: H.03. Decision Making

Support: NIH MH112688

Title: Decision-making strategies are modulated by environmental complexity and mediated by prelimbic cortex

Authors: S. L. HOFFMAN, *U. MUGAN, A. D. REDISH;
Dept. Neurosci, Univ. of Minnesota, Minneapolis, MN

Abstract: Prelimbic Cortex (PL) plays an important role in decision-making strategies and PL disruption causes errors in decision-making. We examined the effects of PL disruption using h4MDi DREADDs under a CAMKIIa promoter in both male and female rats running a spatial decision-making task (active, n = 6; control, n = 6). Rats ran through a central path and made a decision at a choice point (CP) to go left (L) or right (R) for food reward. Reward contingency (L rewarded; R rewarded; rewarded for alternating) changed twice through each 45 minute daily session. The central maze pattern was changed parametrically to modulate the topological complexity of the maze. Rats encountered the same environment for two consecutive days, receiving either agonist (DCZ), or vehicle (saline) on day 1 and saline on day 2.
Control rats performed better than active animals throughout the experiment, and improved their performance on day 2 of encountering the same environment. On day 1, under PL disruption, rats explored less of their environment ($p=0.01$), had more stereotypical paths, and showed less vicarious trial and error (VTE) behavior ($p=0.0002$). On day 2, recovering from PL disruption, active virus rats showed increased VTE behavior ($p=0.001$) and increased exploration ($p=0.07$), suggesting reduced learning from the previous day. After contingency changes, rats showed a decrease in task performance ($p<0.001$), increased exploration ($p<0.001$) and increased VTE behaviors ($p<0.0001$), suggesting a switch to deliberative strategies when reward contingency changed. In more complex environments, which likely require an increased cognitive load, animals explored more ($p=0.0001$) and had less stereotypical paths. Exploratory behavior progressively decreased ($p<0.0001$) through the maze. Average velocity was low at the start of the maze and the choice point, but increased ($p<0.0001$) as animals traversed the maze.
These results suggest that PL is crucial for determining decision-making strategies in complex

and changing environments and that complex environments promote deliberative behaviors while slowing the development of procedural behaviors.

Disclosures: S.L. Hoffman: None. **U. Mugan:** None. **A.D. Redish:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.10

Topic: H.03. Decision Making

Support: NIH Build EXITO UL1GM118964

Title: The role of medial prefrontal cortex vasoactive intestinal peptide interneurons during reward processing

Authors: *B. MARXMILLER[1,2], A. SONNEBORN[3], A. I. ABBAS[4];
[1]Oregon Hlth. and Sci. Univ., Portland, OR; [2]Biol., Portland State Univ., Portland, OR; [3]Behavioral Neurosci., Oregon Hlth. & Sci. Univ. Behavioral Neurosci., Portland, OR; [4]Dept. of Behavioral Neurosci., Vaporhcs/Oregon Hlth. & Sci. Univ., Portland, OR

Abstract: To maximize chances of survival in natural environments, an animal must be able to readily switch between different strategies to obtain food or water when contexts change. This type of reward-based strategy selection is heavily dependent on the medial prefrontal cortex (mPFC), which constantly monitors reward prediction error information, and implements a different strategy when the desired reward does not meet expectations. The circuit mechanisms by which mPFC incorporates contextual and reward information to choose the appropriate strategy are largely unknown, but one potential candidate mechanism involves vasoactive intestinal peptide interneurons (VIP-INs) in the mPFC. These inhibitory interneurons target both pyramidal cells and other interneuron subtypes, which may put them in a good position to suppress existing rule representations and allow for new ones to emerge. Furthermore, these VIP-INs are highly active around positive and negative reward outcomes, suggesting that they can interact with the canonical reward circuitry in some way. Therefore, we set out to study the contribution of VIP-INs in a reward-switching task. Selective optogenetic inhibition of VIP-INs within mPFC resulted in abnormal switching behavior when the reward switches locations. This abnormal behavior correlated with reduced synchrony between the mPFC and ventral tegmental area (VTA), along with aberrant mPFC single unit activity in response to reward outcomes. These results indicate that VIP-INs are involved in helping the mPFC select the appropriate strategy to maximize rewards in dynamic and uncertain environments.

Disclosures: B. Marxmiller: None. **A. Sonneborn:** None. **A.I. Abbas:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.11

Topic: H.03. Decision Making

Support: NIDA (RO1 DA047870)
UCLA Life Sciences Retention Fund (Izquierdo)
NSF GRFP (Aguirre)

Title: Roles of ventrolateral orbitofrontal cortex, basolateral amygdala, and anterior cingulate cortex in flexible stimulus-based learning

Authors: *C. G. AGUIRRE[1], J. L. ROMERO-SOSA[1], J. H. WOO[4], J. MUNIER[2], J. PEREZ[1], G. EDLER[1], M. G. GOLDFARB[1], K. DAS[1], M. G. GOMEZ[1], T. YE[1], J. PANNU[2], P. R. O'NEILL[3], I. SPIGELMAN[2], A. SOLTANI[4], A. IZQUIERDO[1];
[1]Psychology, [2]Section of Biosystems and Function, Sch. of Dent., [3]Shirley and Stefan Hatos Ctr. for Neuropharmacology, Dept. of Psychiatry and Biobehavioral Sci., UCLA, Los Angeles, CA; [4]Dept. of Psychological and Brain Sci., Dartmouth Col., Hanover, NH

Abstract: Reversal learning (RL), impacted in various neuropsychiatric disorders, measures subjects' ability to form flexible associations between cues/stimuli and reward. The contribution of orbitofrontal cortex (OFC), basolateral amygdala (BLA), and anterior cingulate cortex (ACC) to RL and its dependence on the nature of association (i.e., stimulus-based), the modality (i.e., visual), and uncertainty associated with the reward contingencies (i.e., deterministic vs. probabilistic) has not been extensively studied in rats. Here we examined the roles of ventrolateral OFC (a subregion not as often probed as MO and LO), BLA, and ACC in the flexible learning of stimulus-based associations using chemogenetic manipulation. Male and female Long-Evans rats (N=39, 20 females) were prepared with bilateral inhibitory hM4Di DREADDs or eGFP on a CaMKIIa promoter targeting the major output neurons of each region. Rats first met a discrimination criterion, before being tested on both fully predictive deterministic (100/0) and probabilistic (90/10) reversals, during which they selected a visual stimulus associated with a sucrose reward by nosepoking a touchscreen. Thirty minutes prior to each reversal session rats were administered clozapine-N-oxide (CNO) or vehicle (VEH) solution (3mg/kg, i.p.), using a within-subject, counterbalanced design. Specifically, if a rat received CNO on the 1st reversal, it was administered VEH on the 2nd reversal (CNO1-VEH2), or vice versa (VEH1-CNO2). Only animals with confirmed bilateral targeting were included in the behavioral analyses, and efficacy of CNO to reduce activity in these regions was confirmed by ex vivo calcium imaging in slice. Rats learned the initial stimulus discrimination in 8.4 ± 1.5 sessions to 75% for hM4Di and 8.7 ± 0.6 sessions for eGFP. Generalized Linear Models were conducted for each brain region and included all factors (reversal number, virus, drug, drug order, sex) for every measure. Preliminary analyses revealed significant reversal number and/or drug order interactions for learning measures (i.e., probability of choosing the better option, rewards collected), attentional (i.e., initiation latencies), and motivational (i.e., reward latencies) measures for all brain regions. Although post-hoc analyses are ongoing, we found that CNO1-

VEH2 animals exhibit poorer learning and less adaptive strategies compared to VEH1-CNO1 animals following vlOFC and ACC inhibition, but not BLA inhibition. Engagement of vlOFC and ACC during the first experience of a reversal was crucial as it set how future adjustments occurred, consistent with the theorized roles for frontocortical regions in expected uncertainty.

Disclosures: **C.G. Aguirre:** None. **J.L. Romero-Sosa:** None. **J.H. Woo:** None. **J. Munier:** None. **J. Perez:** None. **G. Edler:** None. **M.G. Goldfarb:** None. **K. Das:** None. **M.G. Gomez:** None. **T. Ye:** None. **J. Pannu:** None. **P.R. O'Neill:** None. **I. Spigelman:** None. **A. Soltani:** None. **A. Izquierdo:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.12

Topic: H.03. Decision Making

Support: F31 DA055447
R01 MH117103
R01 DA044297
P50 OD011132

Title: Melanocortin-4 receptor control of striatal-dependent action selection

Authors: *E. C. HEATON, S. L. GOURLEY;
Pediatrics, Emory Univ., Atlanta, GA

Abstract: Goal-directed action refers to behaviors that are dynamic, sensitive to unexpected events, and require the dorsomedial striatum (DMS). Molecular factors underlying an organism's ability to flexibly shift between goal-directed and habitual behavior are incompletely understood. We recently discovered that dorsal striatal melanocortin-4 receptor (*Mc4r)* expression correlates with this behavioral flexibility in adult male and female mice, leading to the hypothesis that the activity of *Mc4r*+ DMS neurons regulates goal-directed action. Stimulation of *Mc4r*+ DMS neurons via Gq-coupled Designer Receptors Exclusively Activated by Designer Drugs (DREADDs) facilitated animals' ability to select actions based on reward likelihood. Meanwhile, inhibition via Gi-DREADDs rendered animals insensitive to changes in reward likelihood, promoting habits. MC4R controls GluA2 AMPA receptor subunit availability at the membrane such that *increasing* MC4R activity should *decrease* glutamatergic excitability of MSNs and mimic the behavioral effects of Gi-DREADDs in *Mc4r*+ neurons. Indeed, administration of MC4R agonist setmelanotide facilitated habit formation, while viral-mediated *Mc4r* knockdown in the DMS enhanced the ability of mice to select actions based on reward likelihood. The DMS receives dense glutamatergic projections from the orbitofrontal cortex (OFC), a region necessary for goal-directed action. Initial trans-synaptic retrograde tracing indicates that DMS *Mc4r*+ neurons receive monosynaptic projections from the OFC, and chemogenetic experimentation

revealed that the OFC is necessary for MC4R-related changes in behavioral flexibility. These results reveal that striatal MC4R may be a key factor in sustaining *versus* "breaking" habits, and thus could serve as a target for treating maladaptive habits that contribute to neuropsychiatric disease.

Disclosures: E.C. Heaton: None. **S.L. Gourley:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.13

Topic: H.03. Decision Making

Support: Canada Institutes of Health Research (CIHR) Grant 156070

Title: Investigating ventral hippocampal to prefrontal pathways involved in approach-avoidance decision-making using a two-alternative, forced-choice operant conflict task

Authors: *J. KATES[1], S. CHEN[2], B. CAVDAROGLU[2], A. C. H. LEE[2], R. ITO[2,1];
[1]Cell & Systems Biol., [2]Psychology, Univ. of Toronto Scarborough, Scarborough, ON, Canada

Abstract: Aberrant approach-avoidance decision-making is a defining feature of several common psychiatric and neurological illnesses (e.g., anxiety). A key aspect of such decision-making - approach-avoidance conflict (AAC) resolution - occurs when an organism must choose between approaching or avoiding stimuli that signal both rewarding and aversive outcomes simultaneously. Mounting evidence in human and rodent literature demonstrates that the ventral hippocampus (vHPC) is critical in regulating AAC decision-making. Moreover, the infralimbic (IL) and medial orbitofrontal cortices (mOFC) - two regions which regulate goal-directed behaviour and encode reward outcomes - are innervated by the vHPC. However, it is currently unclear how vHPC-prefrontal efferents might moderate AAC resolution. This exploratory investigation aims to determine the role of vHPC-prefrontal circuitry in AAC resolution using a novel, two-alternative forced-choice operant task. Adult, male Long-Evans rats were trained to choose between a small reward (sucrose pellet, i.e., reward-only option) versus a larger reward (two sucrose pellets) paired with varying foot-shock intensities (i.e., conflict option). Efferents from the vHPC to the IL or mOFC were inhibited in two separate cohorts (n = 7) using a dual-viral, projection-specific chemogenetic approach (via modified human muscarinic receptor, hM4Di, expression); a third cohort (n = 8) expressed GFP/mCherry as a control. Preliminary results, analyzed using a linear mixed model, suggest that chemogenetic vHPC-IL inhibition via clozapine-N-oxide (CNO) administration increases preference for the conflict option, whereas vHPC-mOFC inhibition does not. The additional use of anxiety assays (e.g., elevated plus maze), which incorporate elements of AAC, finds no effect of CNO administration. Further analysis of choice preference as well as response-times were performed using computational drift-diffusion modeling. This analysis suggests that increasing shock intensity associated with the conflict

option increases the amount of evidence required to formulate AAC decisions; CNO administration, conversely, has no impact on evidence criterion. Taken together, these findings imply differential roles of vHPC projections to the prefrontal cortex in mediating approach-avoidance conflict behaviour, while the use of drift-diffusion modeling increases translatability to human literature.

Disclosures: **J. Kates:** None. **S. Chen:** None. **B. Cavdaroglu:** None. **A.C.H. Lee:** None. **R. Ito:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.14

Topic: H.03. Decision Making

Support: MEXT/JSPS KAKENHI JP 19H03550
MEXT/JSPS KAKENHI JP 19H05224
MEXT/JSPS KAKENHI JP 22H04762
AMED (JP21zf0127004)
Nishinomiya Basic Research Fund, Japan
Takeda Science Foundation
Takahashi Industrial and Economic Research Foundation

Title: A neural basis of decision-making under conflicting predictions of reward and punishment

Authors: K. YOSHIDA[1], M. CHAN[1], Q. LI[2], L. K. PEDRAZA[2], R. O. SIERRA[2], M. MINAMI[1], A. BERENYI[2,4,3], ***Y. TAKEUCHI**[1,2];
[1]Dept. of Biopharmaceutical Sci. and Pharm., Hokkaido Univ., Sapporo, Japan; [2]Dept. of Physiol., Univ. of Szeged, Szeged, Hungary; [3]HCEMM-SZTE Magnetotherapeutics Res. Group, Univ. of Szeged, University of Szeged, Hungary; [4]Neurosci. Inst., New York Univ., New York, NY

Abstract: The proper decision-making with contingency predictions of behaviors is an essential ability for maximizing benefits of individuals and societies. However, the neural basis of such proper decision-making underlying behavioral selections under conflicting predictions of reward and punishment remains unknown. To investigate the neuronal underpinnings of the adaptive behavioral selections, we trained male Long-Evans rats with an operant discrimination task that includes decision-making for reward only or reward followed by footshock punishment while local field potentials in multiple brain regions were recorded. Rats were subjected to discriminate 9 kHz and 4 kHz pure tones in Go or NoGo trials for reward only or reward followed by footshock (0.2 - 0.5 mA for 1 s), respectively. A semi-supervised machine-learning technology called discriminative cross-spectral factor analysis successfully extracted an oscillatory brain activity pattern that decreases several seconds before the reward-taking behaviors in the NoGo

trials happened; we named the oscillatory brain activity pattern as the careful decision-making pattern. The careful decision-making pattern included beta and gamma oscillations in the prefrontal cortices and beta coherence between the medial prefrontal cortex and the amygdala. Chemomgenetic inhibition of the beta and gamma oscillations in the prefrontal cortices and the beta coherence between the medial prefrontal cortex and the amygdala reversibly impaired the proper behavioral selections between Go and NoGo trials. These results suggest a top-down control of impulsive decision-making by the prefrontal cortices on the subcortical limbic structures including the amygdala.

Disclosures: **K. Yoshida:** None. **M. Chan:** None. **Q. Li:** None. **L.K. Pedraza:** None. **R.O. Sierra:** None. **M. Minami:** None. **A. Berenyi:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Amplipex Llc, Neunos Inc. **Y. Takeuchi:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.15

Topic: H.03. Decision Making

Support: FRQS Doctoral Scholarship

Title: A touchscreen-based paradigm for foraging-like behaviour in mice

Authors: *D. LAU[1], D. PALMER[3], S. E. FULTON[2];
[1]Dept. of Neurosci., Ctr. de Recherche du CHUM, Univ. de Montréal, Montréal, QC, Canada; [2]Dept. of Nutr., Ctr. de Recherche du CHUM, Univ. de Montréal, Montreal, QC, Canada; [3]Dept. of Physiol. and Pharmacol., Robarts Res. Institute, Univ. of Western Ontario, London, ON, Canada

Abstract: Decision making during reward-motivated behaviour may be characterized as a cost-benefit trade-off, where costs, including time and effort, are weighed against the amount of reward that could be obtained. Animals foraging in an ecological context may encounter rewards in “patches” of finite, local concentrations of reward that are distributed throughout an environment. In patch-foraging tasks for food, animals may make choices about when to leave a food patch that is being depleted as reward is consumed (exploit), in search of a new patch that may have better reward (explore). How animals manage this explore-exploit dilemma may involve sequential evaluation of “foreground” reward (how much reward is currently being received from exploiting a patch) against the “background” environmental context (for example the overall richness of the environment an animal is occupying). Given growing interest in applying ethologically inspired tasks to evaluate complex relationships between effort, reward, and decision-making that are disrupted in obesity and numerous psychiatric conditions, we have developed a touchscreen-based task for visually guided foraging-like behaviour in mice.

C57BL/6J adult male mice (n=10) were trained to demonstrate their choices by making touch responses to visual cues representing either a “patch”, indicating the possibility to harvest food rewards that deplete over subsequent harvests, or a “travel” cue, indicating the possibility to open a new, replenished food patch. Our preliminary findings demonstrate mice are capable of learning a touchscreen-based task for foraging-like behaviour, with discrimination of touch responding to the active patch above 85%, patch-leaving behaviour that is sensitive to declining foreground reward, and sensitivity to foreground reward across patches with different initial amounts of reward. Ongoing experiments will evaluate extensions of this paradigm to examine more complex food environments in which manipulations of patch richness (foreground reward) are combined with independent manipulations of travel time (background reward) and effort conditions, to study behavioural sensitivity to cost in the context of dynamically varying foreground and background rates of reward.

Disclosures: D. Lau: None. **D. Palmer:** None. **S.E. Fulton:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.16

Topic: H.03. Decision Making

Support: NIH Grant R15DA046375
Cosmos Scholars Grant Program

Title: Neurons in the rat medial frontal cortex are sensitive to deliberation and noise during decision making

Authors: *S. R. WHITE[1], C. B. DARDEN[1], M. MITCHELL[1], M. W. PRESTON, Jr.[3], M. LAUBACH[2];
[2]Neurosci., [1]American Univ., Washington, DC; [3]Neurosciences Grad. Program, Univ. of California San Diego, La Jolla, CA

Abstract: Neural correlates of decision making were examined using a behavioral design in which rats selected lateralized visual stimuli following entries into a central response port. Luminance of the stimuli was associated with reward magnitude. After training with single offer stimuli, rats were tested with dual offers on one-third of trials, with one stimulus predictive of a large value reward and the other predictive of a low value reward. Rats show increased response latencies for dual-offer trials compared to single offers. This deliberation was apparent in the first testing session, and persisted over the period of testing. ExGauss modeling of response latencies revealed that the exponential component, but not the Gaussian component, was sensitive to deliberation and suggests that decisions on dual-offer trials are “noisy”. Neural correlates of decisions were measured in the medial frontal cortex (rostral part of the prelimbic area). Arrays of microwire electrodes were chronically implanted and recordings of single units

and field potentials were analyzed for effects of response location and latency, reward value, and trial type (single or dual offer). Many neurons were modulated during the period between entries into the central and lateralized response ports. Field potentials showed major fluctuations in low frequency rhythms at this time (delta and theta frequencies). Decoding methods (random forest) found that spike activity could be used to predict the location of a response, but surprisingly not reward value. Some neurons discriminated between trials with single and dual offers and/or were predictive of response latencies, especially on dual offer trials. To evaluate effects of "decision noise", suggested by exGauss modeling, we trained classifiers to discriminate between trials with left and right responses using single-offer trials and tested the classifiers with data from other single-offer trials and dual-offer trials. Predictions of side responding for dual-offer trials were less accurate than for single-offer trials for many neurons, suggesting that decision noise is represented in the medial frontal cortex.

Disclosures: S.R. White: None. **C.B. Darden:** None. **M. Mitchell:** None. **M.W. Preston:** None. **M. Laubach:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NIH R15DA046375-01A1.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.17

Topic: H.03. Decision Making

Support: Cosmos Scholars Grant Program

Title: Rostral medial frontal cortex and decision making: Effects of reversible inactivation, mu opioids, and HIV-1 tat

Authors: *J. A. PALMER, K. CHAVEZ LOPEZ, C. B. DARDEN, S. R. WHITE, M. LAUBACH;
American Univ. Program In Behavior, Cognition, and Neurosci. (BCaN), Washington, DC

Abstract: We investigated the role of the rostral medial frontal cortex (MFC) in a value-based decision making task. This part of the rodent MFC has not been studied extensively to date. Rats detected lateralized visual stimuli following entries into a central response port. Luminance was associated with reward magnitude. After training with single offer stimuli, rats were tested with dual offers on one-third of trials, with one stimulus predictive of a large value reward (30 ul of 16% sucrose) and the other predictive of a low value reward (30 ul of 4% sucrose). After training was complete, rats were implanted with chronic guide cannula, targeting the rostral prelimbic area, and tested with reversible inactivation (0.01-1ug/ul of the GABA-A agonist muscimol), the selective mu-opioid agonist DAMGO (1 ug/ul), and an infusion of the HIV-1 tat peptide (15

ug/ul), which is well established to produce inflammation and neurodegeneration resulting cognitive deficits through unknown neuronal mechanisms. Infusions of muscimol into the dorsal, but not the ventral, prelimbic area decreased response latency and choice accuracy. The greatest effects were observed with infusions in more lateral portions of the rostral prelimbic area, adjacent to the medial agranular cortex ("M2"). By contrast, infusions of DAMGO at the same sites increased response latency and choice accuracy. These findings suggest that acute mu opioid receptor activation slows down processing to optimize decisions. Following recovery from the infusions of DAMGO, rats received an infusion of HIV-1 tat. Rats with dorsal infusions showed progressive decreases in response latency over days, indicating that as degeneration occurred in dorsal regions of the rostral prelimbic area, rats increased the speed in which they responded. The rats with ventral infusions in the prelimbic area, just above the medial orbital cortex, showed less obvious effects and consistent response latencies over test sessions. Overall, our findings suggest that the dorsal part of the rostral prelimbic area controls the speed and accuracy of decision making and justifies future studies using neuronal recording methods to understand how opioids and HIV toxins influence cognitive processing in preclinical rodent model systems.

Disclosures: **J.A. Palmer:** None. **K. Chavez Lopez:** None. **C.B. Darden:** None. **S.R. White:** None. **M. Laubach:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NIH 1R15DA046375-01A1, DC CFAR Pilot Award.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.18

Topic: H.04. Executive Functions

Support: ERC StG MEMCIRCUIT 758032

Title: Single-neuron representations during acquisition of instrumental associations in mouse prefrontal cortex

Authors: *L. S. MEHRKE, X.-X. LIN, T. BERNKLAU, S. N. JACOB;
Translational Neurotechnology Laboratory, Dept. of Neurosurg., Klinikum rechts der Isar, Tech. Univ. of Munich, Munich, Germany

Abstract: In an ever-changing world, the ability to adapt to environmental changes is essential for all organisms. The process of acquiring new associations between sensory stimuli, own actions and desired outcomes and flexibly re-learning when necessary is crucial. These functions are known to rely on the prefrontal cortex (PFC), the brain's hub for cognitive control and behavioral flexibility. However, the neuronal mechanisms underlying these cognitive processes

have not been fully understood. Here, we investigated changes in the neuronal representations during association learning and flexible behavior on a single-neuron level in the mouse medial PFC. We trained head-fixed mice (n=6) in an auditory decision-making task with rule switches and measured calcium signals in pyramidal PFC neurons (GCaMP6f expressed under the CamKII promotor) with head-mounted miniature microscopes at beginner, intermediate and expert levels of performance. Animals were first trained to follow the location of an auditory cue, then cue frequency, and finally to respond to cue frequency with reversed actions. We measured a total of 6612 single neurons in 56 sessions. Summed across all sessions, 2013 neurons (30 %) were selective for either cue location or frequency during cue presentation, 3116 neurons (47 %) early after the animals' response and 3006 neurons (46 %) late after the animals' response. Interestingly, as the animals progressed through training and followed the different task rules, the changes in proportion of neurons with selectivity for one of the two cue dimensions allowed for inference of the currently relevant cue dimension only in the epoch following the animals' response, but not during cue presentation. In this epoch, selectivity for both dimensions increased almost monotonically across training sessions and independently of the task rule, possibly reflecting a more general effect of learning. In the response epoch, changes in selectivity for the task-relevant cue dimension associated with rule switches were abrupt, occurring within a single session, and lead to an increase in the number of selective neurons of up to 4-fold compared to the preceding session. In summary, our results support previous findings that PFC neurons adapt to changing behavioral demands and represent task-relevant information. Our chronic recordings from populations of well-identified single neurons will allow us to track the fate of the large proportion of neurons with changing selectivity and help to elucidate the prefrontal mechanisms underlying the ability to flexibly learn and re-learn instrumental associations.

Disclosures: L.S. Mehrke: None. **X. Lin:** None. **T. Bernklau:** None. **S.N. Jacob:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.19

Topic: H.04. Executive Functions

Support: The Centre of Excellence scheme of the Research Council of Norway (Centre for Neural Computation, grant #223262)
The National Infrastructure scheme of the Research Council of Norway NORBRAIN (grant #197467)

Title: General and context-specific tuning in the rat medial prefrontal cortex

Authors: *I. V. RAUTIO[1], F. NEVJEN[2], B. A. DUNN[2], J. R. WHITLOCK[1];
[1]Kavli Inst. for Systems Neurosci., [2]Dept. of Mathematical Sci., Norwegian Univ. of Sci. and Technol., Trondheim, Norway

Abstract: The medial prefrontal cortex has been implicated in diverse cognitive processes such as attention, goal-related navigation, action-planning, social information processing, learning and strategy-switching. Previous work has shown that sensory, motor and cognitive correlates in the mPFC tend to be mixed at the single cell level, necessitating methods to explore higher-dimensional spaces to uncover meaningful behavioral- or task-related correlates in the data. In an effort to better understand the apparent flexibility of this brain area, we implemented a multi-task approach where animals repeatedly experience an open field foraging task and a bait-chasing task in the same arena, as well as a social paradigm in which conspecifics interact on a linear track, while we record from hundreds of neurons simultaneously using Neuropixels probes. Additionally, we employed precise 3D tracking to investigate potential low-dimensional features like pose and movement in addition to higher level features in each context. Single neuron and population responses have so far shown sparse and unstable tuning to posture and movement covariates, whereas allocentric head direction tuning is expressed stably at the single cell level across tasks. In parallel, we identified a separate group of neurons in the social task encoding the direction of the other animal on the linear track. Similarly complex coding properties were elicited in an orthogonal population of neurons in the chasing task, which robustly represent the dynamically changing location of the bait relative to the rat. Spiking responses in each task drove stable correlation structures at the ensemble level. Leveraging the multi-task approach to disentangle behavioral selectivity from task or context, we are a step closer to identifying which types of selectivity are unique and which are common across these seemingly distinct behavioral contexts.

Disclosures: I.V. Rautio: None. **F. Nevjen:** None. **B.A. Dunn:** None. **J.R. Whitlock:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.20

Topic: H.04. Executive Functions

Support: Boehringer Ingelheim Ulm University Bio Center (BIU; grant TPN010)
German Research Foundation (DFG; grant KA 4594/2-1)
Else-Kroener-Fresenius/German-Scholars-Organization Programme for excellent medical scientists from abroad (grant GSO/EKFS 12)
Institute of Applied Physiology, Ulm University.

Title: Excitatory neurons of the anterior cingulate cortex encode distinct choices and outcomes reflecting executive control

Authors: *M. JENDRYKA[1], U. LEWIN[1], S. KAPANAIAH[1], H. DERMUTZ[2], B. LISS[1,3], A. PEKCEC[5], T. AKAM[4], B. GREWE[2], D. KÄTZEL[1];
[1]Inst. for Applied Physiol., Univ. of Ulm, Ulm, Germany; [2]Inst. for Neuroinformatics, ETH Zurich, Zurich, Switzerland; [3]Linacre and New Col., Univ. of Oxford, University of Oxford,

United Kingdom; [4]Dept. of Exptl. Psychology, Univ. of Oxford, Oxford, United Kingdom; [5]Cardio-Metabolic Dis. Res., Boehringer Ingelheim Pharma Gmbh & Co. KG, Biberach, Germany

Abstract: The appropriate control of sustained attention and impulsivity is essential for successful goal-directed behaviour and is impaired in a variety of neuropsychiatric disorders. Both executive functions can be measured by the 5-choice serial reaction time task (5-CSRTT) in humans and rodents. This task requires subjects to temporarily withhold responding and detect briefly presented cues to which they need to respond for gaining rewards. Using this task in mice, we and others have recently identified subsets of excitatory cells in the anterior cingulate cortex (ACC) that modulate forms of attention and impulsivity. However, what these cells encode during conditions that demand executive control, and which aspects of behavioural choices they reflect is currently unknown. We have addressed these questions by using open-source miniaturized endoscopic microscopes (UCLA miniscope) to monitor calcium transients of hundreds of neurons (expressing the genetically encoded calcium indicator GCamp6m) during the 5-CSRTT. To this end, we developed an open-source operant box system that is optimized for mice carrying miniscopes allowing them to perform the task with fast response times and high trial numbers per session. Implanted and pre-trained male adult C57BL/6J mice (n = 16) were exposed to different task conditions evoking impulsivity or inattentiveness, as indicated by an increase in premature or incorrect responses, respectively, and a concomitant decrease of correct responses. Applying supervised machine-learning, a wide range of classifiers were tested in their performance to decode the response type (i.e. correct, incorrect, premature or omitted response) from the neural activity of excitatory ACC neurons, on a trial-by-trial basis. We found that cross-validated prediction of the response type using a linear support vector machine (SVM) classifier reached an average accuracy of up to 90% (vs. 50% chance level). This indicates that each response type chosen on a given trial in the 5-CSRTT is encoded in the ACC. Temporal analysis of decoding performance showed that accuracy peaks approximately 500 ms after the choice, suggesting that ACC neurons encode primarily (but not exclusively) choice *outcome*. A temporal cluster-analysis of response patterns of individual neurons revealed that the ACC comprises distinct populations of neurons that are selectively active before, during or after a specific choice or its outcome. These data demonstrate that the ACC encodes distinct types and temporal phases of response options that reflect different levels of attentional and impulse control.

Disclosures: **M. Jendryka:** None. **U. Lewin:** None. **S. Kapanaiah:** None. **H. Dermutz:** None. **B. Liss:** None. **A. Pekcec:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim. **T. Akam:** None. **B. Grewe:** None. **D. Kätzel:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.21

Topic: H.04. Executive Functions

Support: R00DA042934

Title: Cocaine induced alterations in infralimbic cortex and dorsal lateral striatum neural encoding to reward predictive cues following outcome devaluation

Authors: L. A. ADRIAN[1], T. J. SLOAND[1,2], E. J. CIACCIARELLI[1], M. NIEDRINGHAUS[1], *E. A. WEST[1,2];
[1]Cell Biol. and Neurosci., Rowan Univ. SOM, Stratford, NJ; [2]Rowan Univ. Grad. Sch. of Biomed. Sci., Stratford, NJ

Abstract: The ability to alter behavior in response to changes in consequences is necessary for navigating an ever-changing environment. Substance use disorders (SUDs) are characterized by a continuation of maladaptive behavior despite negative consequences. Thus, characterizing the underlying processes that modulate the ability to change or stop behavior in response to updated expected outcomes is critical for understanding the neurobiological alterations in SUDs. We investigated how cocaine withdrawal leads to aberrant, differential patterns of neural activity in subregions of the rat frontal cortex (infralimbic, IL) and striatum (dorsal lateral striatum, DLS) to reward predictive cues following a decrease in expected outcome value. Mildly water deprived Long-Evans rats were split into two groups. Cocaine rats underwent self-administration for cocaine (i.v., 1 mg/kg/press) and control rats underwent self-administration for saline (i.v.) and water (0.2 ml into a receptacle). for daily 2-hour sessions for 14 days. After 3 weeks of abstinence, rats underwent Pavlovian conditioning for 10 days. They were presented with two distinct cue light patterns as conditioned stimuli (CS+), each predicting a different reward. One CS+ predicted a sugar pellet and the other CS+ predicted a food pellet (10 trials each). Two other light patterns did not predict a reward (CS-). After 10 days of conditioning, rats underwent a devaluation procedure for the sugar pellets to induce a conditioned taste aversion (LiCl, i.p., 0.3M; 7.5 ml/kg). After devaluation, rats were tested on the same Pavlovian task to evaluate their ability to avoid the devalued CS+ in the absence of the rewards (under extinction). IL and DLS electrophysiological recordings showed neuronal populations that were phasic to the CS+ [excited, EXC; or inhibited, INH] or nonphasic (no response to the CS+). Rats in the cocaine group showed higher % of excited phasic neurons compared the control group in the DLS in response to the devalued CS+ (control: 14% of total DLS neurons; cocaine: 37% of total DLS neurons) and nondevalued CS+ (control: 14% of total DLS neurons; cocaine: 26% of total DLS neurons). We also observed a similar pattern in the IL in that the cocaine group showed higher % of excited phasic neurons compared to the control group in response to the devalued CS+ (control: 3% of total IL neurons; cocaine: 18% of total IL neurons) and nondevalued CS+ (control: 7% of total IL neurons; cocaine: 18% of total IL neurons). Thus, we show elevated neural activity in the IL and DLS after a history cocaine to reward predictive cues which may contribute to the deficits observed in the ability of rats to shift behavior.

Disclosures: L.A. Adrian: None. **T.J. Sloand:** None. **E.J. Ciacciarelli:** None. **M. Niedringhaus:** None. **E.A. West:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.22

Topic: H.04. Executive Functions

Support: DA042934 (NIDA)

Title: Prelimbic cortex neural encoding in an Alzheimer's disease rat model during an outcome devaluation task

Authors: *T. J. SLOAND, M. NIEDRINGHAUS, E. A. WEST;
Rowan Univ. SOM, Stratford, NJ

Abstract: Alzheimer's disease (AD) is characterized by the accumulation of neuropathological markers and profound memory loss. AD rats (TgF344-AD) expressing mutant amyloid precursor protein and presenilin-1 exhibit age-dependent progressive AD pathology (plaques, tau pathology, oligomeric Aβ and neuronal loss) and show hypofunction within the prelimbic cortex (PrL) at 6 months. We have shown that PrL neural activity during learning predicts, and is necessary for, rats' ability to suppress behavior following outcome devaluation. We investigated if AD rats would show aberrant neural activity and/or behavioral responding to reward predictive cues following outcome devaluation. AD rats (n=6) and wild-type littermate controls (n=6) were presented with two distinct cues as conditioned stimuli (CS+; predicting a sugar or food pellet) and two cues that did not predict a reward (CS-); 10 trials each. After 10 sessions, rats underwent a devaluation procedure to induce a conditioned taste aversion (LiCl, i.p., 0.3 M; 7.5 ml/kg) to one reward. Rats were then tested on the same Pavlovian task (under extinction) to evaluate their ability to avoid the CS+ associated with the devalued outcome. WT rats spent less time in the food cup during the devalued CS+ (13.6%) compared to the non-devalued CS+ (21.0%). AD rats did not differ in the time spent in the food cup during the devalued CS+ compared to the non-devalued CS+ (10.9% vs. 14.3%). PrL electrophysiological recordings revealed distinct neuronal populations that were "phasic" to the CS+ [excited, EXC; or inhibited, INH]. In WTs, there was a lower % of PrL neurons that were classified as phasic to the devalued CS+ (26.5% of total) compared to the non-devalued CS+ (44% of total), while, in AD rats, the % of PrL neurons that were phasic were the same to both devalued CS+ (26% of total) and non-devalued CS+ (29% of total). Phasic neurons in AD rats were primarily excited to both the devalued (3% INH, 23% EXC) and non-devalued CS+ (9% INH, 20% EXC), whereas the phasic neurons in WT rats were primarily inhibited to both the devalued (20.5% INH, 6% EXC) and non-devalued CS+ (32% INH, 12% EXC). Neuronal population responses in the form of local field potentials were recorded and analyzed. In the 75-85 Hz range, WT rats showed decreased power to the devalued CS+ compared to the nondevalued CS+ while AD rats did not show any difference in power in response to either CS+. AD rats' aberrant behavioral responding may be a result of atypical mPFC encoding.

Disclosures: T.J. Sloand: None. **M. Niedringhaus:** None. **E.A. West:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.23

Topic: H.04. Executive Functions

Support: R00DA042934

Title: Nucleus reuniens to prelimbic cortex circuit is critical for performance on a delayed nonmatch to position task

Authors: E. CIACCIARELLI[1], S. DUNN[1], *M. NIEDRINGHAUS[1], T. J. SLOAND[1,2], T. GOHAR[1,3], L. ADRIAN[1], E. A. WEST[1,2];
[1]Cell Biol. and Neurosci., Rowan Univ. SOM, Stratford, NJ; [2]Rowan Univ. Grad. Sch. of Biomed. Sci., Stratford, NJ; [3]MARC Program, Rutgers University- Camden, Camden, NJ

Abstract: Nucleus reuniens to prelimbic cortex circuit is critical for performance on a delayed nonmatch to position task Ciacciarelli, E.J., Dunn, S.D, Sloand, T.J., Adrian, L.A., Gohar, T., Niedringhaus, M., West, E.A. Working memory is the ability to hold information online to guide behavior and is critical for navigating the environment. The rat prelimbic cortex (PrL) and nucleus reuniens (Re) of the thalamus are two anatomically interconnected structures that are directly involved in working memory. We are interested in the directionality of this involvement and aimed to determine this by utilizing inhibitory DREADDs. Specifically, we aimed to determine if PrL-Re connection and/or the Re-PrL connection were required for performance on a delayed nonmatch to position (DNMTP) task. Fischer 344 rats were injected with a Gi-coupled hM4D DREADD (hM4Di) or control virus into either the PrL (n=8 control, n=12 hM4Di) or Re (n=5 control, n=5 hM4Di). Rats were implanted with cannulae aimed at either the Re (PrL virus) or PrL (Re virus). The task consists of 1) sample phase (one lever extended, e.g., right), 2) time delay (2-32s), and 3) a choice phase, initiated by nosepoke, where the opposite lever (i.e., nonmatch) from the sample phase must be pressed (e.g., left) in order to receive a reward. Failure to press the sample lever, nosepoke, or press the choice lever, resulted in a trial omission. Once criterion (80% correct, 50 trials, 4s delay) was met, rats underwent stereotaxic surgery for bilateral implantation of infusion guide cannulae. Rats were then trained in the task with a 6s delay then were trained in the task with 4 distinct delays (last session 4, 8, 16, 32s). Rats then received bilateral intracerebral infusions of clozapine N-oxide (CNO) or equal volume saline with the order of drug treatment counterbalanced across rats. We found that rats the received CNO in the Re-PrL group (but not in the PrL-Re group) showed worse performance in the DNTMP compared to when those rats received saline. Specifically, we found a significant virus (hm4Di vs control) by drug (CNO vs saline) interaction in our Re-PrL group ($F(1, 8) = 13.26$, $p<0.05$), but not in our PrL-Re ($F(1,18) = 2.236$, $p > 0.1$). These findings suggest a thalamocortical (Re-PrL), but not corticothalamic control (PrL-Re), control of working memory as measured by the delayed nonmatch to position task.

Disclosures: E. Ciacciarelli: None. **S. Dunn:** None. **M. Niedringhaus:** None. **T.J. Sloand:** None. **T. Gohar:** None. **L. Adrian:** None. **E.A. West:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.24

Topic: H.03. Decision Making

Support: CIHR GR006181
CIHR GR008262

Title: Unique characteristics of anterior cingulate cortex (ACC) neural ensembles in impulsive rats

Authors: *J. SEAMANS[1], E. EMBERLY[2], C. C. LAPISH[3];
[1]Univ. of British Columbia, Vancouver, BC, Canada; [2]Simon Fraser Univ., Burnaby, BC, Canada; [3]IUPUI, Indianapolis, IN

Abstract: Cognitive impulsivity is the propensity to accept a smaller immediate reward in lieu of a larger delayed reward. It can adversely affect decision making. We studied how information coding in the anterior cingulate cortex (ACC) changed when rats acted impulsively. A variable payout delayed-discounting task was employed that required rats to chose between pressing one lever (delayed lever press, 'dLP') to receive 6 pellets at a delay or pressing another lever (immediate lever press, 'iLP') to receive a variable number of pellets immediately. Since each dLP increased the payout on iLP trials whereas each iLP decreased it, the optimal strategy was to favor dLPs in spite of the associated delays. The sessions were grouped as either 'impulsive' or 'non-impulsive' based on the rat's propensity for choosing iLPs over dLPs at a given delay. It was possible to accurately simulate the choice behavior on non-impulsive sessions using a Reinforcement Learning model in which the agent relied only on information about past outcomes to guide choice, whereas choice behavior on impulsive sessions was captured by an agent that was prevented from learning about past outcomes or an agent that learned from past outcomes but devalued this information at the time of choice. Regression-based Principle Components Analysis (PCA) revealed that ensembles of neurons exhibited remarkably robust tracking of outcomes across multiple task epochs, yet the strength of this tracking did not vary between groups. When Neural Networks were trained to use ensemble activity from prior outcomes to predict forthcoming lever presses, accuracy was much higher for non-impulsive sessions than impulsive sessions. We therefore conclude that impulsivity does not interfere with the ability of ACC neurons to keep track of prior outcomes but rather the ability to use this knowledge guide choice behavior.

Disclosures: J. Seamans: None. **E. Emberly:** None. **C.C. Lapish:** None.

Poster

565. Cortical Control of Decision Making in Rodents

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 565.25

Topic: H.03. Decision Making

Title: Revealing emotional encoding in the anterior cingulate cortex (ACC) via behavioral discovery

Authors: *A. LINDSAY, I. GALLELLO, B. CARACHEO, J. SEAMANS;
Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Emotion is a critical aspect of Anterior Cingulate Cortex (ACC) function; However, ACC neurons encode many variables related to many modalities within a combined, multiplexed signal and this makes it extremely challenging to segregate and disambiguate signals specifically related to emotion. We approached the issue by recording from groups of individual neurons in the ACC while rats were engaged in a 3-valence task designed to evoke specific emotional responses. In this task, a unique tone preceded a footshock in one context, a food pellet in a second context and null outcome in a third context. We previously reported that ACC ensembles exhibited highly specific activity patterns in each context, presumably because they were tracking the unique emotional reactions they evoked. Yet the contexts also evoked different behavioral responses that were also tracked by ACC neurons. For this and future studies, it was imperative that we are able to dissociate behavioral signals from other signals of interest. Characterizing the neural correlates of behaviour requires consistent and accurate classification of behaviour. To this end, we developed a pipeline of machine learning techniques built on top of existing state-of-art methods to categorize or classify unique sequences of individual limb movements through time from video input. Specifically, the movements of individual limbs were fit using a series of Morlet wavelets spanning various temporal scales. Clustering techniques were then used to find the patterns across limbs that consistently co-occurred (i.e. behaviors) throughout a given session. We then compared automatically discovered behaviours with manually selected behaviours in a shared behavioural space. Much like limb positions (Lindsay et al 2018), behaviours were in general weakly represented at the single neuron level, but were robustly represented across ensembles. Via machine-learning based predictive decoding, we demonstrate accurate prediction of behaviours and emotional contexts, and in addition show by signal regression that disambiguating behaviour from emotional context improves the characterization and predictive strength of emotional signals.

Disclosures: **A. Lindsay:** None. **I. Gallello:** None. **B. Caracheo:** None. **J. Seamans:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.01

Topic: H.03. Decision Making

Support: Wellcome Trust (209558)
Simons Foundation

Title: Encoding models reveal brain-wide signaling of motor activity and reward delivery

Authors: *B. GERCEK[1], J. ARLANDIS[2], J. BENSON[4], N. BONACCHI[3], J. CATARINO[2], G. A. CHAPUIS[6], M. FABBRI[2], M. FAULKNER[2], F. HU[7], J. M. HUNTENBURG[2], A. KHANAL[8], C. KRASNIAK[9], C. LANGDON[9], P. LAU[11], G. T. MEIJER[12], N. J. MISKA[13], J.-P. NOEL[5], A. PAN VAZQUEZ[15], C. ROSSANT[16], N. ROTH[17], Y. SHI[10], K. Z. SOCHA[14], J. W. PILLOW[18], P. DAYAN[19], A. POUGET[20], T. INTERNATIONAL BRAIN LABORATORY[21];
[1]Univ. de Geneve, Geneva, Switzerland; [3]Neurosci., [2]Champalimaud Ctr. for the Unknown, Lisbon, Portugal; [5]Ctr. for Neural Sci., [4]New York Univ., New York, NY; [6]Bioengineering, Imperial Col. London, London, United Kingdom; [7]Div. of Neurobiology, Dept. of Mol. and Cell Biol., Univ. of California, Berkeley, Berkeley, CA; [8]Univ. of California at Los Angeles, Los Angeles, CA; [9]Neurosci., [10]Cold Spring Harbor Lab., Cold Spring Harbor, NY; [11]MRC Harwell Inst., London, United Kingdom; [12]Fundacao Champalimaud, Lisbon, Portugal; [13]Neurosci., [14]Univ. Col. London, London, United Kingdom; [15]Neurosci., Princeton Univ., Princeton, NJ; [16]Ecole Normale Superieure, Paris Cedex 05, France; [17]Biol. Structure, Univ. of Washington, Seattle, WA; [18]Princeton Neurosci. Inst., Princeton, NJ; [19]Max Planck institute for Biol. Cybernetcs, Tuebingen, Germany; [20]Univ. Hosp. of Geneva, Geneva, Switzerland; [21]Intl. Brain Lab., London, United Kingdom

Abstract: Mixed selectivity in neural codes is well documented in multiple brain regions, with individual neurons exhibiting tuning to several variables that are explicit or implicit in behavior. While this mixed selectivity has been observed in multiple brain regions, the scope of such selectivity, and the variables selected for, have never been documented on the scale of the entire brain itself.We examine single neuron firing using neural activity recorded by the international brain lab (IBL) in its brain-wide map: 583 neuropixel penetrations covering 361 brain regions defined by the Allen atlas. The recordings were made in mice performing a task in which mice maximize rewards by exploiting a blockwise stimulus probability governing the appearance of stimuli. The task features auditory inputs, visual inputs, and a variety of behavioral signals which we can examine in the context of single-unit activity.We fit generalized linear models to express single-unit firing as a function of task and behavioral regressors. For each neuron, a model is fit which describes spike counts in bins as a function of stimulus, feedback, wheel speed (absolute value of velocity), block stimulus probability, and first movement onset. The resulting weights governing the predicted response of the model are compared against a statistical null distribution, and the per-region proportions of significantly modulated neurons are reported.Preliminary results show brain-wide sensitivity to wheel speed and reward, and to a lesser extent, the block probability of trial stimulus side. Notably there is very little sensitivity to directional wheel velocity (i.e., signed speed). Global sensitivity to reward delivery is more unexpected, and to our knowledge not previously observed in the literature.The broad sensitivity to block probability within the trial after stimulus is also a surprising result. Because we have overlapping regressors during the within-trial period for both stimulus side and movement direction, it seems unlikely that this result is simply attributable to correlations with those variables. In future work we aim to further investigate the effect of expectation and the prior on neural activity using behaviorally-informed estimates of the animal's internal prior. We also aim to investigate the basis of

widespread responses to reward, and whether those responses can be explained by motor activity like licking not included as model regressors.

Disclosures: **B. Gercek:** None. **J. Arlandis:** None. **J. Benson:** None. **N. Bonacchi:** None. **J. Catarino:** None. **G.A. Chapuis:** None. **M. Fabbri:** None. **M. Faulkner:** None. **F. Hu:** None. **J.M. Huntenburg:** None. **A. Khanal:** None. **C. Krasniak:** None. **C. Langdon:** None. **P. Lau:** None. **G.T. Meijer:** None. **N.J. Miska:** None. **J. Noel:** None. **A. Pan Vazquez:** None. **C. Rossant:** None. **N. Roth:** None. **Y. Shi:** None. **K.Z. Socha:** None. **J.W. Pillow:** None. **P. Dayan:** None. **A. Pouget:** None. **T. International Brain Laboratory:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.02

Topic: H.03. Decision Making

Support: Simons Foundation Grant 543011
Wellcome Trust Grant 216324/Z/19/Z
Fundação para a Ciência e a Tecnologia SFRH/BD/149020/2019

Title: Comparison of neuromodulator signaling in a visual decision-making task

Authors: *K. BOUGROVA[1], N. BONACCHI[2], J. A. CATARINO[1], E. E. DEWITT[3], G. T. MEIJER[1], P. DAYAN[4], Z. F. MAINEN[1], .. INTERNATIONAL BRAIN LABORATORY[5]; [1]Neurosci., [2]Champalimaud Ctr. for the Unknown, Champalimaud Ctr. for the Unknown, Lisbon, Portugal; [3]Neurosci., Champalimaud Ctr. for the Unknown, Lisboa, Portugal; [4]Max Planck institute for Biol. Cybernetcs, Max Planck institute for Biol. Cybernetcs, Tuebingen, Germany; [5]The Intl. Brain Lab., London, United Kingdom

Abstract: The International Brain Laboratory (IBL) is a collaboration of more than 20 experimental and theoretical labs studying decision-making in the mouse. Using a visually-guided decision task, IBL has demonstrated robust, replicable, behavioral and electrophysiological data, along with a growing complement of tailored and richly validated computational models. This provides an unparalleled opportunity to study the activity, and ultimately the function, of neuromodulators, which play a special and important role in theories of learning and decision-making. Dopamine (DA) has been suggested to report a reinforcement prediction error, at least under some circumstances and to some target nuclei. Equally, there is evidence that serotonin (5-HT) modulates learning based on confidence or uncertainty, and in reporting an unsigned or state prediction error. To understand the differential activation of these neuromodulators in the IBL task, we are using fiber-photometry to record bulk Ca2+ activity using GCaMP6 sensors expressed in genetically-defined neural populations in key nuclei. We will compare the activity of the neuromodulators to theoretical predictions and to the IBL behavior and brain wide electrophysiology data sets. As a validation of the approach, we tested

the hypothesis that ventral tegmental area DA neurons report a signed prediction error and dorsal raphe nucleus 5-HT neurons an unsigned prediction error. We used, respectively, 5 and 4 adult mice to target DA and 5-HT, using the isosbestic signal to control the GCaMP6 signal. During the first training stage, habituation, the mouse is being acclimatized to the rig and head-fixation, passively observing a visual stimulus that moves from the side to the center of the screen followed by a water reward delivery. Both DA and 5-HT neurons respond to the unexpected water. Next the mouse must learn to move the stimulus to the center by using a wheel. We observe a divergence in the DA signal associated with the outcome of the choice. DA activity increases upon water reward and decreases when the mouse makes the wrong decision and receives a noise burst. 5HT, in contrast, increases to both reward and noise burst, with a stronger response associated with incorrect trials, consistent with an unsigned prediction error signal. Currently, we are acquiring preliminary data for norepinephrine and acetylcholine, also proposed to play key roles in decision-making, with the goal of recording the activity of the four primary neuromodulators in the same task. In combination with the current IBL data sets, this study provides an unprecedented opportunity to investigate the role of neuromodulators in learning and decision-making.

Disclosures: **K. Bougrova:** None. **N. Bonacchi:** None. **J.A. Catarino:** None. **E.E. DeWitt:** None. **G.T. Meijer:** None. **P. Dayan:** None. **Z.F. Mainen:** None. **.. International Brain Laboratory:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.03

Topic: H.03. Decision Making

Support: Wellcome Trust (216324)
Simons Foundation

Title: International Brain Laboratory brain-wide-map analysis: Standardized Euclidean distance of trial-averaged activity across neuropixel recordings reveals a graded response to task-relevant variables

Authors: *M. SCHARTNER[1], C. LANGDON[2], B. BENSON[3], M. FAULKNER[4], J. M. HUNTENBURG[6], O. WINTER[7], M. J. WELLS[8], N. BONACCHI[9], C. ROSSANT[11], I. R. FIETE[12], M. FABBRI[10], T. INTERNATIONAL BRAIN LABORATORY[13], K. Z. SOCHA[5]; [1]Intl. Brain Lab., Parede, Portugal; [2]Neurosci., Cold Spring Harbor Lab., Cold Spring Harbor, NY; [3]Applied Physics, Stanford Univ., Redwood City, CA; [5]Univ. Col. London, [4]Univ. Col. London, London, United Kingdom; [6]Max Planck Res. Group Neuroanatomy and Connectivity, MPI for Human Cognitive and Brain Sci., Leipzig, Germany; [7]Champalimaud Ctr. For the Unknown, Lisboa, Portugal; [8]Intl. Brain laboratory, London, United Kingdom; [9]Neurosci., [10]Champalimaud Ctr. for the Unknown, Lisbon, Portugal; [11]Ecole Normale Superieure, Paris

Cedex 05, France; [12]Ctr. for Learning and Memory, UT Austin, Austin, TX; [13]CCU, Lisbon, Portugal

Abstract: Euclidean distances between mean responses have been used as a metric for quantifying how task-relevant variables are represented in neural populations. Here we apply this metric to the brain-wide-map of the international brain laboratory (IBL), which comprises hundreds of neuropixel recordings from uniformly sampled regions across the left hemisphere of mice during a biased binary decision making task (the IBL task). Head-fixed mice are trained via water reward to turn a wheel to the left (or right) if a gabor patch appears on a screen before them either on the left (or right). The stimulus side probability is distributed in consecutive blocks that alternate in probability for the left side between 0.2 and 0.8.
We compute the standardized euclidean distance between population responses for different trial conditions with data concatenated across all recording sessions. For each region and trial condition, we consider the temporal evolution of this distance within the trial, such as activity during the first 100 ms after stimulus onset for the stimulus being on the right versus on the left side of the screen. Other trial conditions we consider are windows aligned to post-feedback, pre-stimulus, pre-motion onset and post-motion onset.
Preliminarily, we find a graded response in neural activity across brain regions for each trial condition. For trials sorted by stimulus side and aligned to stimulus onset we see a transient response peaking at 50 ms in visual areas VISl and VISpm, in line with previous studies. For trials sorted by feedback type and aligned to feedback we see peak responses at 25 ms in hindbrain and midbrain regions, including VCO, ICc, CUN, likely reflecting the auditory cue. For trials sorted by choice type (100 ms pre motion onset), we find strong effects in secondary motor and upper limb area of the somatosensory cortex, as well as several subcortical regions, showing a ramping response in time, peaking at motion onset. For trials sorted by action (100 ms window after motion onset), the same areas are found, suggesting that there is a clear overlap of areas planning motion and those executing motion. These preliminary results are consistent with the picture of a decision-making process which is widely distributed across cortical and subcortical regions of the mouse brain.

Disclosures: M. Schartner: None. **C. Langdon:** None. **B. Benson:** None. **M. Faulkner:** None. **J.M. Huntenburg:** None. **O. Winter:** None. **M.J. Wells:** None. **N. Bonacchi:** None. **C. Rossant:** None. **I.R. Fiete:** None. **M. Fabbri:** None. **T. International Brain Laboratory:** None. **K.Z. Socha:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.04

Topic: H.03. Decision Making

Support: Simons Foundation Grant 543011
Wellcome Trust Grant 216324/Z/19/Z

Title: Serotonin modulates neural dynamics and inter-area communication across the mouse brain

Authors: *J. A. CATARINO[1], G. T. MEIJER[1], L. FREITAS-SILVA[1], I. COSTA LARANJEIRA[1], .. INTERNATIONAL BRAIN LABORATORY[2], Z. F. MAINEN[1]; [1]Neurosci., Champalimaud Ctr. for the Unknown, Lisbon, Portugal; [2]The Intl. Brain Lab., London, United Kingdom

Abstract: Serotonin (5-HT) is a central neuromodulator which is implicated in the regulation of cognitive flexibility. 5-HT is released from the dorsal raphe nucleus (DRN), projecting throughout the entire brain. How serotonin is involved in the rapid reconfiguration of neural circuits which may underlie the expression of flexible behavior remains a topic of debate. Therefore, we present an extensive study investigating the modulatory effect of serotonin on neural spiking activity across the mouse brain. We performed acute Neuropixel recordings in eight target locations, recorded in pairs, while serotonin neurons in the DRN were optogenetically stimulated. Targeted cortical areas included frontal, barrel, retrosplenial, piriform, and posterior parietal cortex. Recorded subcortical regions included the hippocampus, thalamus, amygdala, superior colliculus, and tail of the striatum. A total of 4818 neurons were recorded in 64 brain regions across 7 mice. We found that stimulating 5-HT release resulted in a substantial suppression of spiking activity in the hippocampus. The medial prefrontal (mPFC), orbitofrontal (OFC) and secondary motor cortex (M2) showed balanced serotonergic enhancement and suppression of neuronal activity. Serotonin stimulation also changed how brain regions communicated with one another. Communication subspace analysis revealed that the mPFC decreased its communication with neighboring M2 while the OFC showed increased information exchange with M2. Furthermore, we observed an increase in information exchange between thalamus and cortex but a decrease between the thalamus and the hippocampus. These findings shed light on the functional dynamics of the serotonergic projection system, and have important implications for the computational role of serotonin in large-scale neural dynamics across the brain.

Disclosures: **J.A. Catarino:** None. **G.T. Meijer:** None. **L. Freitas-Silva:** None. **I. Costa Laranjeira:** None. **.. International Brain Laboratory:** None. **Z.F. Mainen:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.05

Topic: H.03. Decision Making

Support: Wellcome Trust (216324)
Simons Foundation
The National Institutes of Health (NIH U19NS12371601)

Title: International Brain Laboratory brainwide analysis: decoding of task and behavioral variables from populations of neurons

Authors: *B. BENSON[1], C. FINDLING[2], F. HUBERT[3], M. R. WHITEWAY[5], B. GERCEK[4], J. ARLANDIS[6], J. BENSON[7], D. BIRMAN[9], J. A. CATARINO[6], E. E. J. DEWITT[6], F. HU[11], A. KHANAL[12], C. S. KRASNIAK[13], C. LANGDON[14], P. LAU[16], G. T. MEIJER[6], N. J. MISKA[18], J.-P. NOEL[8], K. NYLUND[10], A. PAN-VAZQUEZ[19], N. ROTH[9], Y. SHI[15], K. Z. SOCHA[17], A. E. URAI[20], T. IBL STAFF[21], P. DAYAN[22], A. POUGET[2], T. INTERNATIONAL BRAIN LABORATORY[21];
[1]Stanford Univ., Stanford, CA; [2]Univ. of Geneva, Geneva, Switzerland; [3]Basic Neurosciences, [4]Basic Neurosci., Univ. de Geneve, Geneva, Switzerland; [5]Neurosci., Columbia Univ., New York, NY; [6]Champalimaud Ctr. for the Unknown, Lisbon, Portugal; [8]Ctr. for Neural Sci., [7]New York Univ., New York, NY; [9]Biol. Structure, [10]Univ. of Washington, Seattle, WA; [11]Div. of Neurobiology, Dept. of Mol. and Cell Biol., Univ. of California, Berkeley, Berkeley, CA; [12]UCLA, Los Angeles, CA; [13]Neurosci., Cold Spring Harbor Lab., New York, NY; [14]Neurosci., [15]Cold Spring Harbor Lab., Cold Spring Harbor, NY; [17]Univ. Col. London, [16]Univ. Col. London, London, United Kingdom; [18]Sainsbury Wellcome Ctr., London, United Kingdom; [19]Princeton Univ., Princeton, NJ; [20]Cognitive Psychology, Leiden Univ., Univ. of Leiden, Leiden, Netherlands; [21]The Intl. Brain Lab., Lisbon, Portugal; [22]Max Planck institute for Biol. Cybernetcs, Tuebingen, Germany

Abstract: Decoding is a popular approach for assessing the information the activity of a neural population contains about externally accessible variables. Decoding analyses are typically limited to examining a small fraction of the brain at high temporal resolution (e.g. using electrophysiology), or a large fraction of the brain at low temporal resolution (e.g. fMRI). Here we use data from hundreds of neuropixel penetrations covering hundreds of brain regions in the Allen atlas to decode task and behavioral variables at unprecedented spatial and temporal resolutions in mice performing a perceptual decision-making task.
In the International Brain Lab (IBL) task, mice are presented with a visual grating stimulus on one side of a screen, and report whether this was left or right by turning a steering wheel; this results in a reward if the chosen side matches the stimulus side. Mice maximize rewards by exploiting a blockwise prior probability governing stimulus side. We decode task variables (blockwise prior probability; stimulus identity; reward) and behavioral variables (choice; wheel speed; whisker movements) from neural activity using maximum likelihood linear and logistic regression. We report decoding measures (R2 or accuracy) on held-out test trials using multi-fold cross validation, assessing statistical significance by comparison with bespoke null distributions. Preliminary results indicate substantial variability in decoding performance of all variables across sessions and brain regions. Despite this, we find several broad trends in the data. The reward signal, motion energy of the whisker pad, and wheel movements are represented across many brain regions. Notably, wheel speed is better decoded than wheel velocity across all brain regions considered. This result indicates that much of the movement-related information in brainwide neural activity is not specific to the exact kinematics of the movement.
We can also decode blockwise prior probability, stimulus, and choice across a number of cortical and subcortical regions. Decoding of pre-stimulus blockwise prior probability, however, is more modest than for stimulus and choice decoding. We find, for example, strong stimulus decoding in VISp, VISpm, and ZI and strong choice decoding in MOp, MOs, CP, and MRN.
In future work, we aim to compare our results with encoding and dimensionality reduction

analyses. We also hope to build on our decoding of the blockwise prior probability to better understand how prior probabilities are represented across brain regions.

Disclosures: **B. Benson:** None. **C. Findling:** None. **F. Hubert:** None. **M.R. Whiteway:** None. **B. Gercek:** None. **J. Arlandis:** None. **J. Benson:** None. **D. Birman:** None. **J.A. Catarino:** None. **E.E.J. DeWitt:** None. **F. Hu:** None. **A. Khanal:** None. **C.S. Krasniak:** None. **C. Langdon:** None. **P. Lau:** None. **G.T. Meijer:** None. **N.J. Miska:** None. **J. Noel:** None. **K. Nylund:** None. **A. Pan-Vazquez:** None. **N. Roth:** None. **Y. Shi:** None. **K.Z. Socha:** None. **A.E. Urai:** None. **T. IBL Staff:** None. **P. Dayan:** None. **A. Pouget:** None. **T. International Brain Laboratory:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.06

Topic: H.03. Decision Making

Support: Wellcome Trust (216324)
Simons Foundation
The National Institutes of Health (NIH U19NS12371601)

Title: Single-cell correlates of sensory, cognitive, and motor variables across the brain

Authors: ***Y. SHI**[1], N. ROTH[3], N. A. STEINMETZ[3], T. A. ENGEL[2], B. BENSON[4], D. BIRMAN[3], N. BONACCHI[6], E. E. DEWITT[7], B. GERCEK[8], C. LANGDON[2], F. HUBERT[9], J. ARLANDIS[10], K. NYLUND[11], K. SOCHA[12], N. J. MISKA[13], M. SCHARTNER[14], M. R. WHITEWAY[17], O. WINTER[18], M. FAULKNER[12], J. M. HUNTENBURG[19], C. ROSSANT[12], M. FABBRI[15], G. A. CHAPUIS[20], P. DAYAN[22], I. R. FIETE[23], S. GANGULI[5], K. HARRIS[12], A. POUGET[21], I. B. WITTEN[24], I. INTERNATIONAL BRAIN LABORATORY[16];
[1]Cold Spring Harbor Lab., [2]Cold Spring Harbor Lab., Cold Spring Harbor, NY; [3]Biol. Structure, Univ. of Washington, Seattle, WA; [4]Applied Physics, Stanford Univ., Redwood City, CA; [5]Stanford Univ., Stanford, CA; [6]Neurosci., Champalimaud Ctr. for the Unknown, Lisbon, Portugal; [7]Neurosci., Champalimaud Neurosci. Programme, Lisboa, Portugal; [8]Basic Neurosci., [9]Basic Neurosciences, Univ. de Geneve, Geneva, Switzerland; [10]Champalimaud Res., Lisbon, Portugal; [11]Washington Univerisity, Seattle, WA; [13]Neurosci., [12]Univ. Col. London, London, United Kingdom; [14]Intl. Brain Lab., Parede, Portugal; [15]Intl. Brain Lab., Lisbon, Portugal; [16]Intl. Brain Lab., London, United Kingdom; [17]Neurosci., Columbia Univ., New York, NY; [18]Champalimaud Ctr. For the Unknown, Lisboa, Portugal; [19]Max Planck Res. Group Neuroanatomy and Connectivity, MPI for Human Cognitive and Brain Sci., Leipzig, Germany; [20]Univ. of Geneva, Geneva, Swaziland; [21]Univ. of Geneva, Geneva, Switzerland; [22]Max Planck institute for Biol. Cybernetcs, Tuebingen, Germany; [23]MIT, Cambridge, MA; [24]Princeton Univ., Princeton, NJ

Abstract: During a decision-making task, neurons correlate not only with behavioral task variables such as vision, action selection, and prior expectation, but also overall task responsiveness and various types of body movement. Neural activity correlated with these behavioral task variables and movement may be distributed across brain regions. Here we use non-parametric statistical tests to delineate the spatial distribution of single-neuron activity relevant for these processes across nearly the whole brain. To achieve this, we introduce a dataset of brain-wide Neuropixels recordings from mice performing a visual discrimination task in which they turn a wheel left or right to indicate the location of a presumed visual stimulus. In each recording session, the probability of stimuli appearing on the left vs. right is 80% (or 20%) in blocks of consecutive trials. As of June 2022, the recordings consist of 49292 neurons recorded from 258 brain regions, combining recordings from 12 laboratories.
To quantify the sensitivity of single neurons to the visual stimulus (left vs. right side of visual stimulus), action selection (left vs. right direction of turning), action initiation (pre-movement vs. baseline) and prior expectation (left vs. right block-side), we computed the sensitivity metric of given condition based on the combined-conditions Mann-Whitney U statistic. We then computed the fraction of neurons in each brain region significant for the behavioral task, visual stimulus, action selection and prior expectation, and identified brain regions that are most relevant for these conditions. We found that about 29% of the brain regions are significant for coding of prior expectation. About 30-50% of brain regions are significant for side of visual stimulus/action selection.
To quantify the responsiveness of neural activity in task epoch, we used Wilcoxon rank-sum test to compare firing rates in baseline (pre-stimulus) and different task periods aligned with stimulus onset, first-movement onset or reward. To measure the behavioral movement correlates of single neurons in the entire recording sessions, we applied a time-shift test to compute the significance of Pearson correlation coefficient between binned spiking activity and time-series of behavior variables. We found that in most brain regions more than half of the neurons have significant task responsiveness and correlations to behavioral movement variables.
Overall, we observed widespread coding of movement and task responsiveness but more restricted representations of task-variable coding. These results suggest that neurons across many brain regions are modulated by performance of a behavioral task.

Disclosures: **Y. Shi:** None. **N. Roth:** None. **N.A. Steinmetz:** None. **T.A. Engel:** None. **B. Benson:** None. **D. Birman:** None. **N. Bonacchi:** None. **E.E. DeWitt:** None. **B. Gercek:** None. **C. Langdon:** None. **F. Hubert:** None. **J. Arlandis:** None. **K. Nylund:** None. **K. Socha:** None. **N.J. Miska:** None. **M. Schartner:** None. **M.R. Whiteway:** None. **O. Winter:** None. **M. Faulkner:** None. **J.M. Huntenburg:** None. **C. Rossant:** None. **M. Fabbri:** None. **G.A. Chapuis:** None. **P. Dayan:** None. **I.R. Fiete:** None. **S. Ganguli:** None. **K. Harris:** None. **A. Pouget:** None. **I.B. Witten:** None. **I. International Brain Laboratory:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.07

Topic: H.03. Decision Making

Support: Wellcome Trust (216324)
Simons Foundation

Title: Distributed neural representations of prior information in mouse decision-making

Authors: *F. HUBERT[1], C. FINDLING[1], B. GERÇEK[1], B. BENSON[2], M. R. WHITEWAY[3], C. KRASNIAK[4], P. DAYAN[5,6], A. POUGET[1], J. ARLANDIS[7], J. BENSON[8], N. BONACCHI[7], J. CATARINO[7], E. DEWITT[7], M. FABBRI[7], F. HU[9], A. KHANAL[10], C. LANGDON[4], P. LAU[11], G. MEIJER[7], N. J. MISKA[12], J.-P. NOËL[8], A. PAN-VASQUEZ[13], C. ROSSANT[7], N. ROTH[14], Y. SHI[4], K. SOCHA[12], A. E. URAI[15], M. WELLS[7], O. WINTER[7], T. INTERNATIONAL BRAIN LABORATORY[16];
[1]Fac. of Med. & Dept. of Basic Neurosciences, Univ. of Geneva, Geneva, Switzerland; [2]Applied Physics, Stanford Univ., Redwood City, CA; [3]Neurosci., Columbia Univ., New York, NY; [4]Neurosci., Cold Spring Harbor Lab., Cold Spring Harbor, NY; [5]Max Planck institute for Biol. Cybernetics, Tübingen, Germany; [6]Univ. of Tübingen, Tübingen, Germany; [7]Champalimaud Ctr. for the Unknown, Lisbon, Portugal; [8]New York Univ., New York, NY; [9]Div. of Neurobiology, Dept. of Mol. and Cell Biol., Univ. of California, Berkeley, Berkeley, CA; [10]Univ. of California, Los Angeles, CA; [11]MRC Harwell Inst., Harwell Campus, United Kingdom; [12]Neurosci., Univ. Col. London, London, United Kingdom; [13]Princeton Univ., Princeton, NJ; [14]Biol. Structure, Univ. of Washington, Seattle, WA; [15]Cognitive Psychology, Leiden Univ., Univ. of Leiden, Leiden, Netherlands; [16]The Intl. Brain Lab., Lisbon, Portugal

Abstract: Despite numerous studies, the neural basis of approximate Bayesian inference, and, in particular, how prior information impacts decisions, remains unclear. A dominant hypothesis is that prior information is incorporated in decision-making at a late stage of processing, in high-order areas such as OFC, ACC or LIP, right before motor commands are issued. Alternatively, information may be broadcast throughout the brain with top-down influences all the way to sensory areas.
To address this question, we examined brainwide neuropixel recordings collected by the International Brain Lab (IBL). In the IBL task, mice are trained to indicate the location of a visual grating stimulus (left or right). Crucially, the prior probability that the stimulus appears on the left flips between 20% and 80% between blocks of variable length.
We found that mice leverage the prior probability over the block to improve their decision accuracy. In particular, they perform better than chance (using this prior) when the grating contrast is set to zero. As a crude approximation to their computation, we therefore designed a Bayes optimal algorithm for estimating the block probability on a trial by trial basis given the specific set of trials experienced by each animal in each session. We then decoded this Bayes optimal estimate from 361 brain regions in the Allen atlas, using carefully quality-controlled recordings of the activity of over 200 000 putative single neurons.
For each brain region, we used cross-validated Lasso linear decoders. Statistical significance was assessed by comparing our result to a null distribution designed to account for potential spurious correlations between blocks and neural drift or other slow changes.
In both inter-trial and within-trial periods, we observed that the prior is widely represented throughout the mouse brain. Consistent with previous work, it is present in particular in high level cortical areas such as ACC or OFC. However, it is also seen throughout substantial portions of the rest of the brain, including early sensory cortical and subcortical regions, such as primary

visual cortex or superior colliculus. Overall, we find that around 24% of regions reflect the prior, both in cortical and subcortical regions.
This widespread representation of the prior argues for a neural model of Bayesian inference involving loops between areas, as opposed to a model in which the prior is incorporated only in decision making areas. This study offers the first brain-wide perspective on prior encoding, underscoring the importance of using large scale recordings on a single standardized task.

Disclosures: **F. Hubert:** None. **C. Findling:** None. **B. Gerçek:** None. **B. Benson:** None. **M.R. Whiteway:** None. **C. Krasniak:** None. **P. Dayan:** None. **A. Pouget:** None. **J. Arlandis:** None. **J. Benson:** None. **N. Bonacchi:** None. **J. Catarino:** None. **E. DeWitt:** None. **M. Fabbri:** None. **F. Hu:** None. **A. Khanal:** None. **C. Langdon:** None. **P. Lau:** None. **G. Meijer:** None. **N.J. Miska:** None. **J. Noël:** None. **A. Pan-Vasquez:** None. **C. Rossant:** None. **N. Roth:** None. **Y. Shi:** None. **K. Socha:** None. **A.E. Urai:** None. **M. Wells:** None. **O. Winter:** None. **T. International Brain Laboratory:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.08

Topic: H.03. Decision Making

Support: NIH Grant R15DA046797
NIH Grant F31DA050458

Title: Dopaminergic and noradrenergic modulation of sensitivity to delayed punishment

Authors: G. L. MINNES, A. J. WEINER, A. S. PISAHL, T. J. QUALLS, A. LILEY, D. B. GABRIEL, *N. W. SIMON;
Univ. of Memphis, Memphis, TN

Abstract: Decision-making involves careful consideration of all potential positive and negative outcomes. Importantly, negative outcomes often occur later in time, leading to underestimation, or “discounting,” of these consequences. The Delayed Punishment Decision-making Task (DPDT) was developed to study sensitivity to delayed vs immediate punishment during cost/benefit decision-making in rats. Rats choose between two levers, one resulting in a small, single-pellet reward with no foot shock punishment, and the other resulting in a larger, three-pellet reward followed by a mild foot shock punishment. This punishment is preceded by a systematically increasing delay as the blocks progress (0, 4, 8, 12, 16 s). DPDT revealed that rats choose the punished option more as the delay in punishment increases, indicative of discounting of delayed punishment. Here, we examined the effects of systemic administration of catecholaminergic drugs on sensitivity to delayed punishment discounting in male and female adult rats. We found that acute cocaine did not affect choice of rewards with immediate punishment, but caused a dose-dependent reduction in choice of delayed punishment.

Interestingly, this effect was more prominent in females than males. Neither activation nor blockade of the D1 dopamine receptor affected decision-making, whereas activation but not blockade of the D2 dopamine receptor abolished the discounting of delayed punishment. We next found that D2 receptor blockade did not alter the acute effects of cocaine during DPDT, suggesting that cocaine's affects on decision-making are not regulated by the D2 receptor. Finally, we observed that atomoxetine, a norepinephrine reuptake inhibitor, decreased choice of the punished reward regardless of punishment delay. Overall, these data demonstrate that dopamine and norepinephrine transmission differently modulate sensitivity to delayed vs. immediate punishment.

Disclosures: G.L. Minnes: None. **A.J. Weiner:** None. **A.S. Pisahl:** None. **T.J. Qualls:** None. **A. Liley:** None. **D.B. Gabriel:** None. **N.W. Simon:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.09

Topic: H.03. Decision Making

Support: NIH Grant R15DA046797
NIH Grant F31DA050458-02

Title: Orbitofrontal cortex encoding of delayed punishment during decision-making

Authors: *A. LILEY, D. B. K. GABRIEL, G. L. MINNES, N. W. SIMON;
Univ. of Memphis, Memphis, TN

Abstract: One characteristic of many psychiatric illnesses, such as substance use disorder and attention deficit hyperactivity disorder, is the undervaluation of future negative outcomes. However, there is currently a paucity of knowledge on the neuronal mechanisms governing sensitivity to delayed punishment. To evaluate this maladaptive behavior in a rodent model, we developed the Delayed Punishment Decision-making Task (DPDT). During the task, rats are given the choice between an immediate large reward succeeded by a foot shock or an immediate small reward that is safe. As the task progresses, an incrementally increasing delay is added preceding the foot shock. Rats avoid large rewards associated with punishments that occur immediately, but increasingly favor large rewards succeeded by delayed punishments, indicative of the underestimation, or discounting, of delayed punishment.
Previous findings from our lab using bilateral pharmacological inactivation suggest that the lateral orbitofrontal cortex (lOFC) influences cost-benefit decision-making with delayed punishment. However, it is unclear how OFC activity encodes information during this form of decision-making. Here, we investigated time-specific neuronal encoding of information during DPDT using single-unit electrophysiological recording with drivable microwire electrode arrays unilaterally implanted into left or right lOFC (counterbalanced across subjects). As previously,

we observed that rats shifted preference toward the punished reward when punishment was delayed. We recorded 150 units from 2 Long Evans male rats. It was observed that different subsets of units were sensitive to different task events, with a large percentage of neurons either activated or suppressed prior to a decision. Interestingly, there was a significant attenuation of suppressed units prior to choice of delayed compared to immediate punishment, which may reflect discounting of the impending punishment. Additionally, a subset of neurons demonstrated either a sustained inhibition or activation during the delay preceding punishment, suggesting that these neurons may be bridging the gap between action and delayed punishment. This research represents the first neurophysiological investigation of delayed vs immediate punishment, which may have implications for developing biological interventions to improve decision-making in psychopathology.

Disclosures: **A. Liley:** None. **D.B.K. Gabriel:** None. **G.L. Minnes:** None. **N.W. Simon:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.10

Topic: H.03. Decision Making

Support: NIH Grant R15DA046797
NIH Grant F31DA050458

Title: Effects of maternal separation on risky decision-making

Authors: A. J. WEINER, A. S. PISAHL, ***G. L. MINNES**, B. A. BASKHAIROUN, N. W. SIMON;
Univ. of Memphis, Memphis, TN

Abstract: Early life isolation and separation from one's mother, termed maternal separation (MSEP), is an increasingly common occurrence in society, particularly in individuals with low socio-economic status. This form of early life adversity has enduring effects on brain development and causes a myriad of neurobiological and behavioral deficits. To develop treatments to mitigate the effects of MSEP, it is critical to determine which aspects of cognition are vulnerable and when these disturbances manifest across the lifespan. To this end, we tested the effects of an established MSEP protocol on risky decision-making in both adolescence in adulthood. During MSEP, pups were separated from their mother for four hours a day for two weeks (PD11-PD21). Then, rats trained in the Risky Decision-Making Task (RDT), in which subjects chose between a small, safe reward and a larger reward accompanied by dynamic risk of a mild foot shock (0%, 25%, 50%, 75%, 100%). We observed that rats that underwent MSEP were significantly riskier than controls during adolescence. Interestingly, females exposed to MSEP were substantially riskier during adolescence than adulthood, whereas males displayed comparable risk-taking during these periods. Collectively, these data confirm that MSEP can

engender increased risk-taking in adolescence. In addition, we observed that the development of risk-taking across the lifespan differs based on sex, which has important implications for the development of both behavioral and biological treatments to improve decision-making in psychopathology.

Disclosures: A.J. Weiner: None. **A.S. Pisahl:** None. **G.L. Minnes:** None. **B.A. Baskhairoun:** None. **N.W. Simon:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.11

Topic: H.03. Decision Making

Support: NIH Grant F31DA050458
NIH Grant R15DA046797

Title: Lateral orbitofrontal cortex encodes punishment risk and reward magnitude separately during risky decision-making

Authors: *D. GABRIEL, A. LILEY, I. AGUILAR, N. SIMON;
Univ. of Memphis, Memphis, TN

Abstract: Effective decision-making requires rapid evaluation of outcomes expected from available choices. Desirable outcomes commonly involve an element of risk, and excessive risk-taking can result in persistent reward seeking despite likely consequences. The rat risky decision-making task (RDT) measures punishment-based risky behavior, and reveals that rats typically shift preference away from risky rewards as probability of punishment increases. While neural substrates of risky behavior have been identified, the functional neuronal activity underlying punishment guided risky decision-making is largely unknown. We recorded neuronal activity in the rat lateral orbitofrontal cortex (lOFC), a brain region implicated in decision-making and sensitivity to punishment. Activity of 554 individual units within lOFC was recorded from six male rats behaving in RDT. Analyses focused on neuronal activity during three discrete decision-making stages: deliberation, choice, and outcome anticipation. As observed previously, choice shifted away from large rewards when risk of punishment was introduced. Deliberation before choice of large nonrisky rewards evoked selective responses (phasic inhibition/activation) from distinct neuronal subpopulations in lOFC. Introducing risk of punishment attenuated net inhibition and increased selective activations, suggesting that lOFC activity encodes the presence of risk prior to choice. A similar alteration in selectivity was evident during outcome anticipation, wherein anticipation of risky outcomes evoked a net attenuation of selective inhibition. Interestingly, reduced selective inhibition was evident regardless of whether the subject had chosen the risky or safe outcome. Regardless of risk level, distinct neuronal sub-populations activated during choice of large vs small rewards, suggesting separate units encode

differences in reward magnitude. Finally, Random Forest classifiers used neural features to predict risk presence or choice. The presence of risk was most accurately classified by neuronal activity during pre-choice deliberation, while activity during deliberation, choice, and outcome anticipation all classified choice equally. Collectively, these data suggest lOFC flexibly encodes multiple features of risky decision-making, most prominently signaling risk of punishment prior to choice. This signal may inform downstream regions involved with action selection to guide decision-making. These data suggest manipulation of phasic lOFC activity during deliberation before risky choice may serve as a therapeutic strategy for disorders associated with maladaptive risky behavior.

Disclosures: **D. Gabriel:** None. **A. Liley:** None. **I. Aguilar:** None. **N. Simon:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.12

Topic: H.03. Decision Making

Support: Einstein foundation Berlin
Human Brain Project
Deutsche Forschungsgemeinschaft, SFB1315
European Research Council

Title: Flow of cortical information during evidence accumulation in mouse neocortex.

Authors: *M. NASHAAT[1], H. ORABY[1], S. TENG GOH[1], M. BOSC[2], S. KÖRNER[1], S. KARAYEL[1], A. KEPECS[3], M. E. LARKUM[1];
[1]Humboldt Univ. of Berlin, Berlin, Germany; [2]Kepecs Lab., Cold Spring Harbor Lab., Cold Spring Harbor, NY; [3]Cold Spring Harbor Lab., St. Louis, MO

Abstract: A major challenge in understanding cortical mechanisms of decision-making is disentangling the confounding contribution of multiple microcircuits across different time scales of the decision process. During evidence accumulation, sequential sampling of noisy information is a fundamental strategy for optimal decision making that requires integrating sensory evidence over time. Previous studies have investigated the contribution of different brain regions during evidence accumulation, however there is a lack of a mechanistic view that amasses the different findings together. Most studies are limited by technique used, e.g only one brain region can be recorded at a time, or the requirements imposed by advanced neural acquisition techniques, e.g head-fixation. Furthermore, a considerable body of work in human and non-human primates have utilized the random-dots kinematics (RDK/RDM) task. While in rodents a variety of stimuli have been used, RDK has not been investigated. Such variations yield a conservative transfer of findings across species.
In this study, we aim to disentangle the spatio-temporal contribution of different cortical areas

during evidence accumulation in the RDK task. We developed a novel nose-poking head-fixation system that enables advanced neural acquisition while retaining classical nose-poking behavioral structure. Psychophysical report of the head-fixed mice shows that they can optimally accumulate evidence across time. Our analysis of wide-field imaging in mice expressing calcium fluorescence in L2/3 and L5 pyramidal cells reveals a distinct fronto-parietal activity pattern during evidence accumulation. We accordingly optogenetically inhibited the implicated regions. Our findings identified distinct temporal profiles for different fronto-pareital cortical areas. The results bridge the gap between studies in primates and rodents, proposing a refined model for the flow of information through the neocortex during evidence accumulation.

Disclosures: **M. Nashaat:** None. **H. Oraby:** None. **S. Teng Goh:** None. **M. Bosc:** None. **S. Körner:** None. **S. Karayel:** None. **A. Kepecs:** None. **M.E. Larkum:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.13

Topic: H.03. Decision Making

Support: McDonell Center for Systems Neuroscience Small Grants Program

Title: The serotonergic psychedelic DOI specifically alters mouse behavioral strategy on a value guided dynamic foraging task

Authors: ***A. CHRISTENSEN**[1], S. STAROSTA[1], E. PHELAN[1], E. STEVENS, Jr.[1], M. BERGSTROM[1], S. J. BRUNWASSER[1], K. HENGEN[1], P. OSTEN[2], A. KEPECS[1]; [1]Washington Univ. in St. Louis, St. Louis, MO; [2]Certerra Inc., Farmingdale, NY

Abstract: Serotonergic psychedelics have shown remarkable potential to treat a variety of mental health disorders, and they reliably lead to profound changes in acute human subjective experience. Incredibly, these psychological and therapeutic effects are widely thought to be mediated via a single, specific biological substrate - agonism of the 5-hydroxytryptamine receptor subtype 2a (5HT2AR). These receptors are broadly expressed across the brain and next to nothing is known about whether a specific subset of those neural circuits mediate the effects of psychedelic drugs. Here, we began by designing a cellular-resolution brain-wide c-fos screen to comprehensively identify candidate brain regions and cell types activated by acute psychedelic agonism of 5HT2AR. To accomplish this, we screened for cfos expression after administration of the psychedelic 2,5-Dimethoxy-4-iodoamphetamine (DOI), and in two control conditions. 1. After administration of the selective 5HT2AR antagonist MDL100907, and 2. After administration of the non-psychedelic partial 5HT2AR agonist Lisuride Maleate. As expected, specific frontal cortical regions like the lOFC and anterior insular cortex traditionally associated with psychedelic action were particularly modulated by DOI, along with key regions in the mesolimbic dopamine system. Next, we sought to develop a behavioral paradigm for study of

psychedelics that was amenable to cross-species comparison. Previous studies have shown changes in human behavior on reinforcement learning tasks after psychedelic administration, and our lab developed a dynamic foraging behavior that is dependent on a network of frontal and mesolimbic circuits that overlaps substantially with our cfos-identified regions. Thus we hypothesized that DOI might acutely alter behavior in this dynamic foraging task. We found that after psychedelic administration, mouse reinforcement-learning strategy was significantly and specifically altered. Taken together, these results provide 1. a resource of candidate regions for follow-up in future work, and 2. a novel behavioral paradigm with a specific behavioral phenotype induced by psychedelic administration.

Disclosures: **A. Christensen:** None. **S. Starosta:** None. **E. Phelan:** None. **E. Stevens:** None. **M. Bergstrom:** None. **S.J. Brunwasser:** None. **K. Hengen:** None. **P. Osten:** None. **A. Kepecs:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.14

Topic: H.03. Decision Making

Support: Deutsche Forschungsgemeinschaft STA 1544/1-1

Title: Win-leave, loose-stay: counterintuitive foraging behavior under diminishing returns optimizes average reward rate

Authors: *S. STAROSTA[1], S. SHUVAEV[2], A. SIEBELS[3], D. KVITSIANI[4], A. KOULAKOV[2], A. KEPECS[1];
[1]Washington Univ. St. Louis, St. Louis, MO; [2]Cold Spring Harbor Lab., Cold Spring Harbor, NY; [3]Neurosci., Albert Einstein Col. of Med., New York, NY; [4]Mol. Biol. and Genet., Aarhus Univ. / DANDRITE, Aarhus C, Denmark

Abstract: We are continually confronted with decisions about whether to stay engaged with the current option or to switch to a new one. For instance, when to give up looking for the last berry on a raspberry bush and move on or when to settle down with a partner for life all involve “one choice” decisions about staying or switching. These decisions have been extensively studied as foraging decisions under diminishing returns, but little is known about what neuronal algorithms might support these.Here, we studied foraging decisions in mice facing the choice when to leave a depleting reward source. To identify the choice strategy, we devised several reward manipulations and observed, unexpectedly, that mice tend to leave a depleting source earlier after a higher than expected reward. Critically, this observation allowed us to distinguish between different classes of models because only one that implemented a decision rule comparing the next expected reward to the average of the previous rewards predicted this result. We show that this decision rule may be learned via a reinforcement learning method called R-

learning, but is not consistent with classical V-, or Q-learning models. Critically, the R-learning method explaining choice behavior optimizes average rate reward. To understand the neural underpinnings of choice behavior we performed optical recordings of dopamine neuron activity in the Ventral Tegmental Area (VTA). We observed that phasic VTA responses are best accounted for by a reward prediction error (RPE) based on R-learning, pointing to a potential learning mechanism that optimizes stay-or-leave choices.Our work offers an algorithmic decision rule and neuronal implementation for an ethologically relevant, understudied form of decisions, - foraging under diminishing returns.

Disclosures: S. Starosta: None. **S. Shuvaev:** None. **A. Siebels:** None. **D. Kvitsiani:** None. **A. Koulakov:** None. **A. Kepecs:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.15

Topic: H.03. Decision Making

Support: 5-R01-MH115027-04

Title: Acute psilocybin enhances behavioral flexibility via its action at 5HT2A receptors

Authors: *A. TORRADO PACHECO, R. OLSON, G. GARZA, B. MOGHADDAM; OHSU, Portland, OR

Abstract: The serotonergic psychedelic psilocybin has received considerable attention for its therapeutic potential in treating symptoms of psychiatric disorders when used in conjunction with psychotherapy. The biological mechanisms underlying this effect are not understood, but a potential hypothesis is that psilocybin enhances cognitive flexibility. The evidence for this hypothesis, however, is scant.
We addressed this question using a behavioral task designed to assess cognitive flexibility, in which animals are required to perform an instrumental choice according to one of two rules that involve different perceptual dimensions. Several extradimensional shifts occur during the task, requiring rats to detect the change in environmental contingencies and switch behavior accordingly to match the new rule. This task has been previously characterized and performance in it is sensitive to manipulations that are known to modulate cognitive flexibility. We evaluated the behavior of male and female rats in the task during baseline sessions after stable performance had been reached and compared it to sessions during which rats received treatment with psilocybin (1 mg/kg i.p.) and a series of other pharmacological manipulations.
Our results show a robust effect of psilocybin in improving performance in this task, that was not observed in saline-treated control animals. Interestingly, treatment with the serotonin 2A receptor (5HT2AR) agonist 2,5-Dimethoxy-4-iodoamphetamine (DOI) had no beneficial effect on cognitive flexibility, suggesting psilocybin’s pharmacological profile has unique and selective

effects. Since the major receptor targets of psilocybin are the 2A and 2C serotonin receptors, we repeated the experiment while blocking these receptors to determine psilocybin's mechanism of action. Pre-treatment with the 5HT2CR antagonist SB242084 did not impact psilocybin's effect on behavioral flexibility. Conversely, the 5HT2AR antagonist ketanserin blocked psilocybin's ability to improve performance in the task.
These data support the hypothesis that psilocybin positively impacts cognitive flexibility and further demonstrate that this effect is likely mediated by 5HT2A but not 5HT2C receptors.

Disclosures: A. Torrado Pacheco: None. **R. Olson:** None. **G. Garza:** None. **B. Moghaddam:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.16

Topic: H.03. Decision Making

Support: EMBO ALTF 384-2020
U19 NIH-NINDS BRAIN Initiative Award 5U19NS104648
SCGB AWD543027
K99MH120047

Title: How do cortical areas interact during evidence-based decision making?

Authors: *E. DIAMANTI[1], L. PINTO[2], O. KARNIOL-TAMBOUR[1], S. Y. THIBERGE[1], C. D. BRODY[1], J. PILLOW[1], D. W. TANK[1];
[1]Princeton Neurosci. Inst., Princeton Univ., Princeton, NJ; [2]Dept. of Neuroscience, Feinberg Sch. of Med., Northwestern Univ., Chicago, IL

Abstract: Complex behaviors require the involvement of distributed neural populations across the cortex (Pinto et al. 2019), with diverse areas often found to exhibit similar neural dynamics (Koay et al. 2022). Indeed, during a navigation-based decision-making task requiring evidence accumulation and post-stimulus memory, cortical areas form broad functional modules that are recruited throughout the decision-making process. Is this broad recruitment and the seemingly indistinguishable neural dynamics across the dorsal cortex the result of strong interactions between areas? And what is the nature and directionality of these interactions? We recorded simultaneously from 3 areas along the cortical hierarchy, visual area AM, retrosplenial cortex and premotor area M2 during an evidence accumulation task and used a switching non-linear dynamics model to estimate interareal communication signals. Recordings were performed using a random-access two-photon mesoscope (Sofroniew et al. 2016) to simultaneously image excitatory neurons across the three areas in 9 transgenic mice expressing the Calcium indicator GCaMP6s. Mice navigated a T-maze in virtual reality to receive water rewards from one out of two end arms (Pinto et al. 2018). In the first 200 cm of the maze's central stem, the 'cue region',

mice sampled salient visual cues presented on the side walls. The cue region was then followed by a 100 cm long ‘delay region’, after which mice had to make a turn to indicate the side with the highest total number of cues. Consistent with previous findings (Koay et al. 2022), activity across the population in each of the three areas formed sequences that spanned the whole trial. Support vector machines produced significant decoding of choice and evidence from the population activity of each area at each time point in the trial. Notably, we found only minor differences in decoding performance between areas. We then turned to the model to infer communication profiles between areas. Despite the apparent similarity in neural activity and decoding performance across areas, communication profiles differed. For instance, we found strong communication from M2 to AM but not vice versa. We are currently investigating these communication profiles, how do they evolve during the trial and how they vary with the animal’s behavioral state. Answering these questions will provide new insight into how diverse cortical areas work together to inform complex behaviors.

Disclosures: **E. Diamanti:** None. **L. Pinto:** None. **O. Karniol-Tambour:** None. **S.Y. Thiberge:** None. **C.D. Brody:** None. **J. Pillow:** None. **D.W. Tank:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.17

Topic: H.03. Decision Making

Support: Simons Collaboration on the Global Brain 543005
HHMI
Wellcome Trust
Pew Scholars Program
NIH Grant NS112312
NIH Grant NS113110
R01EB028171

Title: Brain-wide neural activity underlying memory-guided movement

Authors: *S. CHEN[1], Y. LIU[2], Z. WANG[3], L. LIU[5], H. HOU[6], N.-W. TIEN[7], S. DRUCKMANN[4], N. LI[5], K. SVOBODA[8];
[1]Howard Hughes Med. Inst., Howard Hughes Med. Inst., Ashburn, VA; [2]Stanford Univ., Sunnyvale, CA; [3]Stanford Univ., Palo Alto, CA; [4]Stanford Univ., Stanford Univ., Stanford, CA; [5]Baylor Col. of Med., Baylor Col. of Med., Houston, TX; [6]Janelia Res. Campus, Janelia Res. Campus, Ashburn, VA; [7]Natl. Inst. of Hlth., Natl. Inst. of Hlth., Bethesda, MD; [8]HHMI / Janelia Farm Res. Campus, Ashburn, VA

Abstract: Our brains can maintain information over timescales of a few seconds to guide future actions. Such 'short-term memory' (STM) serves as the brain's post-it note and is essential for

cognition, such as motor planning, decision-making, and foraging. The neural correlates of STM have been studied mostly one brain region at a time. For instance, neurons in the frontal cortex and related brain regions show persistent and slowly varying changes in spike rate that correlate with the maintenance of STM. Yet, how coordinated neuronal activity across inter-connected brain regions collectively maintains STM remains poorly understood. Anterior lateral motor cortex (ALM; part of M2) is an essential hub for the planning and execution of memory-guided directional licking. Taking an anatomy-guided approach, we targeted regions that form a brain-wide multi-regional network with ALM, including nuclei in the thalamus, basal ganglia, midbrain, medulla and others. We used up to 5 Neuropixels probes to simultaneously record the activity of hundreds of neurons across ALM connected brain regions during a memory-guided movement task. Neurons encoding sensory stimulus, choice, movement, and outcome, were widely distributed across the brain. Choice-selective neurons were more prevalent in the ALM connected network. Movements were encoded more strongly in premotor regions including brainstem and midbrain as compared to frontal cortex and thalamus. At the level of population activity, and on a trial-by-trial basis, functional interactions between brain areas with different task-related activities were recruited dynamically and changed over behavioral epochs. Inter-area correlation along a choice dimension ramped up during the motor planning phase and collapsed when the animal initiated movement, suggesting dynamical gating of functional coupling between brain areas according to cognitive and behavioral needs. Our data provide a foundation for understanding how STM and other neural computations are produced by multi-regional interactions across the brain.

Disclosures: S. Chen: None. **Y. Liu:** None. **Z. Wang:** None. **L. Liu:** None. **H. Hou:** None. **N. Tien:** None. **S. Druckmann:** None. **N. Li:** None. **K. Svoboda:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.18

Topic: H.03. Decision Making

Support: KAKENHI JP16H06567 (to M.M.)
JST CREST JPMJCR1853 (to M.M.)
KAKENHI JP19K16890 (to M.N.)

Title: Optogenetic stimulation of dopamine signals transmitted to the ventral striatum during ongoing decision-making process affects economic choice behavior in macaque monkeys

Authors: *M. NEJIME[1], M. YUN[1], T. KAWAI[1], J. KUNIMATSU[1], H. YAMADA[1], K.-I. INOUE[2], M. TAKADA[2], M. MATSUMOTO[1];
[1]Fac. of Med., Univ. of Tsukuba, Tsukuba, Ibaraki, Japan; [2]Primate Res. Institute, Kyoto Univ., Inuyama, Aichi, Japan

Abstract: Midbrain dopamine (DA) neurons are thought to play a crucial role in economic decision-making. These neurons are activated when the outcome of choice is better than expected, and their activation reinforces choices leading to better-than-expected outcomes. Notably, this type of DA neuron activation occurs after decisions are made. On the other hand, DA neurons are activated or suppressed when choice options are offered (i.e., before decisions are made). It remains unclear whether and how DA signals evoked by options affect upcoming choice behavior. In the present study, we focused on the DA pathway from the midbrain to the ventral striatum (VS), which receives dense projections from DA neurons, in macaque monkeys and examined the causal relationship between choice behavior and option-evoked DA signals transmitted through this pathway. The monkeys were trained to perform an economic decision-making task in which an option was offered and the animals were required to decide whether to choose this option based on the value. Using optogenetics applicable to the monkey brain, we stimulated DA axon terminals in the VS when the option was offered. We found that the monkeys more often chose the option when DA axon terminals were stimulated than not stimulated. To investigate the electrophysiological basis of the causal contribution of DA signals transmitted to the VS to choice behavior, we next recorded single-unit activity from DA and VS neurons in the monkeys performing the task. We found that both DA and VS neurons represented diverse signals related to economic decision-making. Some neurons represented the value of the offered option (value type), some represented whether or not the monkeys would choose the option (choice type), and some represented a combination of the value and choice (intermediate type). Notably, the dynamics of these signals were corresponding to the economic decision-making process. Many DA and VS neurons encoded the value of the option after it was offered, then gradually changed their activity to represent the animal's upcoming choice. However, the dynamics of DA and VS neurons were somewhat different. While the proportions of the three types of DA neurons increased with shorter latencies after the onset of the option and decreased before the monkeys executed an action to choose the option, those of the three types of VS neurons gradually increased with longer latencies and kept increasing even after the onset of the action. Our findings suggest that DA neurons linking to the VS form a key circuit that transforms value information into choice command, and that DA signals transmitted to the VS causally contribute to this decision-making process.

Disclosures: **M. Nejime:** None. **M. Yun:** None. **T. Kawai:** None. **J. Kunimatsu:** None. **H. Yamada:** None. **K. Inoue:** None. **M. Takada:** None. **M. Matsumoto:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.19

Topic: H.03. Decision Making

Support: R01-EY-022411

Title: Perceptual decision-related single-neuron activity in the monkey subthalamic nucleus

Authors: K. ROGERS[1], *L. DING[2];
[1]Dept. of Neurosci., Univ. of Pennsylvania, Phliadelphia, PA; [2]Dept. of Neurosci., Univ. of Pennsylvania, Philadelphia, PA

Abstract: The basal ganglia (BG) consist of multiple nuclei and are thought to influence cognitive functions like motor planning, learning, and perceptual decision-making. The subthalamic nucleus (STN) is a part of the indirect and hyperdirect pathways in the BG. Several hypotheses have been proposed for STN's involvement in perceptual decision making, based largely on single-neuron recordings of STN activity in animals performing non-perceptual decision-making tasks and aggregate neural recordings (such as fMRI and local field potential signals) in human subjects. These hypotheses include: 1) STN's output is a nonlinear summation of accumulated evidence across choice options (Bogacz and Gurney, 2007). This model translates to a gradual increase in STN activity over time equally for both choices; 2) STN provides transient suppression that disappears over time for both choices (Ratcliff and Frank, 2012). This translates to an initial burst in STN activity followed by a gradual decrease; and 3) STN inhibits SNr activity until enough evidence has been accumulated to overcome this inhibition and trigger a decision commitment (Wei, et al. 2015). This translates to a rise in STN activity during evidence accumulation for both choices until shortly before decision commitment with a plateau occurring for the ipsilateral choice. These predictions of STN activity patterns during decision formation have not been tested directly.
In this study, we recorded single-unit STN activity in two monkeys performing a random-dot visual motion discrimination saccade task. The monkeys made saccades at a self-chosen time to indicate their perception of the global motion direction of a random-dot kinematogram. For each trial, the motion strength and direction were randomly chosen from 5 values and two directions, respectively. The monkey was given a liquid reward for a correct choice. We screened for neurons with activity modulation during the task and obtained recordings from 99 STN neurons. In this sample, we observed the presence of all three types of activity patterns described above, with the second type being more dominant. Certain neurons also showed activity patterns that deviate from all three predictions. These results suggest complex involvement of the STN in perceptual decision making, which may be best captured by a revised model that incorporates previous hypotheses.

Disclosures: K. Rogers: None. **L. Ding:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.20

Topic: H.03. Decision Making

Support: R01 EY015260

Title: Context-dependent sensory adaptation in cortical area MT as a substrate of adaptive inference in uncertain and unstable environments

Authors: *K. D. MCGAUGHEY[1], N. TARDIFF[3], K. SCHAPIRO[3], H. LEFUMAT[4], J. I. GOLD[2];
[1]Univ. of Pennsylvania Neurosci. Grad. Group, PHILADELPHIA, PA; [2]Dept Neurosci, Univ. of Pennsylvania Neurosci. Grad. Group, Philadelphia, PA; [4]Dept. of Neurosci., [3]Univ. of Pennsylvania, Philadelphia, PA

Abstract: Visual decisions typically require accumulation of uncertain sensory information to make inferences about the state of the world. This inference process is complicated by the fact that the state of the world can undergo unpredictable changes, which often require adjustments in how sensory information is accumulated. Previously, we modeled optimal, adaptive inference in these changing environments. One key feature of our model is a leakage of accumulated evidence, where the rate of the leak depends on environmental stability. For example, in increasingly unstable environments, where unexpected changes can affect the relevance of recently accumulated evidence, the leak rate increases. This increased leak rate effectively removes older, pre-change evidence and attenuates beliefs about the pre-change state that would hinder choice accuracy. Whereas this context-dependent leak can describe human decision-making behavior, it remains unknown exactly where or how it may be implemented in the brain. We propose the leak is implemented, at least in part, via sensory adaptation during evidence encoding. Specifically, we hypothesize that the rate of change of incoming sensory information modulates the dynamics of evidence encoding, enabling the decision process to adapt to expectations about environmental stability.To test this hypothesis, we trained two rhesus macaques on a random-dot motion direction-discrimination task in which we manipulated sensory uncertainty (via motion coherence) and instability (via “change-points” or abrupt switches in motion direction). Preliminary data (*n = 32* sessions Monkey Ch; *n = 36* sessions Monkey An) suggest that the monkeys, like human subjects, can adapt their decision-making behavior to the stability of the current stimulus context. Specifically, the monkeys were less biased by pre-change evidence and less sensitive to current evidence in more unstable environments (more change-points), which is consistent with a leak in evidence accumulation that depends on stimulus context. We collected preliminary electrophysical data *(n = 12* sessions Monkey An) to explore whether this leaky evidence accumulation
could be implemented via context-dependent sensory adaptation in middle temporal area (MT). Single-unit responses can adapt to preferred-direction motion and the time course of this adaptation can depend on the recently experienced switch rate of the motion stimulus, suggesting that the stability of recent stimulus statistics affects evidence encoding. Together, this work implies that context-dependent adaptation of sensory encoding can help to optimize decision-making in unstable and uncertain environments.

Disclosures: K.D. McGaughey: None. **N. Tardiff:** None. **K. Schapiro:** None. **H. Lefumat:** None. **J.I. Gold:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.21

Topic: H.03. Decision Making

Support: NIDA DA051977
NIDA DA051598

Title: Improvements in decision-making during adolescence predict cocaine-taking behaviors in adulthood

Authors: *P. VILLIAMMA[1,2], J. CASBY[3], S. M. GROMAN[1,2];
[1]Neurosci., [2]Med. Discovery Team On Addiction, [3]Mol. Pharmacol. and Therapeut. Grad. Program, Univ. of Minnesota, Minneapolis, MN

Abstract: Drug-taking is known to disrupt reward-based decision-making but not all individuals become addicted or have the same extent of changes in decision-making. Poor performance in reward-based decision-making tasks is associated with greater self-administration of drugs of abuse. Although the mechanisms leading to these decision-making disruptions are not known, our recent work has found that the degree of improvement in reward-based decision-making during adolescence is predictive of decision-making in adulthood. These data suggest that deviations in select adolescent neurodevelopmental trajectories may enhance drug use susceptibility in adulthood. We hypothesized, therefore, that poor adolescent decision-making trajectories would be predictive of greater cocaine-taking behaviors in adulthood. To test this hypothesis, we trained and tested female and male Long Evans (N=56) rats on a three-choice, spatial reversal-learning task. Rats were assigned to either a cross-sectional study (N=40) and underwent a single round of testing on the reversal-learning tasks at P35, P55, or P75, or to a longitudinal study (N=16) and were repeatedly tested on the reversal-learning tasks at the same ages (P35→P55→P75). All rats then underwent a single assessment in adulthood (P120). A subset of rats (N=34) was trained to self-administer cocaine in 6h daily sessions for 14 days. We found that reversal-learning performance improved during adolescence in both longitudinal and cross-sectional groups. This improvement in performance was due specifically to an increase in positive-outcome updating, and not negative-outcome updating or the retention of value. We then examined whether individual differences in the degree of improvement in positive-outcome updating during adolescence would predict decision-making and cocaine-taking behaviors in adulthood. We found that rats with a more attenuated positive-outcome updating trajectory performed worse in the reversal-learning task and took significantly more cocaine compared to rats with a steeper trajectory. These relationships were specific to the rate of change in positive-outcome updating during development. Positive-outcome updating at any singular adolescence age from the cross-sectional study was not a predictor of cocaine-taking behaviors in adulthood. These data suggest that the neurodevelopmental changes that occur during adolescence are critical regulators of addiction susceptibility. Our ongoing studies are utilizing immune, proteomic, and genomic approaches to investigate the neurodevelopmental changes mediating decision-making trajectories and drug use susceptibility.

Disclosures: P. Villiamma: None. **J. Casby:** None. **S.M. Groman:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.22

Topic: H.03. Decision Making

Support: NIDA DA051977
NIDA DA051598

Title: Oral oxycodone self-administration is greater in female rats compared to male rats and associated with disruptions in positive feedback updating

Authors: *K. P. LAROCCO[1,2], P. VILLIAMMA[1,2], S. M. GROMAN[1,2,3];
[1]Dept. of Neurosci., [2]Med. Discovery Team on Addiction, [3]Dept. of Psychology, Univ. of Minnesota, Minneapolis, MN

Abstract: Oxycodone is one of the most highly prescribed medications for pain and because of its rampant misuse, is believed to be the primary driver of the ongoing opioid epidemic. Not all individuals that are prescribed oxycodone, however, misuse the drug or become addicted. Evidence suggests that some individuals may be more vulnerable to developing an opioid use disorder (OUD) compared to others. Identification of individuals at risk for developing an OUD could be an important step in preventing oxycodone addiction. The current study sought to develop a self-administration paradigm that would be able to model this vulnerability in rats and determine if decision-making phenotypes assessed before drug use would predict oxycodone-taking behaviors. Long Evans rats (78 M/78 F) were trained and tested on a three-choice, probabilistic reversal-learning (PRL) paradigm. Rats were then trained to orally self-administer either a saccharin solution (0.05%; N=41) or oxycodone in a saccharin solution (0.05 mg/kg/infusion; N=115) in 3 h daily sessions. On the ninth session, the dose of oxycodone increased, and saccharin was gradually removed from the solution. Self-administration behaviors were assessed for an additional 14 days followed by tests of motivation, extinction, and cue-induced reinstatement. Rats found oral oxycodone to be highly reinforcing and continued to self-administer the drug in the absence of any sweetener. There was significant variation in the degree to which rats escalated their oxycodone use: ~33% of rats increased their intake, ~33% decreased their intake, and ~34% maintained a stable intake. Notably, rats that increased their oxycodone intake responded more during a progressive ratio test, extinction, and cue-induced reinstatement. Our oral self-administration paradigm, therefore, appears to capture the transition from use to misuse in some individuals. We then examined the differences between male and female rats. Female rats took significantly more oxycodone and were more likely to escalate their oxycodone intake compared to males. Moreover, females responded more during tests of motivation, extinction, and reinstatement. Females are, therefore, more likely to transition from oxycodone use to misuse compared to males. Female rats also performed worse than males in the PRL task and this was due to disruptions in value updating following a positive outcome. We propose that greater oxycodone-taking behaviors in female rats are due to pre-existing disruptions in positive-feedback updating. Our novel oxycodone self-administration paradigm

provides a unique platform for conducting biological and systems-level analyses of OUD vulnerability.

Disclosures: K.P. LaRocco: None. **P. Villiamma:** None. **S.M. Groman:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.23

Topic: H.03. Decision Making

Support: NIH/NIDA IRP

Title: Effects of cariprazine and buprenorphine on economic choice between remifentanil and food in squirrel monkeys.

Authors: *J. HECKER, A. AMIRALI, D. EFFINGER, R. MONTORO, C. VOGT, A. NEWMAN, C. SCHINDLER, H. JEDEMA, C. BRADBERRY;
NIH, NIDA IRP, Baltimore, MD

Abstract: Opioid maintenance therapy (OMT) utilizes mu-opioid agonists such as buprenorphine to clinically treat those suffering from opioid use disorder (OUD). Buprenorphine is a partial mu-opioid agonist and has a longer half-life than fast-acting opiates such as heroin, rendering it useful for alleviating withdrawal symptoms. With its low receptor intrinsic activity, it may be a preferred treatment as it has a lower rate of overdose-related deaths. Using such an established treatment on a new task allows us to compare data with a variety of non-conventional therapeutics for OUD. For example, cariprazine is a partial D2/D3 agonist currently used as an atypical antipsychotic to treat Schizophrenia and Type 1 Bipolar Depression, and recent studies suggest that it improves symptoms associated with comorbid substance use disorders. Furthermore, cariprazine is currently being evaluated in clinical trials to examine its impact on drug use in patients with comorbid cocaine and opioid use disorders. In the present study we compared the effects of pretreatment with buprenorphine and cariprazine on opioid preference in an economic choice task that tracks a subject's valuation and choice of drug and non-drug rewards. Adult male squirrel monkeys received clinically relevant doses of buprenorphine (0.1-0.32mg/kg) or cariprazine (10-100µg/kg) intravenously for 5 consecutive days approximately 22 hours prior to a touch screen choice task in which subjects selected between differing quantities of remifentanil, a fast-acting opioid, and sweetened condensed milk. Monkeys that received buprenorphine treatment showed a significant reduction in drug preference compared to baseline choice allocation during the preceding week, consistent with its demonstrated efficacy in OMT. In contrast, cariprazine treatments did not show significant reductions in drug preference in the dose range tested. Future studies will determine if plasma concentrations for cariprazine and its metabolites appear adequate based on other preclinical and clinical studies. We also plan to assess the potential of additional non-opioid treatments by evaluating their effect on drug choice.

Disclosures: **J. Hecker:** None. **A. Amirali:** None. **D. Effinger:** None. **R. Montoro:** None. **C. Vogt:** None. **A. Newman:** None. **C. Schindler:** None. **H. Jedema:** None. **C. Bradberry:** None.

Poster

566. Neural Mechanisms of Decision Making: Choice: Single Unit to Brain Wide Effects

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 566.24

Topic: H.03. Decision Making

Support: T32 Fellowship MH018399-35

Title: Population Activity of Single Units in Cortex and Striatum Drive Behavior Toward Optimal Choice in Rats Performing a Temporal Discounting Task

Authors: *M. J. FRANCOEUR, D. RAMANATHAN;
Psychiatry, UC San Diego, La Jolla, CA

Abstract: Information about actions and their outcomes are used to create value representations and stored to inform future choices. Past research identifies the role of discrete brain areas such as ventral striatum, prefrontal, and orbitofrontal cortex in value-based decision making. These areas are the primary projections in the cortico-striatal pathway and therefore likely work in a dynamic network to inform and update value representations. Previously, we have observed beta-frequency oscillations which reliably signal reward outcome throughout areas of the cortico-striatal circuit. In a temporal discounting task, we find that cortical brain regions are most positively correlated with high value choice whereas nucleus accumbens is negatively correlated with low reward choice. Thus, we predict there may be two different functional circuits responsible for driving behavior toward the optimal high value choice. To further explore reward processing in the cortico-striatal network we used 32-CH Harlan microdrives to record neural activity from 18 male Long-Evans rats performing a temporal discounting task. Single units were collected from subdivisions of medial prefrontal cortex, orbitofrontal cortex, and nucleus accumbens by driving from dorsal to ventral and splitting tetrode placement between medial/ lateral divisions when needed (4 tetrodes medial, 4 tetrodes lateral). We recorded ~2,000 neurons from dorsal/ventral prefrontal cortex (N=6 rats; 400 units), medial/lateral orbitofrontal cortex (N=5 rats; 800 units), nucleus accumbens core/ shell (N=7 rats; 800 units). In our temporal discounting task, rats choose to either wait for a high value (3x water) reward or to receive a low value (1x water) reward delivered immediately (500ms). In the first block of trials ($\leq$ 65 trials) the high value reward is delayed 2s, and in the second block (>65 trials) is delayed 10s. Rats do show behavioral differences based on temporal delay (main effect of block $p<0.001$). On average, rats made 76.4 +/ 22.5% high value choices in block 1 (2s delay) and only 21.8+/ 34.0% high value choices in block 2 (10s delay). We find activity related to action, reward anticipation, delay length, reward outcome and magnitude discrimination. A subset of units showed magnitude preferences and modulated activity from between blocks. Moreover, we find spiking activity is coherent with oscillations during reward outcome specifically at beta-frequencies (15-

30Hz), further supporting our hypothesis that beta oscillations reflect the local population activity of neurons and may represent a widely distributed value signal throughout the cortico-striatal network.

Disclosures: M.J. Francoeur: None. **D. Ramanathan:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.01

Topic: H.03. Decision Making

Support: NIH Grant R00DA041493
Bruce/Jones Graduate Fellowship

Title: Cocaine intake correlates with subsequent risk-taking behavior and affects estrous cycling in female rats.

Authors: *L. TRUCKENBROD[1], E. M. COOPER[2], C. A. ORSINI[3];
[1]Univ. of Texas, Austin, TX; [2]The Univ. of Texas at Austin, Austin, TX; [3]Psychology, The Univ. of Texas at Austin, AUSTIN, TX

Abstract: Substance use disorders (SUDs) are characterized by forms of compromised decision making, such as elevated risk taking. Understanding the mechanisms by which risk taking is increased in individuals with these disorders is crucial to developing effective treatments and reducing risk of relapse. Previous work using animal models has corroborated increased risk taking observed in men and women with SUDs. Level of drug intake, however, varies across individuals (humans and rodents alike), raising the possibility that those individuals that consume more drugs may display a greater elevation in risk taking. The main goal of this project was to therefore test whether level of drug intake correlates with risk taking during abstinence. Male and female Sprague-Dawley rats were implanted with jugular catheters and underwent 14 days of long-access cocaine (or sucrose) self-administration. Three weeks after the cessation of self-administration, rats were trained and tested in a rodent model of risky decision making ("Risky Decision-making Task"; RDT) in which rats chose between a small, "safe" reward and a large, "risky" reward accompanied by an increasing probability of a mild footshock delivery. Throughout each phase of the experiment, the estrous cycle of females was monitored to determine whether chronic cocaine exposure disrupted hormonal cyclicity. Our findings show that greater cocaine intake during self-administration was positively correlated with greater choice of the large, risky reward (risky choice), a relationship that was absent in sucrose controls. Additional analyses compared risk taking between females categorized as "high cocaine intake" and "low cocaine intake" (based on a median split) and revealed that females in the "high intake" group chose the large, risky reward significantly more than females in the "low intake" group. This relationship between cocaine intake and risky choice was unique to females, as identical

analyses in males did not reveal significant correlations between cocaine intake and risky choice. Cocaine exposure also caused significant disruptions in estrous cyclicity in females, irrespective of intake group. During self-administration and abstinence, rats displayed repetitive estrus phases, long periods of diestrus, and few proestrus phases, and this estrous cycle irregularity persisted into RDT behavioral testing. These findings are consistent with previous work showing that cocaine exposure disrupts estrous cyclicity and suggests that cocaine-induced increases in risk taking in females may be due to disruption of the natural fluctuations of ovarian hormones across the estrous cycle.

Disclosures: L. Truckenbrod: None. **E.M. Cooper:** None. **C.A. Orsini:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.02

Topic: H.03. Decision Making

Support: R21DA053462
R00DA041493

Title: Fentanyl self-administration increases risk-taking behavior in male rats.

Authors: *A.-R. WHEELER[1], L. M. TRUCKENBROD[2], E. M. COOPER[3], B. SETLOW[5], C. A. ORSINI[4];
[1]Univ. of Texas at Austin, Austin, TX; [2]Univ. of Texas, Austin, TX; [3]The Univ. of Texas at Austin, Austin, TX; [4]Psychology, The Univ. of Texas at Austin, AUSTIN, TX; [5]Dept. of Psychiatry, Univ. of Florida, Gainesville, FL

Abstract: Risky decision making involves the ability to weigh risks and rewards associated with different options in order to make adaptive choices. Individuals with opioid use disorder display impaired risky decision making, exhibiting exaggerated risk-taking behavior, which may render them more vulnerable to relapse during periods of abstinence. To better understand causal relationships between opioid use and risk taking, our lab employs a rodent model of decision making involving risk of explicit punishment (the Risky Decision-making Task; RDT). In this task, rats make discrete choices between a small, safe food reward and a larger food reward that is accompanied by an increasing risk of mild footshock punishment across the behavioral test session. Using this model, we have shown that chronic cocaine exposure increases choice of the large, risky reward (increased risk taking) in males and females. The goal of this experiment was to determine if chronic exposure to the synthetic opioid fentanyl has similar effects on risk taking in males. Male Sprague Dawley rats were implanted with jugular catheters, and half of the rats underwent long-access fentanyl self-administration (6h/day for 14 days) and the other half underwent sucrose self-administration. Following self-administration, rats remained undisturbed in their home cages for 3 weeks before being trained on the RDT to assess fentanyl-induced

changes in risk taking. Rats that self-administered fentanyl chose the large, risky reward significantly more than rats that self-administered sucrose. Increased risk taking was accompanied by a reduction in lose-shift behavior in the fentanyl group, suggesting that the fentanyl-induced increase in risk taking is due to insensitivity to punishment. To confirm that the increased risk taking in the fentanyl group is not secondary to changes in food motivation or shock sensitivity, rats were then tested in control assays that assessed willingness to work for food and shock reactivity. There were no differences between groups on either measure. Lastly, to determine if increased risk taking was due to behavioral inflexibility, rats were tested on a probabilistic reversal learning task. Surprisingly, rats in the fentanyl group completed significantly more reversals than rats in the sucrose group, indicating that cognitive inflexibility does not account for fentanyl-induced increases in risk taking. Future experiments will extend this work to females to determine whether there are sex differences in fentanyl-induced changes in risk taking.

Disclosures: A. Wheeler: None. **L.M. Truckenbrod:** None. **E.M. Cooper:** None. **B. Setlow:** None. **C.A. Orsini:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.03

Topic: H.03. Decision Making

Title: Evaluating cognitive deficits as a non-motor symptom in Parkinson's disease using a 6-hydroxydopamine lesion in the substantia nigra

Authors: *T. AGUIRRE, H. CHAMSEDDINE, E. RUSNAK, K. VAZQUEZ, C. ARGENBRIGHT, P. N. FUCHS;
The Univ. of Texas at Arlington, Arlington, TX

Abstract: Motor symptoms in Parkinson’s Disease (PD) have been well studied, however, non-motor symptoms, including pain, fatigue, insomnia and cognitive deficits, are still being further analyzed. PD is caused by a loss of dopaminergic neurons in the substantia nigra, which result in the rigid, stiff and slow movement associated with PD. It is also well known the substantia nigra and dopamine play a role in reward and motivation, both key components in higher cognitive processes. The purpose of this project is to potentially expose a connection between the depletion of dopamine in the substantia nigra, as depicted in PD, and a decrease in cognitive abilities in a rodent model. In this study, forty Sprague Dawley female rats were lesioned with a neurotoxin, 6-hydroxydopamine (6-OHDA), in the substantia nigra to evaluate a non-motor symptom associated with PD: cognitive deficits. Decision-making was the specific cognitive impairment being examined using a novel version of the Rat Gambling Task (RGT). All forty animals underwent operant training to learn to single and dual lever press and then learned the RGT to establish a baseline. Once completed, all forty rats underwent stereotaxic surgery to microinject a

total of 1 microliter saline (20 control animals) or the vehicle solution containing 6-OHDA (8 ug/ul) and 0.1% ascorbic acid dissolved in saline (20 experimental animals). After two weeks, the animals were subjected to test for RGT, as well as the Open Field Task. Upon completion of testing, animals underwent a transcardial perfusion to fix brain tissue, which was then extracted, sliced and stained for tyrosine hydroxylase using immunohistochemistry. Results found that after two weeks, there was a significant difference between the saline and 6-OHDA lesioned animals, in which the 6-OHDA lesioned animals performed worse on the Rat Gambling Task, perhaps indicating that the depletion of dopamine within the substantia nigra did have an impact on the motivation and reward needed to produce advantageous decisions. With more research, we could come to a better understanding of these neural pathways that involve cognitive processes when a neurodegenerative disease is present.

Disclosures: **T. Aguirre:** None. **H. Chamseddine:** None. **E. Rusnak:** None. **K. Vazquez:** None. **C. Argenbright:** None. **P.N. Fuchs:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.04

Topic: H.03. Decision Making

Support: China Scholarship Council - University of St Andrews Scholarship (PhD programmes): 201708060006

Title: Midbrain dopamine neuron number correlated with loss-stay behavior under risk in male rats, and did not increase with extra environmental enrichments

Authors: *Y. LI, S. VENTURA, D. TAIT, J. AINGE, E. BOWMAN;
Sch. of Psychology and Neurosci., Univ. of St Andrews, St Andrews, United Kingdom

Abstract: Midbrain dopamine neurons project extensively to the rest of the brain and play a crucial role in learning and reward-processing. Previous research from our laboratory focused on loss-stay behavior during risky decision-making—the repeating a choice that had led to a loss immediately previously. Huttunen (2016, unpublished doctoral dissertation) discovered a significant correlation between loss-stay behavior in male rats and dopamine neuron number in the Substantia Nigra *pars compacta* (SNpc)—a structure involved in reinforcement computations during trial-and-error learning. This was surprising because the homeostatic dopaminergic system would be expected to prevent individual variations in neuron number from creating observable behavioral differences. We aimed to further investigate the link between dopamine structure and function in risk decision-making, and to explore the possible influence of age and environmental enrichment on this relationship. Across three experiments, we tested a total of 64 Lister Hooded rats (11 females, 53 males) in a rat Balloon Analogue Risk task to assess their risk- and loss- related behaviors. Rats were randomly assigned to the standard or extra

enrichment conditions. The latter received more enriching items in their home cages and 1 hour of playtime every weekday for at least 3 months. By the end of the experiments, 12 rats were young (*c.*16 weeks), 38 were middle-aged (*c.*47 weeks) and 14 were aged (*c.*60 weeks). We quantified the number of putative dopamine neurons in the midbrain using immunohistochemical staining with *Tyrosine Hydroxylase*. Results showed a significant correlation between loss-stay behavior and dopamine neuron number in the SNpc, and this correlation was only significant when age was controlled in the analysis. Environmental enrichment preserved rats' visual object recognition memory against age-related decline, but it did not alter dopamine neuron number, hippocampal neurogenesis or other risk- and stress-related behaviors. The lack of enrichment effect on dopamine neuron count contradicted the *c.*17% dopamine neuron increase in male mice after 2 weeks of environmental enrichment (Aumann, Tomas & Horne, 2013). In conclusion, dopamine neuron number in the SNpc correlated loss-stay behavior, which was mediated by age but not environmental enrichment. A candidate mechanism underlying our findings may be neurotransmitter phenotype switching.

Disclosures: Y. Li: None. **S. Ventura:** None. **D. Tait:** None. **J. Ainge:** None. **E. Bowman:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.05

Topic: H.03. Decision Making

Support: NIH Grant R01DA051295
NIH Grant R25MH081482

Title: Sex-dependent effects in dopaminergic modulation of risky decision-making in rats

Authors: *S. M. AYOUB[1], A. M. LIBSTER[1], S. DULAWA[1], J. YOUNG[1,2];
[1]Psychiatry, Univ. of California San Diego, San Diego, CA; [2]Res. Service, VA San Diego Healthcare Syst., San Diegp, CA

Abstract: Psychiatric disorders known to impair DA functioning (i.e., schizophrenia, bipolar disorder) worsen performance in risky decision-making tasks, such as the Iowa Gambling Task, while DA agonist treatment, e.g., pramipexole (PPX), to people with Parkinson's disease, result in gambling problems. Using a recently developed task, Zalocusky (2016) demonstrated the risk preference of male rats can be increased by systemic administration of PPX and decreased by optogenetic silencing of DA-2 receptor (DA-2R) expressing neurons in the nucleus accumbens (NAc). Sex-dependent differences were not examined however, and the role of sex in risky decision-making remains unclear. Here, we trained female and male rats in the same task to explore sex-differences in risk preference at baseline and in response to pharmacological challenges of PPX, and the DA-2R antagonist sulpiride (SUL). In operant boxes animals could

choose from one of two nose-pokes, one that delivered a 50 µl strawberry milkshake reward (safe-option), and the other a 10 µl reward with a 75% probability and 170 µl reward with a 25% probability (risky-option). Once trained to a stable baseline of risk preference, rats were treated with PPX (0.15 or 0.3 mg/kg; Experiment 1) or SUL (30 mg/kg; Experiment 2) for 3 days, each separated by a saline washout. Baseline: females were less risk-adverse/more risk-prone than males. Experiment 1: collapsed across drug and saline tests, there was a main effect of drug on percent risk choice (%RC) change from baseline [$F(1,18) = 10.5$, $p < 0.01$], with PPX increasing %RC. When analyzed across each testing day, a main effect of session [$F(6,108) = 3.6$, $p < 0.005$] was observed, as was a session * sex * drug interaction [$F(6,108) = 2.2$, $p < 0.05$]. *Post hoc* analyses revealed females differed from males in the timing of their response to PPX based on the dose administered. Experiment 2: collapsed across drug and saline tests, there was a main effect of drug on %RC change from baseline [$F(1, 6) = 20$, $p < 0.01$], with SUL decreasing %RC. There was also a trend toward a drug * sex interaction [$F(1, 6) = 3.6$, $p = 0.11$], with more pronounced attenuation of %RC by SUL in females. A similar pattern was observed when data was analyzed across all drug and saline tests. Together this data indicate a sex-specific modulation of baseline risk preferences, and female rats may be more sensitive to DA manipulations on risky decisions, highlighting the necessity of tracking sex-based differences in such tasks. Ongoing studies will determine whether DA-2R NAc activity reveal a similar sex-specific change in DA during risk-preference.

Disclosures: **S.M. Ayoub:** None. **A.M. Libster:** None. **S. Dulawa:** None. **J. Young:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.06

Title: WITHDRAWN

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.07

Topic: H.03. Decision Making

Support: NIH Grant DA053014

Title: Distinct roles of striatal dSPNs and iSPNs in flexible decision making under uncertainty

Authors: *J. BAHUGUNA[1], J. K. BADYNA[2], K. BOND[1], M. CLAPP[1], C. VICH[4], E. A. YTTRI[3], J. E. RUBIN[5], T. D. VERSTYNEN[2];
[1]Psychology and Neurosci. Institute, Dept. of Psychology, [3]Biol. Sci., [2]Carnegie Mellon Univ., Pittsburgh, PA; [4]Univ. de les Illes Balears, Majorca, Spain; [5]Mathematics, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Striatal direct pathway (dSPN) and indirect pathway spiny projection neurons (iSPNs) are critical in controlling and shifting decision policies in response to environmental feedback. We studied the role of striatal dSPNs and iSPNs by optogenetically stimulating them in D1-cre and A2A-cre mice resp. while they performed a two-armed bandit task with probabilistic rewards (conflict) and sudden changes in action-outcome contingencies (volatility). We first showed that the behavioral data from mice for all levels of conflict and volatility could be reproduced with a biologically based spiking neural network model of the cortico-basalganglia-thalamic (CBGT) circuit, with a dopamine dependent spike timing dependent plasticity rule. The behavioral data (accuracy, reaction times) from the CBGT network and mice were mapped onto a drift diffusion model (DDM) that describes the process of decision making as a noisy accumulation of evidence up to a decision threshold. We observed that, in both model and mice, a switch in reward contingencies (block change point) induced a sharp drop in drift rate followed by a recovery indicating a slowing down of evidence accumulation on encountering errors and a return to the original rate of evidence accumulation over the next few trials. Higher probabilities of reward for a correct choice led to larger drops in drift rate at the change point. The boundary height showed a slight decrease at the block change point and on average showed a slight decrease for higher volatility. In the CBGT model, changes in drift rate were associated with differences in the balance of dSPN and iSPN activation levels, both within and across action channels, while boundary height was associated with overall iSPN firing rates. In mice, dSPN stimulation induced an increase in average drift rate at change point and recovery, as predicted by the model, along with a decrease in boundary height compared to control animals. Interestingly, iSPN stimulation yielded a decrease in average drift rate at change point and recovery and increase in boundary height, as predicted by the model, as compared to control animals. Overall, the parameter alterations that we find suggest that change points switch behavior into an exploratory regime. The exaggerated increase in the boundary height and decrease in drift rates induced by iSPN stimulation indicate a shift to a slow exploration policy, in contrast with dSPN stimulation, which pushes the animal into a regime of much faster exploration. These results align with findings from our CBGT model, highlighting how direct and indirect pathways may drive distinct changes in information processing in response to changes in the environment.

Disclosures: J. Bahuguna: None. **J.K. Badyna:** None. **K. Bond:** None. **M. Clapp:** None. **C. Vich:** None. **E.A. Yttri:** None. **J.E. Rubin:** None. **T.D. Verstynen:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.08

Topic: H.03. Decision Making

Support: 111 project (Base B16018)
The National Natural Science Foundation of China (NSFC)

Title: The rat frontal cortex encodes a value map in support of economic decisions under risk

Authors: X. ZHU[1], ***C. BAO**[2], J. MOLLER-MARA[1], J. LI[2], S. DUBROQUA[1], J. C. ERLICH[2]; [1]NYU Shanghai, Shanghai, China; [2]Sainsbury Wellcome Ctr., Univ. Col. London, London, United Kingdom

Abstract: Neurons in frontal and parietal cortex encode task variables during decision-making, but causal manipulations of the two regions produce strikingly different results. For example, silencing the posterior parietal cortex (PPC) in rats and monkeys produces minimal effects in perceptual decisions requiring integration of sensory evidence, but silencing frontal cortex profoundly impairs the same decisions. Here, we tested, for the first time, the causal roles of the rat frontal orienting field (FOF) and PPC in economic choice under risk. On each trial, rats chose between a lottery and a small but guaranteed surebet. The magnitude of the lottery was independently varied across trials and was indicated to the rat by the pitch of an auditory cue. As in perceptual decisions, both unilateral and bilateral PPC muscimol inactivation produced minimal effects. FOF inactivation produced substantial changes in behavior even though our task had no working memory component. We quantified control and bilateral inactivation behavior with a multi-agent model consisting of a mixture of a ’rational‘ utility-maximizing agent ($U=V^{\rho}$) with two ’habitual‘ agents that either choose surebet or lottery. Silencing PPC produced no significant shifts in any parameters relative to controls. Effects of FOF silencing were best explained by a decrease in ρ, the exponent of the utility function. This effect was parsimoniously explained by a dynamical model where the FOF is part of network that performs sensory-to-value transformations. To test our model prediction, we recorded neural activity from rat FOF during the risky-choice task. In line with our predictions, single FOF neurons encoded the value of the lottery even when controlling for choice. These results together showed that FOF is a critical node in the neural circuit for decision under risk.

Disclosures: X. Zhu: None. **C. Bao:** None. **J. Moller-Mara:** None. **J. Li:** None. **S. Dubroqua:** None. **J.C. Erlich:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.09

Topic: H.03. Decision Making

Support: ERC
SGSSS

Title: Spatiotemporal characterization of the neural correlates of social risk and risk-prediction error signals in humans

Authors: *R. KOSTOVA, M. G. PHILIASTIDES;
Sch. of Psychology and Neurosci., The Univ. of Glasgow, Glasgow, United Kingdom

Abstract: Differences between expected and experienced outcomes lead to prediction errors, which drive learning. Under conditions of uncertainty, organisms must build representations of variables that aid future choices, such as expected value and risk. Although decision making is heavily driven by different risk propensities, the neural correlates of risk and risk prediction errors (RiPEs) have received less attention compared to predicted rewards (PRs) and reward prediction errors (RPEs). Furthermore, the literature to date has focused primarily on understanding the processing of risk signals in non-social (i.e. probabilistic) contexts.
We used a passive-observation task (n = 40) with socially relevant individually-specific cues of rewards with fixed expected value (EV) and overall SD (predicted risk) while varying trial-wise SD (current risk). RiPEs were defined as the difference between the current risk and predicted risk. We wanted to assess the extent to which social risk and RiPEs will be reflected in the EEG signal and compare to recent EEG work with a non-social probabilistic task. Before the main experiment, each participant rated how much money (£10-110) they believed each of 30 faces (trustees) returned to a hypothetical investor. Seven of these faces were used in the main task and were paired in individual trials to form four different current risk levels (predicted risk = 26.46), and an EV of 60. On each trial, one face stimulus was highlighted to indicate the final reward outcome. A single-trial multivariate discriminant analysis was used to identify spatial EEG weightings that optimally discriminated between the two extreme levels along three distinct dimensions: current risk, signed RiPE and unsigned RiPE.
We found separate representations for the three signals, with largely distinct spatiotemporal response profiles. The current risk was discriminated from 170 ms post-stimulus, with an average peak at 272 ms, while unsigned RiPE emerged at 200 ms and peaked at 280 ms. Finally, signed RiPE peaked later around 250 ms and remained sustained between 270 and 510 ms, indicating greater temporal inter-subject variability. These onset times are consistent with an earlier risk representation, required for computing the signed and unsigned attributes of the RiPE. We, further, verified the components' dissociation by showing significant parametric effects between each component's amplitudes and the four levels of the related factors.
These results suggest that there are three separate signals coding risk and RiPEs that are likely to generalise across domains, therefore pointing towards a common currency of coding social and non-social uncertainty.

Disclosures: R. Kostova: None. **M.G. Philiastides:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.10

Topic: H.03. Decision Making

Title: Evaluating the correlation between altruism, risk-taking behavior and personality

Authors: *C. H. CHEN[1], P. H. WU[1], C. J. LIN[1,2,3], C. H. LIN[1,2];
[1]Psychology, [2]Res. Ctr. of Nonlinear Analysis and Optimization, [3]Nursing, Kaohsiung Med. Univ., Kaohsiung, Taiwan

Abstract: Altruism, risk-taking behavior, and personality trait are all critical factors in predicting decision-making behaviors in the real world. There were a few studies that mostly assessed risk-taking behavior via descriptive gamble to pinpoint the correlation issue between the three factors. However, few studies further explored the correlation between the three factors utilizing the experience gamble to measure the risk-taking behavior. In this study, we hypothesized that the descriptive vs. experience gambles may cause different outcomes. Therefore, the current study aims to investigate the association between the three factors. This study used Dictator Game (DG) as a behavioral measure for altruism, and Balloon Analogue Risk Task (BART) was administered as the experience gamble and an index of risk-taking behavior. Furthermore, the Big Five Inventory-15 in Taiwanese (BFI-15) was utilized here as the index of personality traits. We predicted that risk-taking behavior would be significantly associated with altruistic attitudes and also correlated with certain personality traits. To examine the above issue, this study recruited 128 participants (43 males and 85 females). They first completed BFI-15, then completed BART and DG in random order. The result indicated that the correlation between altruism, risk-taking behavior and personality traits was observed. Number of explosions in BART (less risk-taking), not BART SCORE, was correlated to allocations to the recipient in DG (more altruism) ($r=-.18$, $p<.05$). In contrast to the previous descriptive studies, we found a negative correlation between the number of explosions in BART and allocations in DG. And we also found a negative relationship between neuroticism and allocations to the recipient ($r=-.203$, $p<.05$), in which a clear relationship was observed between personality traits and altruism. Notably, the distribution in DG of this study is very different from that of the Western studies (Engel, 2011). This study showed that most of the participants had made an equal allocation and only a small number of participants kept all their money to themselves. The present study found less risk-taking behavior and neuroticism probably correlated with more altruism. The correlation between the three factors should be valuable to explore further. Moreover, the cultural difference between Western findings and the present study in DG is also interesting and needed to verify deeper.

Disclosures: C.H. Chen: None. **P.H. Wu:** None. **C.J. Lin:** None. **C.H. Lin:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.11

Topic: H.03. Decision Making

Title: Laterality quotient as a predictor of willingness to engage in risky behaviors

Authors: *S. S. COELHO;
The Univ. of Texas at Austin, Austin, TX

Abstract: Laterality has been continuously studied over the years, but a consensus as to its evolutionary role in the human species is seemingly far from being achieved. There is neuropsychological evidence of the correlation between brain lateralization, language processing, and the incidence of neuropsychological conditions such as schizophrenia. Cross-cultural stigmatization often assigns overly positive or negative attributes to left-handed individuals, but researchers have found conflicting results when evaluating handedness through patterns of intelligence, epigenetic expression, in utero development, and psychopathological genealogy. This study aimed to analyze the behavioral implications of lateralization and hemisphere specialization, most specifically highlighting the underlying correlations between handedness, decision-making, and neurofunction. The main goal was to understand potential correlations between cognitive development and subsequent willingness to take risks throughout life as a function of brain lateralization. Willingness to take risks was measured through the DOSPERT test, and participants' laterality quotients were calculated through the Edinburgh Handedness Inventory (EHI). Laterality quotients were crossed with behavioral patterns recorded in the DOSPERT test through statistical modeling to determine predictive factors between handedness and risk-related decision-making. We found that laterality quotients were highly correlated with willingness to take risks when there was a strong preference for one hand over the other. These findings support the hypothesis that brain lateralization and motor coordination might be functionally correlated with human behavioral patterns and decision-making abilities. Investigating the ontogenesis of laterality and its interaction with behavior can provide a further understanding of brain connectivity and function, which may facilitate the advancement of neuropsychological treatments.

Disclosures: S. S. Coelho: None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.12

Topic: H.03. Decision Making

Title: Risk Attitudes, and mood, are affected by our sleep duration

Authors: *Z.-Y. YAN[1], P. W. GLIMCHER[2];
[1]New York Univ., New York City, NY; [2]New York Univ., New York, NY

Abstract: Environmental states affect our day-to-day emotions and decisions, including our choices under risk and uncertainty. Previous work has found that an unexpected positive outcome can increase risk-taking, such as a sunny day after a streak of cloudy days. To extend

these findings to more personal daily life activities at the within-subject level, we conducted a longitudinal examination of the correlations between a variety of daily activities and risk attitudes. We defined risk attitude using a novel PCA-based approach that allowed us to search for general patterns without relying on any one specific measure of risk-taking. Using this approach, we hoped to determine how features of daily life impact risk preferences and mood. One hundred and twenty-three subjects were recruited in the US on CraigsList and the NYU subject recruitment systems. In total 115 subjects finished the experiments. We employed a mobile smartphone-based experimental platform (Datacubed Health: Linkt) to gather daily data about the lives, states, and traits of our participants. Each participant was asked to complete a time-use diary once per week over a two-month period. The following instruments were also delivered at least once every week: a risky choice task (Levy et al. 2010), a delay discounting task (Kable & Glimcher, 2007), and the self-report positive and negative affect (Kahneman et al. 2004). In the time use diary, we asked subjects to complete a timeline detailing everything they did on that day, and we categorized these activities into Sleep, Work, Home, Leisure, and Social times. In addition, we gathered 13 personality inventories: attitude toward uncertainty, temporal discounting tendency, impulsivity, and the level of psychological distresses to better understand the individual difference in the risk attitude. Our goal was to examine how the amount of time spent on a particular activity in previous weeks influenced risk attitude-related variables, in a way that did not depend on any one specific measure of risk attitude. To do that, we employed a Principal Component Analysis (PCA) to define the collinearities of a suite of risk-related measurements. We then used these aggregated risk-ontologies to perform a standard Granger-style time-series autoregression. We found that Sleep-time significantly predicts a generalized risk vector and reported negative mood in the coming week. We did not find any correlation between the other activities we examined and mood and decisions. With more sleep time, subjects have fewer negative emotions and become more risk-tolerant. Our results indicate that simple bio-behavioral variable changes can influence mood and risk preferences.

Disclosures: Z. Yan: None. **P.W. Glimcher:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Datacubed Health.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.13

Topic: H.03. Decision Making

Support: CONACYT Scholarship

Title: Decision making and early life stress: The role of autonomic reactivity

Authors: *G. VEGA-OCEGUERA, C. VEGA-MICHEL;
ITESO, Guadalajara, Mexico

Abstract: Decision making and early life stress: The role of autonomic reactivity
Author: Vega-Oceguera, G. & Vega-Michel, C.Disclosure: Vega-Oceguera, G., NONE. & Vega-Michel, C., NONE
Tema: H. Cognition. H.03 Decision making H.03.g Neural circuits of risk and ambiguityKeywords: Decision making, early life stress, stress profile, autonomic reactivity
Learning to select efficiently among available alternatives to achieve specific goals is a skill linked to quality of life from different perspectives. Although there are multiple elements involved in this process, in the study of decision making, the relevance of psychophysiological responses to stress in the selection of alternatives, risk analysis and reinforcement-based learning has been recurrently emphasized. If psychophysiological variations in the face of straining conditions in the general population appear to be relevant when understanding decision making, what about this influence in people with a history of early-life stress? Previous research has reported that the activity of autonomic nervous system and brain circuits associated with stress-adaptation changes dramatically when people have been exposed to conditions of harm or deprivation. To understand this better, our goal was to evaluate the predictive capacity that autonomic reactivity has on decision making in adolescents with early-stage stress.We worked with a sample of 30 participants between 12- and 15-years old living in a social welfare institution. In two sessions, the BART task was solved, and an assessment of autonomic reactivity took place using the stress profile procedure developed by Thompson and Thompson (2003). With the data acquired, we develop a multiple linear regression model with five input variables: heart rate, skin conductance, muscle tension, respiration rate and distal temperature, and with the covariance index of the BART task, which usually provides information on risk propensity, as the output variable.Results: The predictive model reached statistical significance, suggesting that the autonomic nervous system response generates an important effect on the resolution of a decision-making task when there is a history of early-life stress. Of the five predictor variables considered, skin conductance and heart rate contribute the most weight to the model.Conclusion: The data suggest that autonomic reactivity comes to significantly influence performance when solving decision-making activities in people with a history of early-life stress. This may have clinical implications since these data allow us to establish lines of work to improve risk detection in a population with these characteristics.

Disclosures: G. Vega-Oceguera: None. **C. Vega-Michel:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.14

Topic: H.03. Decision Making

Support: BRAIN initiative grant R01MH117040
Takeda Science Foundation Overseas Research Fellowship

Title: The role of venrolateral prefrontal cortex in the probabilistic choices

Authors: *A. FUJIMOTO[1], C. ELORETTE[1], S. H. FUJIMOTO[1], D. FOLLONI[1], N. BIENKOWSKA[1], L. FLEYSHER[2], B. E. RUSS[3,1,4], P. H. RUDEBECK[1];
[1]Neurosci., [2]BioMedical Engin. and Imaging Inst., Icahn Sch. of Med. at Mount Sinai, New York, NY; [3]Nathan Kline Inst., Orangeburg, NY; [4]Psychiatry, New York Univ. at Langone, New York, NY

Abstract: Identifying the best course of action requires exploiting the value of known options but also learning about the value of novel options to maximize rewards. Despite this, the distinct brain mechanisms that support these processes are poorly understood. Recent studies suggested that ventro-lateral prefrontal cortex (vlPFC) is critical for learning reward values (Chau et al., *Neuron*, 2015; Rudebeck et al., *Neuron*; 2017). However, it is unclear how this area represents newly learned stimulus-reward value associations as well as well-known associations. Using functional magnetic resonance imaging (fMRI) we assessed how vlPFC interacts with other areas during choices between novel or familiar options in macaque monkeys. We trained four female macaque monkeys (*Macaca mulatta*) to perform a probabilistic choice task in a 3T MRI scanner. In this task the animals chose between two visual stimuli associated with either 90%, 50% or 30% of juice reward on each trial using eye movements. The stimuli used for each block (100 trials) were either novel at the beginning of each block ('Novel' condition) or were highly familiar to the subjects ('Familiar' condition), and each monkey performed at least 2 blocks with novel and familiar stimuli per day. The imaging data obtained were analyzed using customized version of AFNI pipelines (Cox et al., *Comp Biomed Res*, 1996). All four monkeys performed at a very high level in Familiar block (> 82%). Whereas, performance in the Novel block varied across monkeys (62-76% in the latter half of a block) and all animals showed distinct learning curves. Correct performance was higher in trials in which higher value options were paired compared to the trials with lower value pairs, in both conditions, suggesting that the animals tracked the total value of available options regardless of the trial type. Whole-brain fMRI analysis showed that several areas including bilateral vlPFC, medial thalamus, and insular cortex were engaged during the task. Although the activation was observed in both conditions, vlPFC activation was stronger in the Novel condition compared to the Familiar condition, suggesting more engagement during the learning of new stimuli. Our data demonstrate that vlPFC tracks the value of choice options not only during learning but also in the choice between extensively learned options, suggesting a critical role of vlPFC in both learning and decision-making.

Disclosures: A. Fujimoto: None. **C. Elorette:** None. **S.H. Fujimoto:** None. **D. Folloni:** None. **N. Bienkowska:** None. **L. Fleysher:** None. **B.E. Russ:** None. **P.H. Rudebeck:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.15

Topic: H.03. Decision Making

Title: Focal human hippocampal lesions impact evidence accumulation underlying approach-avoidance conflict decision making

Authors: *W. LE DUC[1], C. R. BUTLER[2], G. P. ARGYROPOULOS[3], S. CHU[1], R. ITO[4], A. C. LEE[4];
[1]Psychological Clin. Sci., Univ. of Toronto Scarborough, Toronto, ON, Canada; [2]Dept. of Brain Sci., Imperial Col., London, United Kingdom; [3]Cognitive Neurosci. and Neuropsychiatry Section, UCL Great Ormond Street Inst. of Child Hlth., London, United Kingdom; [4]Psychology, Univ. of Toronto, Toronto, ON, Canada

Abstract: AApproach-avoidance conflict (AAC) refers to situations in which a goal stimulus cues concurrent possible outcomes of reward and punishment. Recent work in rodents and in humans has implicated the hippocampus (HPC) in resolving AAC, culminating in a view of this structure as a behaviour inhibitor under AAC. However, the existing human research on AAC has relied largely on neuroimaging findings. Patient studies have recruited only a single patient with focal HPC damage, patients with extensive focal lesions beyond the HPC, or individuals suffering from temporal lobe epilepsy. These findings are therefore limited in the extent to which they allow for causal conclusions about the specific involvement of the HPC in AAC. Additionally, this work has tended to focus primarily on behavioural response measures (e.g., proportion of approach/avoid decisions) that provide limited insight into the underlying psychological processes involved (e.g., evidence accumulation). In the current study, we recruited six patients (all male; age: M = 66.00, SD = 8.00) with focal HPC lesions, as well as 18 controls (six female, 12 male; age: M = 70.56, SD = 7.96) and administered an AAC task to both groups. Consistent with rodent findings on the impact of HPC inhibition, patients approached significantly more often than did controls when faced with AAC. Hierarchical drift diffusion modelling revealed that patients showed slower evidence accumulation toward avoidance decisions compared to controls, and a baseline propensity for approach decisions under AAC. These findings provide causal evidence that the human HPC is involved in AAC behaviour and offer novel insight into the associated latent decision-making processes.

Disclosures: W. Le Duc: None. **C.R. Butler:** None. **G.P. Argyropoulos:** None. **S. Chu:** None. **R. Ito:** None. **A.C. Lee:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.16

Topic: H.03. Decision Making

Support: NSF/GRFP
NIH/NINDS 2 R01NS021135

Title: Human hippocampal theta is elevated during approach-avoidance conflict in a Pacman game

Authors: *B. R. STAVELAND[1], O. KIM-MCMANUS[2], P. BRUNNER[3], J. J. LIN[4], M. HSU[5], R. T. KNIGHT[6];
[1]Helen Wills Neurosci. Inst., Univ. of California Berkeley, Berkeley, CA; [2]Dept. of Neurol., UC San Diego, San Deigo, CA; [3]Dept. of Neurosurg., Washington Univ., Saint Louis, MO; [4]Dept. of Neurol., UC Davis, Davis, CA; [5]Haas Sch. of Business, California Clin. Trials, Berkeley, CA; [6]Dept. of Psychology, UC Berkeley, Berkeley, CA

Abstract: Funding: NSF GRFP (BRS); NIH/NINDS 2 R01NS021135 (RTK)
Choosing to approach or avoid actions or stimuli which represent both rewarding and aversive outcomes is both characteristic of everyday decisions and frequently induces anxiety. In humans, excessive avoidance is a feature of generalized anxiety disorder, PTSD, and agoraphobia. Research in rodents and primates has implicated the hippocampus (HC) in approach-avoidance conflict (AAC). Further, anxiolytics reduce HC theta band activity in anxiogenic contexts. However, human fMRI studies provide conflicting results about hippocampal activation during AAC. Here we tested two presurgical epilepsy patients (n=18 HC electrodes) on a novel, continuous approach-avoidance conflict decision-making game based on the arcade game Pacman (240 trials). The decision to move towards the center of the corridor was associated with potential gains (eating "dots", resulting in points) and potential losses (ghost attack, resulting in loss of the Pacman life). 20% of trials were no-conflict trials, where the patient was free to collect dots without the threat of the ghost. We focused on theta power (3-7Hz) by condition (ghost vs no-ghost), by performing t-tests on the averaged signal between the two conditions and FDR corrected for the number of time points. We found that theta power was elevated in both patients during ghost trials compared to no-ghost trials prior to turning around (patient 1: 996-1534 milliseconds, $p < .05$, corrected; patient 2: 1036-1442 milliseconds and again briefly from 401-571 milliseconds prior to turning around ($p < .05$, corrected for both windows). These results provide evidence for human theta hippocampal involvement during approach-avoidance conflicts.

Disclosures: **B.R. Staveland:** A. Employment/Salary (full or part-time):; Helen Wills Neuroscience Institute, UC Berkeley. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NSF GRFP. **O. Kim-McManus:** A. Employment/Salary (full or part-time):; UC San Diego Department of Neurosciences, Rady Children's Hospital, Division of Neurology. **P. Brunner:** A. Employment/Salary (full or part-time):; Department of Neurology, Albany Medical College, National Center for Adaptive Neurotechnologies, Department of Neurosurgery, Washington University School of Medicine, St. Louis. **J.J. Lin:** A. Employment/Salary (full or part-time):; Department of Neurology, UC Davis, Center for Mind and Brain, UC Davis. **M. Hsu:** A. Employment/Salary (full or part-time):; Helen Wills Neuroscience Institute, UC Berkeley, Haas School of Business, UC Berkeley. **R.T. Knight:** A. Employment/Salary (full or part-time):; Helen Wills Neuroscience Institute, UC Berkeley, Department of Psychology, UC Berkeley. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report

that research relationship even if those funds come to an institution.; NIH/NINDS 2 R01NS021135.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.17

Topic: H.03. Decision Making

Support: MRC G1000183
K.Z is supported by the Institute for Neuroscience at Cambridge University, The Alan Turing Institute and Downing College, Cambridge.

Title: The road not taken: investigating the behavioural and neural correlates of changing one's mind

Authors: *K. ZUHLSDORFF[1], J. DALLEY[2], T. ROBBINS[3], S. MOREIN-ZAMIR[4]; [1]Dept. of Psychology, [2]Univ. Of Cambridge, Cambridge, United Kingdom; [3]Dept. of Psychology, Univ. of Cambridge, Cambridge, United Kingdom; [4]Sch. of Psychology and Sport Sci., Anglia Ruskin Univ., Cambridge, United Kingdom

Abstract: Cognitive flexibility is the ability to adapt to our constantly changing environment. Reduced flexibility can lead to worse life outcomes and heightened cognitive decline with age. However, a full picture of the behavioural and neural underpinnings of cognitive flexibility has not yet been established, as it is a multifaceted construct. Paradigms commonly used to study flexibility include task-switching, attentional set shifting and reversal learning. In daily life, cognitive flexibility can involve a shift in behaviour being elicited volitionally. Individuals often choose to change behaviours based on environmental signals, but these are frequently accompanied by noise from the surroundings. We report results from a novel 'change your mind' task, which assesses proactive switching under uncertainty, without the need for ongoing rule-based learning. Participants completed a two-alternative forced choice task and following spurious feedback, were presented with the same stimulus again. They could repeat their previous response or change it, acting of their own volition. To our knowledge, no existing task provides participants with the opportunity to repeat their choice and assess whether they subsequently choose the 'road not taken'. We report findings from forty healthy participants who completed the task whilst undergoing functional MRI (fMRI) imaging. Behaviourally, participants predominantly repeat their choice but change their response choice when the first response was incorrect, or when negative feedback was presented. Greater activations were evident in the anterior insula (AI), anterior cingulate cortex and dorsolateral prefrontal cortex on change trials compared with repeat trials. These findings indicate that this task can be used to study volitional switching, and that the neural circuits have overlapping and diverging features to other subtypes of flexibility.

Disclosures: **K. Zuhlsdorff:** None. **J. Dalley:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Boehringer-Ingelheim GmbH, GlaxoSmithKline. **T. Robbins:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; GlaxoSmithKline, Shinogi. F. Consulting Fees (e.g., advisory boards); Cambridge Cognition, Greenfield Bioventures, Cassava, Arcadia. **S. Morein-Zamir:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.18

Topic: H.03. Decision Making

Support: NINDS 2 R01 NS021135

Title: Behavioral and electrophysiological evidence of moment-to-moment changes in expectations

Authors: *D. MARCIANO, L. BELLIER, I. MAYER, M. GOOD, M. HSU, R. T. KNIGHT; Univ. of California, Berkeley, Berkeley, CA

Abstract: Expectations are often dynamic: any sports fan knows that expectations change rapidly as games unfold. Yet expectations have traditionally been studied as static. Here we provide parallel electrophysiological and behavioral evidence that expectations change from moment to moment. In Study 1 (EEG, N=37, 150 trials each) subjects played a realistic slot-machine game. They chose one item on the left reel, the right reel spun and decelerated to a stop. If the items on the payline *matched*, subjects *lost* $0.25; otherwise, they won $0.10. We classified wins by whether the machine stopped 1 item before a match (Narrow Escape Before, NEB), 1 item after (NEA) or more (Full Escape, FE). The feedback related negativity (FRN) was enhanced for losses (Fig.1.a $p<.001$). The P3 was larger for NEB than for other wins ($p<.001$), suggesting a bigger reward prediction error. Prior to the slot machine reveal, EEG differed for NEB vs. NEA and FE in the [-500ms-0] window ($p<0.01$), reflecting the expectations elicited by the possibility of losing (NEB) vs. assured winning as the wheel spins past match (NEA/FE). Study 2 (online, N=30, 36 trials each) used a new task to behaviorally measure expectations. On each trial, subjects chose between betting on a slot machine or a sure amount. Subjects could change their choice as often as they wanted during the trial. Bonuses depended on their choice at a random timepoint, thus incentivizing subjects to report their true expectations at each moment. We averaged choices across trials and subjects for each timepoint to build expectation trajectories for the 4 outcomes assessed in the EEG study. Notably, these trajectories were strikingly similar to the EEG traces in the last second before the machine stopped (Fig.1, c d). A

timeseries regression analysis confirmed that behavioral expectations predicted EEG activity at Cz ($p<.001$). These studies provide the first evidence that moment-to-moment expectations can be behaviorally measured and are tracked by EEG activity. Our finding of dynamic expectations provides a new dimension to our understanding of how we predict the future.

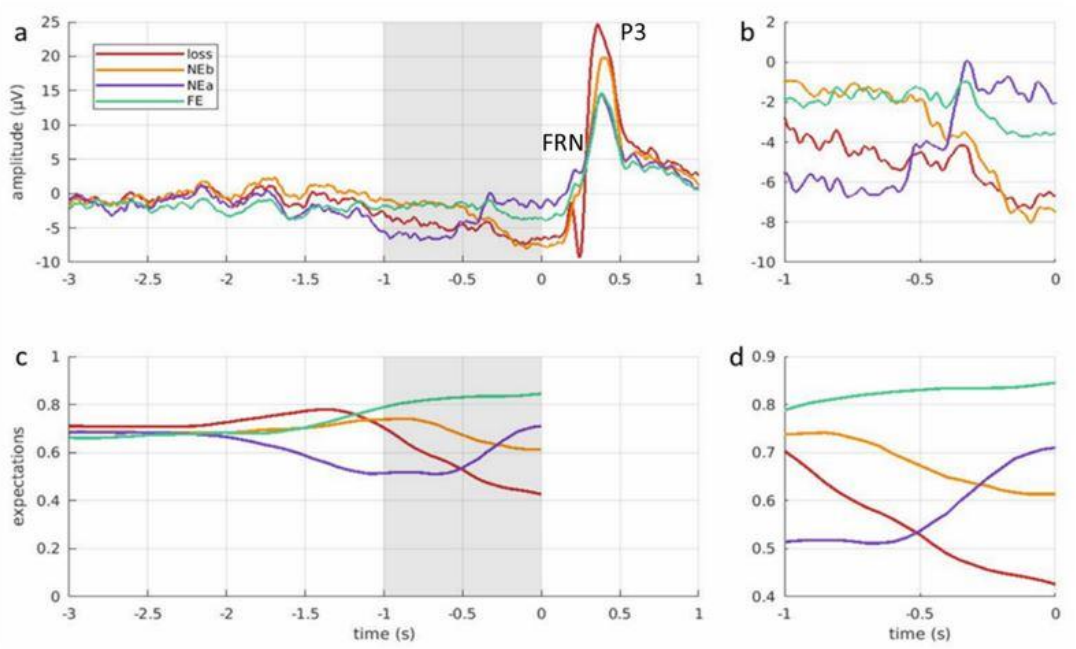


Fig. 1. a) Study 1 grand-average ERPs at Cz. t=0: machine stops spinning. On average, deceleration started 3.3 second before the machine stopped. Gray shaded area (last second of deceleration) is blown up in panel b. c) Study 2's expectations trajectories. Gray shaded area is blown up in panel d. All outcomes have similar expectations trajectories up to 2 sec before the machine stops. Note how Panels c) and d) show similar time-courses.

Disclosures: D. Marciano: None. **L. Bellier:** None. **I. Mayer:** None. **M. Good:** None. **M. Hsu:** None. **R.T. Knight:** None.

Poster

567. Neural Mechanisms of Decision Making: Risk and Ambiguity

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 567.19

Topic: H.03. Decision Making

Support: NIH R21AG049293
NIH R56AG058769

NSF BCS-1829439
Gruber Foundation Fellowship

Title: Qualitative Decision-Making Under Uncertainty

Authors: *A. RICH[1], R. JIA[1], T. FRIED[2], I. LEVY[1];
[1]Dept. of Comparative Med., [2]Dept. of Intrnl. Med., Yale Sch. of Med., New Haven, CT

Abstract: Behavioral and neuroimaging studies of decision-making under uncertainty typically use monetary outcomes. Though convenient for experimental paradigms, many decisions involve non-monetary outcomes. Life choices that are not directly quantifiable remain largely unstudied in part due to the large challenge that they pose to models of valuation and choice. Qualitative decision-making may exhibit significant demographic, behavioral, and neural variability that is not captured by quantitative outcomes. To investigate this, we presented healthy young (n=34, mean age 25.7±4.5, 18 female) and older adults (n=15, mean age 71.3±5.5, 6 female) with a monetary lottery task and a novel verbally-described medical decision-making paradigm while they underwent fMRI. Participants made monetary and medical choices under two conditions of uncertainty-risk and ambiguity. To assess the influence of uncertainty on choice behavior, the proportion of trials in which participants chose the uncertain option was calculated at each uncertainty level in both the monetary and medical domains. We found that, in both age groups and domains, participants chose the risky option more as outcome probability increased and the ambiguous option less as the level of ambiguity increased, though older adults were more ambiguity averse in the monetary domain than young adults (t(29)=-2.04, p=.050). Additionally, among older adults, men exhibited greater ambiguity aversion in the monetary domain than women (t(12) =-2.48, p=.03). To investigate the behavioral consistency of qualitative and quantitative decision-making, we correlated choice probabilities within subjects across domains. For young adults, medical and monetary choice probabilities were significantly correlated with one another under both risk (r=.56, p<.001) and ambiguity (r=.65, p<.001); however, in older adults, choice probabilities within the medical and monetary domains were statistically similar under risk (r=.54, p=.04) but not ambiguity (r=.19, p=.50). To explore the neural underpinnings of qualitative choice, we plan to conduct a univariate analysis by general linear model using binary predictors representing each uncertainty level in both decision domains. Additionally, we plan to conduct second-level region-of-interest analyses in the right posterior parietal cortex, previously shown to relate to risk attitudes. At present, our results demonstrate the importance of ecologically relevant decision-making paradigms and suggest that the synchrony between qualitative and quantitative decision-making may differ as a function of demographic variables such as age and gender.

Disclosures: A. Rich: None. **R. Jia:** None. **T. Fried:** None. **I. Levy:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.01

Topic: H.05. Working Memory

Support: NIH Grant R01-027925
NYU MacCracken Fellowship

Title: Effort impacts neural representations of spatial working memory

Authors: *S. L. MASTER, C. E. CURTIS;
New York Univ., New York Univ., New York, NY

Abstract: Mental effort is defined as the amount of cognitive work required by a given task. However, when subjects are properly incentivized, they can apply mental effort to improve their performance, independent of task difficulty. That people often need to be incentivized to exert cognitive effort suggests that it is costly to use. It remains unclear why cognitive effort is costly to use, given that its use is beneficial and adaptive. Here, we aim to elucidate the neural mechanisms of cognitive effort exertion and the downstream consequences of effort in terms of the quality of neural representations. Using a spatial working memory (WM) task, eye-tracking, fMRI, and Bayesian decoding, we test the hypothesis that cognitive effort impacts the quality of WM representations stored in the human brain. Participants performed a visual-spatial WM task with two trial types which required more (hard) or less (easy) memory precision. The amount of precision on each trial type was determined within-subject through two separate staircase procedures. Importantly, during the WM delay period, the only difference between trial types was the participant's expectation of the difficulty of the upcoming judgment. We found that on hard compared to easy trials, participants were slower to respond and had larger pupil sizes during the memory delay, suggesting that subjects exerted more cognitive effort on those trials. Delay period BOLD activation was greater on hard relative to easy trials in visual field maps throughout the dorsal visual stream, suggesting that the exertion of cognitive effort increases neural response gain. These changes in gain across trial types were particularly strong in frontal cortex (retinotopic areas inferior and superior precentral sulcus). In addition, even voxels with receptive fields not containing the WM stimulus exhibited increased activity on difficult trials. These changes in BOLD activation were accompanied by increases in WM representational quality. Using a Bayesian decoder, we found evidence for increased representational accuracy in some retinotopic ROIs, like visual area V3AB. These results demonstrate that the neural mechanisms of subjective constructs like cognitive effort can be studied effectively. They also suggest one mechanism through which the use of cognitive effort may be costly, which is that its exertion is accompanied by widespread increases in neural activation even in task-irrelevant cortical areas.

Disclosures: S.L. Master: None. **C.E. Curtis:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.02

Topic: H.05. Working Memory

Support: NIH Grant R01 EY-016407
NIH Grant R01 EY-027925
Swartz Foundation Postdoctoral Fellowship

Title: Drift-like dynamics of working memory representations in the human brain

Authors: H.-H. LI, W. MA, ***C. E. CURTIS**;
Dept. of Psychology, NYU, New York, NY

Abstract: Working memory (WM) allows the brain to store information for a brief period of time and thereby increases the duration of neural representations available to support decision-making and actions. The neural processes underlying WM are noisy and memory error increases with delay duration. Even though neurophysiological studies on animals and theoretical works have investigated how WM deteriorates over time, the formation of memory errors in the human brain remains largely unexplored. Here with fMRI, we studied how memory errors evolve across time in a classical memory-guided saccade task. During the experiment, in each trial, participants were presented with a brief WM target positioned at a random polar angle with a fixed eccentricity (12° from the fixation point). Participants were required to remember the location of the target, and after a 12-second delay, report their memory by making a saccadic eye movement. We used a Bayesian decoder (TAFKAP; van Bergen and Jehee, 2021) to decode the location (polar angle) of the target using the voxel activity pattern measured at each single time point during the delay. We found that the stimulus information, quantified as the (circular) correlation between the target location and the decoded location, can be decoded as early as 1.5 seconds from delay onset and peaks at about 3.75 seconds. Moreover, the errors made by the decoder were predictive of behavioral memory. The correlation between decoding error and behavioral error became significant at a later time point (~7.5 seconds at IPS0) and ramped-up over time during the delay. This error-error correlation emerged earliest in IPS0 and the dorsal exstrastriate cortex V3AB, and only became significant at the end of the delay in primary visual cortex. By sorting the trials based on the direction (clockwise vs. counterclockwise) of memory error, we found that the decoding error increased over time in the direction of behavioral memory error. Overall, our results are consistent with theories that model memory error as drift-like dynamics. These unique temporal population dynamics suggest that memory error accumulates in IPS and high-level visual cortex, and that WM content in primary visual cortex reflects feedback signals.

Disclosures: H. Li: None. **W. Ma:** None. **C.E. Curtis:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.03

Topic: H.05. Working Memory

Title: Linking behavioral and neural estimates of trial-by-trial working memory information content

Authors: *Y. ZHOU, D. FOUGNIE, K. K. SREENIVASAN;
Div. of Sci. and Mathematics, New York Univ. Abu Dhabi, Abu Dhabi, United Arab Emirates

Abstract: How is working memory (WM) information represented in the brain? Neural and computational models have used data aggregated over hundreds of trials to argue for different perspectives on how population neural activity encodes individual memories. These proposals range from the notion that memories are point estimates (e.g., a specific shade of red) or that memories include measures of uncertainty (e.g., a reddish color), or that memories are complex probability distributions over feature space. The use of aggregate data represents a key inferential bottleneck that critically limits the ability to adjudicate between different theories of information coding in WM. In this study, we overcome this limitation using a powerful method to link behavioral and neural estimates of WM representation on a trial-by-trial basis. We asked 7 human subjects to memorize the direction of a dot-motion stimulus and hold this information in WM over a brief delay. At the end of the delay, instead of making a single report about the memorized motion direction, they indicated their memory for motion direction by placing 6 "bets", resulting in a distribution over 360° direction space that reflected subjects' probabilistic memory distribution on a per-trial basis. Additionally, we used a recently-developed probabilistic decoding method (TAFKAP; van Bergen & Jehee, 2021) to estimate the posterior probability distribution of motion direction given the BOLD fMRI signal in V1-V3. While this approach yields a probability distribution on a per-trial-basis, in practice it has been used to generate only aggregate mean and variance measures, due to difficulty linking trial-specific neural evidence with behavior. We compared trial-wise behavioral and neural distributions using Kolmogorov-Smirnov (KS) tests on each trial and found that the KS value was significantly smaller than chance (permutation test), suggesting that the variation in probabilistic neural representations matches that of behavioral reports. Moreover, the asymmetries in behavioral and neural distributions matched; the KS value for the real data was significantly smaller than the KS value for surrogate distributions with flipped asymmetry, suggesting that the asymmetry in neural representations was reflected in behavioral responses. These results indicate (1) that individual WM representations are complex probability distributions that contain more information than that can be deduced from aggregate WM data, and (2) that asymmetries in neural probability distributions influence behavior.

Disclosures: Y. Zhou: None. **D. Fougnie:** None. **K.K. Sreenivasan:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.04

Topic: H.05. Working Memory

Support: National Natural Science Foundation of China 31930052
Beijing Municipal Science & Technology Commission Z18110000151800

Title: Neural representation of hierarchical rank information in auditory working memory

Authors: *Y. FAN[1], N. DING[2], H. LUO[1];
[1]Peking Univ., Peking Univ., Beijing, China; [2]Zhejiang Univ., Zhejiang Univ., Hangzhou, China

Abstract: Natural auditory sequences always organize into hierarchical structures. For example, when listening to speech, the brain has to encode the order of syllables to recognize a word and encode the order of words to understand a sentence. How the brain maintains such ordinal information of hierarchical sequences in working memory (WM) remains elusive. Our previous works have shown that the ordinal rank information could selectively be reactivated during the delay period of a WM task, by both a post-cue and a task-irrelevant neutral impulse. In the present study, using the same approach, we aimed to probe the neural representation of the hierarchical rank information retained in WM. We recorded electroencephalography (EEG) activities on human subjects while they memorized a sequence of words and report both global order of words and local order of syllables within a word. By employing a time-resolved multivariate decoding approach, we demonstrate that during the delay period, an auditorily presented probe word reactivates both its global and local rank information. A neutral impulse, however, only reactivates global but not local rank information, suggesting a predominance role of global structure in WM organization. Furthermore, global and local ranks are encoded by distinct response patterns, implying potentially orthogonal representations. Finally, rank reactivations correlate with behavioral global precedence effect. Taken together, the global and local ranks of a hierarchical auditory sequence are separately represented in the WM system, wherein the global rank might serve as a primary scaffold to organize information storage.

Disclosures: Y. Fan: None. **N. Ding:** None. **H. Luo:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.05

Topic: H.05. Working Memory

Support: DFG SFB 1089
SPP2205
MO 930/4-2
Volkswagen Foundation: 86 507
BMBF 031L0197B, Tübingen AI Center
FKZ: 01IS18039A

Title: Theta-based spike-phase coding supports temporal-order working memory in the human MTL and recurrent neural networks

Authors: *S. LIEBE[1], J. NIEDIEK[3], M. PALS[2], T. P. REBER[4], J. MACKE[2], F. MORMANN[5];
[1]Univ. of Tuebingen, Tübingen, Germany; [2]Univ. of Tuebingen, Tuebingen, Germany; [3]Edmond and Lily Safra Ctr. for Brain Sci., The Hebrew Univ. of Jerusalem, Jerusalem, Israel; [4]Psychology, UniDistance Suisse, Brig, Switzerland; [5]Univ. Clin. of Bonn, Bonn, Germany

Abstract: Memories are composed of discrete events across time. However, which neural mechanisms underlie remembering the temporal order of events remains an open question. In this study, we probed a prominent theory proposing that temporal order of items in working memory is reflected in sequential neural activity at different phases of theta oscillations. We simultaneously recorded spiking activity and Local Field Potential (LFP) in the medial temporal lobe of neurosurgical patients performing a multi-item working memory task. We additionally trained Recurrent Neural Network Models (RNNs) in an analogous paradigm. During the memory maintenance period, we observed enhanced oscillatory theta power (2-8 Hz) as well as theta-phase-related spiking that reflected stimulus position and, importantly, depended on memory performance. Similarly, theta oscillations emerged in RNNs after training, and model units showed similar phase-dependent activity related to item position as recorded neurons. However, in contrast to the theory, the ordering of preferred phase of firing did not reflect the serial order of encoded memory items, both for recorded and model units. Instead, our modeling linked temporal order to stimulus timing and oscillation frequency, which we subsequently confirmed in our neural recordings. Taken together, our findings implicate spike-phase coding in the theta range as a potential mechanism to maintain sequential information within biological and artificial neural networks.

Disclosures: S. Liebe: None. **J. Niediek:** None. **M. Pals:** None. **T.P. Reber:** None. **J. Macke:** None. **F. Mormann:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.06

Topic: H.05. Working Memory

Title: Increasing Delay Period in Working Memory Tasks Leads to More Stable Information Encoding

Authors: *A. SIMONOFF, M. C. ROSEN, D. J. FREEDMAN;
Univ. of Chicago, Univ. of Chicago, Chicago, IL

Abstract: Working memory, which encompasses the ability to maintain and manipulate information in short-term memory, requires a complex interplay between several brain areas ranging from sensorimotor to executive function. The primary neural correlate of WM is persistent neural activity while animals hold information in memory in the lateral interparietal cortex (LIP), prefrontal cortex (PFC), and the dorsolateral PFC (dlPFC). The function of this

persistent activity and the mechanisms through which it occurs remain key areas of inquiry. Here, as part of an investigation of the mechanism of persistent activity, we conducted a theoretical investigation using recurrent neural networks (RNNs) to understand how persistent activity may occur and what it reflects, and we trained two monkeys to prepare for task performance and recordings. Average network task accuracies was greater than 97%, but decoding accuracy varied from 12% to 99% using support-vector machines (SVMs). The networks robustly demonstrated that information encoding occurs in short-term synaptic plasticity (STSP), especially when the delay duration is significantly shorter than the time constant of STSP, as decoding accuracy decreases in those circumstances. This was also demonstrated in the stark changes in the principal component analyses (PCAs) as delay duration increases: the first two principal components change from being overlapping to becoming spatially distinct as soon as the delay duration is larger than the neurotransmitter release duration. Finally, this is confirmed by the cross-temporal analyses, where this behavior is also observed during decoding of the sample during the delay period. As such, we conclude that information must be maintained in silent processing, i.e. STSP, during those times that information is unable to be decoded from the persistent activity of the networks but the networks still demonstrate high accuracy. We will investigate this further *in vivo* using semi-chronic electrode array recordings from surface cortical areas (such as the dorsolateral prefrontal cortex, dlPFC) and acute recording from sulcal areas (such as the lateral interparietal cortex, LIP).

Disclosures: A. Simonoff: None. **M.C. Rosen:** None. **D.J. Freedman:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.07

Topic: H.05. Working Memory

Support: ESF-Sachsen Anhalt

Title: Trpc4 channel in the hippocampal ca1 area supports working memory, persistent firing and theta-gamma coupling

Authors: *B. SABER MAROUF[1], A. REBOREDA[2], F. THEISSEN[4], R. KAUSHIK[3], A. DITYATEV[5], M. SAUVAGE[6], M. YOSHIDA[7];
[1]Leibniz-Institut für Neurobiologie, Magdeburg, Germany; [2]Functional Architecture of Memory, [3]Leibniz Inst. For Neurobio., Magdeburg, Germany; [4]LIN, Magdeburg, Germany; [5]Mol. Neuroplasticity, DZNE: German Ctr. for Neurodegenerative Dis., Magdeburg, Germany; [6]Functional Architecture of Memory Dpt, Leibniz Inst. for Neurobio., Magdeburg, Germany; [7]LIN Magdeburg, Magdeburg, Germany

Abstract: Working memory requires a maintenance of information in a readily accessible form in various cognitive tasks. Persistent firing of neurons is believed to be one of the

neurophysiological mechanisms to retain information during working memory. However, it still remains unclear whether persistent firing is supported by intrinsic properties of individual neurons or by the synaptic network among them. Based on our previous work indicating the role of transient receptor potential canonical 4 (TRPC4) channels in persistent firing in individual hippocampal CA1 pyramidal cells, here we studied the role of TRPC4 channels on the spatial working memory and electrophysiological activity in vivo. A spatial-delayed alternation task (DNMTP-T-maze) was used to study the effect of TRPC4 knockdown on working memory in mice implanted with tetrodes in the CA1 area (12 weeks old, C57BL/6J). The behavioral results showed a significant performance attenuation in the TRPC4 knockdown group compared to the control group that was injected with a scramble shRNA virus. Local field potential analysis has shown significantly lower theta-high gamma coupling in the TRPC4 KD group compared to the control group. Single unit data analysis has depicted a significant decrease in elevated firing rate in the delay period of the working memory task (persistent firing). In addition, phase precession was reduced in TRPC4 KD group. In conclusion, our data suggests that hippocampal TRPC4 channels support spatial working memory through persistent firing, theta-high gamma nesting and phase precession.

Disclosures: **B. Saber Marouf:** None. **A. Reboreda:** None. **F. Theissen:** None. **R. Kaushik:** None. **A. Dityatev:** None. **M. Sauvage:** None. **M. Yoshida:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.08

Topic: H.05. Working Memory

Support: VA RR&D Grant RX000825

Title: Removing items from working memory engages large-scale brain networks and is sensitive to dopamine tone

Authors: A. J. WESTPHAL[1], *A. S. KAYSER[2];
[1]Univ. of California, San Francisco, Univ. of California, San Francisco, San Francisco, CA;
[2]Univ. of California San Francisco, Univ. of California San Francisco, San Francisco, CA

Abstract: Control over the contents of working memory is critical to ongoing cognition. While much attention has focused on the encoding and manipulation of such memoranda, the ability to actively remove items from this limited capacity system is also crucial. Currently it is unknown whether removal has the same dependencies as other working memory processes on dopaminergic tone, and whether it requires both local and large-scale changes in the connectivity of frontoparietal brain regions. In this study, subjects (N = 30) received either placebo or the catechol-O-methyltransferase (COMT) inhibitor tolcapone in randomized, counterbalanced, double-blind fashion while performing a working memory task in which they encoded three

faces on each trial. Concomitantly with the third face, a retrospective cue (retrocue) alerted subjects to remember either one (low load condition) or all three (high load condition) of the faces. Behaviorally, performance assessed by the sensitivity index, d', was significantly greater in the low-load than the high-load condition ($F(1, 30) = 119.642$, $p < 0.0001$, adj. $R^2 = 0.7928$), indicating that subjects successfully utilized the retrocue. Consistent with the active nature of removal processes, corresponding BOLD responses in the frontoparietal and fusiform regions were significantly larger in the low-load than the high-load condition at cue, and activity in a face-sensitive region, the left fusiform cortex, scaled with d' (all BOLD comparisons significant at $p < 0.05$, corrected for multiple comparisons). Concomitantly, a measure of network integration (node degree) was significantly greater in the frontoparietal network at cue for the low-load compared to the high-load condition. Although tolcapone did not significantly affect d', activity in the right caudate and putamen, as well as bilateral anterior cingulate cortex, demonstrated a significant interaction between load and tolcapone. Together these results emphasize the active contributions of frontoparietal and higher visual areas to the removal process, and they suggest a dopaminergic influence on associated neural mechanisms.

Disclosures: A.J. Westphal: None. **A.S. Kayser:** F. Consulting Fees (e.g., advisory boards); Boehringer-Ingelheim.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.09

Topic: H.05. Working Memory

Support: NIH Grant MH064498

Title: Priority-based representational transformation in RNN simulation of the double serial retrocuing working memory task

Authors: *Q. WAN[1], B. R. POSTLE[2];
[1]Dept. of Psychology, [2]Departments of Psychology and Psychiatry, Univ. of Wisconsin-Madison, Madison, WI

Abstract: What is the fate of information that is held "in" working memory (WM), but out of the focus of attention? The double serial retrocuing (DSR) WM task begins with the presentation of two samples, followed by a retrocue (cue1) designating one the "prioritized memory item" (PMI) that will be tested at recall1. The uncued item (unprioritized memory item; UMI) can't be forgotten, however, because with $p = .5$ cue2 might designate it for recall2. Previous fMRI and EEG studies of DSR have shown that multivariate evidence for the UMI can drop to baseline levels, suggesting that the UMI might be retained in an activity-silent format. More recently, evidence has also emerged that the UMI might be retained in an active, but transformed, representational format. For example, in an EEG study of the 2-back WM task, stimulus

reconstructions with multivariate inverted encoding modeling (IEM) underwent a “flip” when items took on UMI status. To explore this, we trained recurrent neural networks (RNNs) with an LSTM architecture to perform the 2-back task and applied Principal Component Analysis (PCA) and demixed (d)PCA to hidden layer activity and observed that stimulus representation also flips as a function of its priority status. Here, we assessed the generalization of this finding to a task that, unlike the 2-back, challenges performance with overt, unpredictable prioritization cues. We trained RNNs (N = 10) with an LSTM architecture to perform DSR with > 95% accuracy on an independent test set. PCA visualization of LSTM hidden layer activity revealed priority-based representational transformation: the amount of representational change in the PC space is 10.68 times (SD = 10.02) greater on “switch” trials, in which the item designated a UMI by cue1 is prioritized by cue2, than on “stay” trials in which the same item retains PMI status across the trial. Next, we applied dPCA to hidden layer activity to obtain state-specific subspaces. PMI and UMI subspaces were separated by an angle of 86.76° (SD = 2.15°), a geometry that would minimize interference between the two. PMI and decision subspaces were separated by an angle of 83.26° (SD = 3.19°), UMI and decision by 83.23° (SD = 3.71°). Although there are many differences between RNNs and the human brain, these results demonstrate the plausibility of the idea that the representational transformation of stimulus information may be a core computation supporting the flexible control of information held in WM.

Disclosures: **Q. Wan:** None. **B.R. Postle:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.10

Topic: H.05. Working Memory

Support: NIH Grant MH064498

Title: Neural mechanisms of active removal from working memory

Authors: *J. SHAN, B. R. POSTLE;
Dept. of Psychology, Univ. of Wisconsin-Madison, Madison, WI

Abstract: The ability to remove no-longer-useful information from working memory (WM) is important for the flexible control of behavior. In addition to the passive decay that results from the withdrawal of attention, behavioral data from an “ABC-retrocuing” task also provide evidence for an active removal process (Shan and Postle, 2022). ABC-retrocuing trials begin with the presentation of two oriented grating stimuli (“A” and “B”) followed by a cue indicating which of the two might be tested at the end of the trial (for this example, A). On trials that encourage “passive dropping” of the uncued item, item C then appears at a previously unoccupied location. On trials that encourage active removal of the uncued item, item C appears at the location where B had also appeared. In the present study we used functional magnetic

resonance imaging (fMRI) of healthy adult humans (male and female) performing ABC retrocuing to assess evidence for each of three proposed mechanisms: hijacked adaptation, context breaking, and mental-context shifting. The hijacked-adaptation model posits an adaptation-like modification of perceptual circuits combined with a weak activation of the to-be-removed item. Evaluated with multivariate inverted encoding modeling (IEM), it predicts a cue-triggered flipping of the IEM reconstruction of stimulus information (Lorenc, Vandenbroucke et al., 2020; Sahan, Sheldon, et al., 2020). The context-breaking model posits a breaking of the stimulus-to-context binding that corresponds to "holding" information in WM. It predicts a cue-triggered rapid decline to baseline of the IEM reconstruction of the actively removed item. The mental-context shifting model posits that interference from to-be-removed information is minimized via an abrupt change of mental context (such that the context associated with new information is maximally dissimilar to that associated with the to-be-removed information). It will be tested by using representational similarity analysis (RSA) to assess the rate of contextual shift under the active vs. the passive condition. This study is preregistered (osf.io/unrz9/) as a Stage 1-accepted Preregistered Research Report, with a target N=30. Preliminary results (N=5) are more consistent with the hijacked-adaptation than the context-breaking model. In early visual cortex, on active-removal trials, IEM reconstruction of the location of the to-be-removed item flips (to a significantly negative reconstruction) in response to the removal cue, before later returning to baseline in time for the encoding of new information at that location.

Disclosures: J. Shan: None. **B.R. Postle:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.11

Topic: H.05. Working Memory

Support: NIAAA Grant T32-AA007468
OHSU Physician Scientist Grant

Title: Differential medial prefrontal cortex subregion and layer involvement during distinct phases of working memory

Authors: *A. SONNEBORN[1], L. BARTLETT[2], A. I. ABBAS[3];
[1]Behavioral Neurosci., Oregon Hlth. & Sci. Univ., Portland, OR; [2]Behavioral Neurosci., OHSU, Portland, OR; [3]Dept. of Behavioral Neurosci., Vaporhcs/Oregon Hlth. & Sci. Univ., Portland, OR

Abstract: The rodent medial prefrontal cortex (mPFC) can be divided into several layers and subregions based on both anatomical location and input/output connectivity. This region as a whole is thought to be essential for controlling working memory (WM), an active process in which an animal can hold recent sensorimotor information in mind with the goal of manipulating

it or protecting it from interference for future use. Activity within isolated layers or subregions has been differentially implicated in mediating distinct phases of WM (e.g. information encoding, maintenance, or retrieval). However, how all layers and subregions interact in real time as an animal performs a WM task is completely unknown. Here, we simultaneously recorded from deep and superficial layers down the entire dorsal/ventral axis of the mPFC as mice complete a non-match to place WM task. Our initial findings indicate that spatial information enters the mPFC (encoding) in a distributed manner across all layers and subregions. It then undergoes a qualitative change so that it can be maintained in the superficial layers of dorsal mPFC during a delay period, and is subsequently transferred again to a more distributed representation before the animal needs to make a decision. To our knowledge, these findings are the first to detail the dynamic information flow between all mPFC layers and subregions within the same experiment.

Disclosures: A. Sonneborn: None. **L. Bartlett:** None. **A.I. Abbas:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.12

Topic: H.05. Working Memory

Support: Simons Collaboration for the Global Brain Pilot Award 872599SPI
NIH BRAIN Initiative R00 MH120047

Title: Intrinsic timescale gradients across the dorsal cortex vary by cortical layer and are modulated by behavioral state

Authors: *P. SALVINO, L. PINTO;
Dept. of Neurosci., Northwestern Univ., Chicago, IL

Abstract: The mammalian cortex appears to be functionally organized along a hierarchy of intrinsic timescales, in which posterior sensory areas integrate input over shorter time windows than frontal association areas. These timescales are affected in aging and several neuropsychiatric diseases, and have been implicated in high-level cognitive processes such as working memory and evidence accumulation. However, the mechanisms underlying the generation of these timescales within local circuits and the degree to which they are modulated by behavioral state remain unclear. Here, we used mesoscale widefield calcium imaging of the whole dorsal cortex to begin investigating the circuit mechanisms underlying intrinsic timescales. Using transgenic mouse lines that preferentially label subpopulations of excitatory neurons, we examined how these timescales manifested within different cortical layers. Specifically, we imaged GCaMP6s from mouse lines that preferentially label layer II/III, layer V, or layer VI neurons in mice running spontaneously. We observed timescale gradients in all layers. Interestingly, however, the timescales themselves decreased systematically with cortical

depth, with layer II/III displaying the longest timescales across all cortical areas. Further, these timescales were modulated by the behavioral state of the mice as indexed by running. This was true in all layers but modulation was strongest in layer V. We are currently investigating how timescales within and across layers and areas are expressed in individual neurons using cellular resolution two-photon microscopy. Taken together, our findings indicate that intrinsic timescales across the cortex are dynamic. Further, the laminar differences place important constraints on models of how intrinsic timescales arise in cortical circuits, and on our understanding of how cortical circuits are functionally organized to support behavior.

Disclosures: P. Salvino: None. **L. Pinto:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.13

Topic: H.05. Working Memory

Support: National Science and Technology Innovation 2030 Major Program 2021ZD0204103
National Natural Science Foundation of China (31930052)
China Postdoctoral Science Foundation (2020M680166)

Title: Co-occurrence of past and present shifts current neural representations and mediates serial biases

Authors: *H. ZHANG, H. LUO;
Sch. of Psychological and Cognitive Sciences, IDG/McGovern Inst. for Brain Sci., Peking Univ., Beijing, China

Abstract: The regularities of the world render an intricate interplay between past and present. Even across independent trials, current-trial perception can be automatically shifted by preceding trials, namely the ‘serial bias’. Meanwhile, the neural implementation of the spontaneous shift of present by past that operates on multiple features remains unknown. In two auditory pitch categorization experiments with human electrophysiology recordings, we directly examined how various past-trial features (pitch, category, and motor response) influence the present perceptual decision-making. Specifically, human subjects were required to report whether a given pitch belonged to the learned “High” or “Low” categories by pressing two buttons, wherein the categorical report and the motor response were dissociated by a choice-response cue presented either after (Experiment 1) or before (Experiment 2) the tone. First, within the same task, decision-making exhibits ubiquitous serial biases — the present category report being repelled away from the previous pitch but biased towards the previous categorical report, and the motor response showing a bias of alternation. Using multivariate decoding, we next demonstrate that serial bias arises from the co-occurrence of past-trial neural reactivation and the neural encoding

of current-trial features. Notably, the past-trial feature can only be reactivated by the corresponding present-trial feature, resulting in a synchronized temporal profile of neural representation over time as revealed by the cross-correlation analysis. Our novel representation-shift analysis further reveal that the meeting of past and present shifts the neural representation of current-trial features towards the direction consistent with the feature-specific behavioral biases and modulates serial bias behavior. In addition, the neural shift of features occurs at different temporal latencies, relatively early for pitch and motor response, but later for category, suggesting that serial bias operates at different stages of decision-making, from sensory processing to choice formation and motor execution. In summary, past-trial features that constitute an 'event-file' keep their respective identities in working memory and are only reactivated by the corresponding features in the current trial, giving rise to dissociated feature-specific serial biases in neural representation and behaviors. This 'event-file' reactivation might constitute a fundamental mechanism for adaptive past-to-present generalizations over multiple features and facilitate the automatic interaction between memory and decision-making.

Disclosures: H. Zhang: None. **H. Luo:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.14

Topic: H.05. Working Memory

Support: Swiss National Science Foundation (SNSF)

Title: Does the future use matter? Memorized information and representations in memory

Authors: *P. FIAVE[1], K. JORDAN[1], K. UITTENHOVE[2], L. BARTSCH[3], E. VERGAUWE[1]; [1]Fac. of Psychology and Educational Sci., Univ. of Geneva, Geneva, Switzerland; [2]Inst. of Psychology, Univ. of Lausanne, Lausanne, Switzerland; [3]Dpartment of Psychology, Univ. of Zurich, Zurich, Switzerland

Abstract: A key element of human cognition is working memory (WM), a limited capacity system that keeps small amounts of information available for ongoing tasks. Previous WM studies have primarily focused on where stored information is represented in the brain, with little consideration for the implications of future use of the memorized information on these representations. Here, we used fMRI and multivoxel pattern analysis (MVPA) to investigate the effects of the future use of the memorized information on WM representations. Human subjects (18-35years old) performed WM tasks requiring active reproduction or recognition of a previously encoded stimulus (color/shape) while their brain responses were collected with a 3T MRI scanner. Consistent with the various phases of the experimental paradigm (encoding, maintenance/retention and memory test), we observed robust univariate activations within the occipitotemporal, parietal, motor and prefrontal cortices. Multivariate analysis of brain responses

showed distinct representations for visual encoding of stimuli mainly within the early visual areas. In addition, the maintenance of information differed as a function of memory test (reproduction vs. recognition) in broad brain regions including visual, parietal, motor and prefrontal cortices. Our results suggest that neural representations in WM are task-dependent and may be contingent on the future use of the memorized information.

Disclosures: **P. Fiave:** None. **K. Jordan:** None. **K. Uittenhove:** None. **L. Bartsch:** None. **E. Vergauwe:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.15

Topic: H.05. Working Memory

Title: Effects of cerebellar theta burst stimulation (TBS) on working memory

Authors: *N. RAIES, J.-F. NANKOO, R. CHEN;
Univ. Hlth. Network (UHN), Toronto, ON, Canada

Abstract: The cerebellum is well-known for its role in motor control, but recent evidence suggests that the cerebellum is also involved in cognitive functions. Lesions of the cerebellum can lead to changes in working memory, which involves a system that temporarily holds and manipulates information. The contribution of the cerebellum to working memory is achieved through its connections with the prefrontal cortex (PFC). Previous studies showed that theta burst stimulation (TBS), a form of repetitive transcranial magnetic stimulation (rTMS), on the cerebellum changes its functional connectivity with the PFC. Specifically, excitatory intermittent TBS (iTBS) increases, whereas inhibitory continuous TBS (cTBS) decreases this functional connectivity. This study aims to further explore the cerebellar contribution to working memory by investigating the effects of cerebellar iTBS and cTBS on working memory performance. We expect that TBS will modulate the cerebellar contribution to working memory. We hypothesize that iTBS on the cerebellum will improve working memory, whereas cTBS will disrupt it. Nine participants (6 women and 3 men; age range: 42 - 79) participated in this ongoing study. Bilateral cerebellar stimulation was applied with a figure-of-eight coil at 3 cm lateral and 1 cm below the inion. The participants received iTBS, cTBS, and sham iTBS in three separate sessions in random order. Within 30 minutes after TBS, the participants performed three types of working memory tasks: letter 2-back, digit span forward (DSF), and digit span backward (DSB). The score ranges were 0-16 points for DSF and 0-14 points for DSB. For the 2-back task, we calculated the discriminability index (d-prime), which takes into account hits (percent correct) and false alarms (responses to non-target trials). The repeated measures ANOVA revealed a significant effect of the type of stimulation (iTBS/cTBS/Sham) on performance in the DSB task ($F(2, 16) = 3.85$; $p = 0.04$; $\eta_p^2 = 0.33$). Thus, we performed a planned comparison (a priori contrast), and it showed that scores in the cTBS condition were significantly lower than in the

sham condition ($t(16) = 2.67$; $p = 0.02$). iTBS and cTBS did not affect performance in the 2-back and DSF tasks compared to sham. The findings support the hypothesis that the cerebellum is involved in working memory, and this contribution may be disrupted by cTBS. The study is ongoing and more participants are being recruited.

Disclosures: **N. Raies:** None. **J. Nankoo:** None. **R. Chen:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

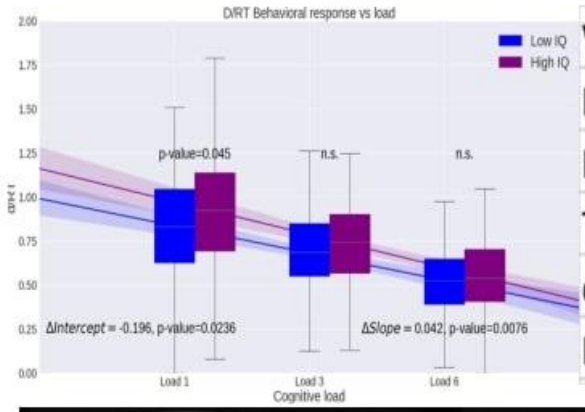
Program #/Poster #: 568.16

Topic: H.05. Working Memory

Title: An fmri study of delayed item recognition task supports neural efficiency hypothesis

Authors: *A. FERNANDEZ GUERRERO[1], Y. STERN[2], S. NAYAK[1], S. HOJJATI[1], R. RAZLIGHI[1];
[1]Weill Cornell Med., New York, NY; [2]Columbia, New York, NY

Abstract: The neural efficiency hypothesis posits that more intelligent individuals require less level of brain activation to perform a task successfully than less intelligent ones. Previous findings based on fMRI indicated that more intelligent individuals exhibit lower inhibition of the Default mode network (DMN) (Basten et al., 2013) and there were IQ-related differences in the brain cortical activation which tend to disappear by controlling for subjective difficulty (Dunst et al., 2014).We aimed to provide evidence for the neural efficiency hypothesis using fMRI activation/deactivation pattern during a delayed item recognition task (Letter Sternberg task or LS), in a group of 273 healthy adults aging from 20 to 80 years. We associated fMRI activations to IQ within 3 different large-scale brain networks (dorsal-attention network (DAN), DMN and visual) controlling for performance (ratio of d' to response time), brain structure (cortical thickness) and task load. The LS behavioral data indicates that high IQ individuals have better overall performance (Δintercept =0.192, $p<0.02$), whereas they have steeper slope in load-related performance decay (Fig. 1a). At the neuronal level, controlling for brain structure, task-load, and individual performance level, we found that during the probe phase higher IQ was significantly associated with less brain activation in the DAN ($p<0.004$) (Fig. 1b). In addition, greater cognitive load was associated with more DMN suppression, while DAN and visual cortex tended to increase its activation with greater load (Fig. 1c). In conclusion, greater cognitive load corresponds, in general, to greater activation in visual and DAN whereas it is associated with stronger deactivation in DMN. Furthermore, higher IQ were associated with less activation at least in probe phase of the LS task in support of neural efficiency hypothesis.



Variable	coef	t	P>|t|
Intercept	233.7335	2.7037	0.0071
IQ	-1.1626	-2.8945	0.004
Thickness	42.3726	1.6107	0.1078
d/RT	22.5005	2.2115	0.0274
Load	-6.9191	-4.1454	0.00001

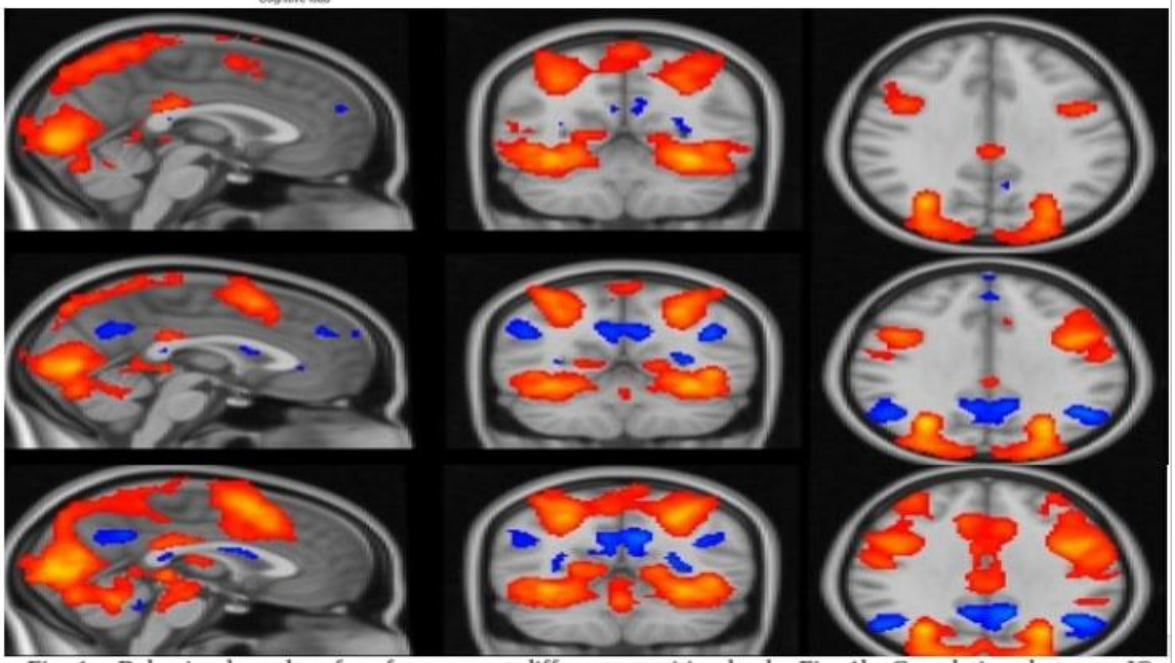


Fig. 1a: Behavioral results of performance at different cognitive loads. Fig. 1b: Correlations between IQ, thickness and performance on Pro activation over DAN. Fig. 1c: DAN and DMN networks on the stimulus presentation of 6 items (highest load).

Disclosures: **A. Fernandez Guerrero:** None. **Y. Stern:** None. **S. Nayak:** None. **S. Hojjati:** None. **R. Razlighi:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.17

Topic: H.05. Working Memory

Title: Pupil Size Tracks Graded Functional States of Working Memory Maintenance

Authors: *Y. DONG, A. KIYONAGA;
Cognitive Sci., UCSD, La Jolla, CA

Abstract: The temporary maintenance of information with working memory (WM) enables goal-oriented behavior and interacts with perceptual processing. Behavioral and physiological evidence suggest that WM items at varying levels of task relevance (or 'priority') might be held in distinct representational states, distributed across the brain. Yet it remains debated what role sensory systems play in WM maintenance. Recent studies have shown that pupil size reflects the brightness of an item that is internally maintained in WM - remembering a darker WM item elicits larger pupil dilation than brighter items, even when sensory input is equivalent. Like spatial attention processes, this WM-mediated pupillary response is likely controlled by the oculomotor orienting circuit, including Frontal Eye Field and Superior Colliculus. This pupil effect raises the question of whether peripheral sensory apparatus serve a functional role in supporting WM. To address this, here we test whether the WM-mediated pupil response reflects the features and attentional state of WM content. During eye-tracking, participants completed a retrocue WM task for dark and bright WM stimuli. We manipulated the relative priority state among two WM items by varying the reliability of the retrocue across blocks. Behaviorally, memory recall was better for cued (prioritized) than uncued items, and this effect was larger in blocks with most predictive cues. Physiologically, we replicated previous findings that remembering a darker WM item is associated with larger pupil size (vs. a bright item). Additionally, this effect was modulated by the degree of prioritization of the WM item: the WM-mediated pupil dilation during maintenance was greatest in the condition when the retrocue was most reliable. These initial results suggest that pupil modulations track the neural representational state of WM content, with graded degrees of pupil modulation for items at varying priority levels. This experiment demonstrated that manipulating WM can modulate the pupils, while follow-up experiments will manipulate pupil dilation to probe the effects on WM. Together, these experiments will illuminate the role of early visual processing in WM maintenance, as well as the feasibility of using pupillary measures as an index of WM.

Disclosures: Y. Dong: None. **A. Kiyonaga:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.18

Topic: H.05. Working Memory

Support: NIH Grant F32MH124268

Title: Causal contribution of cortico-cerebellar regions to visual working memory

Authors: *J. BRISSENDEN, T. LEE;
Univ. of Michigan, Ann Arbor, MI

Abstract: Working memory (WM) enables the short-term maintenance of mental representations in support of ongoing cognitive operations. A substantial body of literature has sought to reveal the specific brain structures and mechanisms that support WM. To date, much of this literature has used correlational measurement techniques to investigate the neural substrates of WM. We sought to investigate the causal role of brain areas across cerebral cortex and cerebellum in WM using a combination of functional magnetic resonance imaging (fMRI) and transcranial magnetic stimulation. Across multiple sessions we applied continuous theta-burst stimulation (TBS) to disrupt activity in parietal cortex, frontal cortex, cerebellum, and a control site immediately prior to the performance of a continuous report spatial working memory task on a computer or in an fMRI scanner. A baseline fMRI session collected structural MRI, population receptive field mapping and resting-state scans. We examined the effect of this disruption on parameters of a variable precision mixture model fit to the recall error distribution for each session. We also investigated how the perturbation of each area affected the probabilistic decoding of remembered location from BOLD activity across the brain. TBS to each of our regions of interest resulted in reduced recall precision as well as increased precision variability relative to control stimulation. The probability of guessing was unaffected by TBS. We further showed that stimulation of our regions of interest decreased decoding accuracy and increased decoding uncertainty relative to the control site (van Bergen et al., 2015). These results provide evidence for a causal contribution to working memory maintenance by a distributed network of areas spanning both cerebral cortex and cerebellum.

Disclosures: J. Brissenden: None. **T. Lee:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.19

Topic: H.05. Working Memory

Support: GACR grant 21-44843L

Title: High frequency ripple oscillations in human memory encoding and recall

Authors: *J. CIMBALNIK[1], **L. JURKOVICOVA**[1,2,3], J. DOLEZAL[5], M. LECH[6], M. PAIL[3], R. ROMAN[7,2,4], M. T. KUCEWICZ[8];
[1]Intl. Clin. Res. Center, St. Anne's Univ. Hosp., Brno, Czech Republic; [2]Behavioral and Social Neurosci. Res. Group, CEITEC – Central European Inst. of Technology, Masaryk Univ., Brno, Czech Republic; [3]Brno Epilepsy Center, Dept. of Neurology, St. Anne’s Univ. Hospital, Fac. of Medicine, Masaryk Univ., Brno, Czech Republic; [4]Brno Epilepsy Center, Dept. of Neurology, St. Anne’s Univ. Hospital, Fac. of Medicine, Masaryk Univ., Brno, Cyprus; [5]Czech Inst. of Informatics, Robotics and Cybernetics, Czech Tech. Univ. in Prague, Prague, Czech Republic; [6]Brain and Mind Electrophysiology laboratory, Multimedia Systems Department, Fac. of Electronics, Telecommunications and Informatics, Gdansk Univ. of Technol., Gdansk, Poland;

[7]Dept. of Biomed. Engineering, Intl. Clin. Res. Center, St. Anne's Univ. Hosp. Brno, Brno, Czech Republic; [8]Multimedia Systems, Gdansk Univ. of Technol., Gdansk, Poland

Abstract: High frequency ripple oscillations (HFO, 80 - 200 Hz) are induced by cognitive processing and likely represent underlying neuronal activity. These events can be directly recorded from the human brain of epileptic patients who undergo monitoring with implanted intracranial or subdural electrodes as part of their epilepsy diagnosis. In this study, we analyzed the occurrence of HFO across brain structures during word presentation and memory encoding tasks. We automatically detected HFO in intracranial EEG data sampled at 5 kHz from 7 patients who performed two cognitive tasks. In the word screening (WS) task 180 distinct nouns were presented in 5 pseudorandomly ordered trials (i.e. each word 5 times). During the free recall (FR) task the patients were asked to remember 12 words presented one at a time, and then to recall them in any order after a short distractor task with simple algebraic equations. 15 trials were performed by each patient (180 words in total matched with the WS task).To determine HFO active channels in performed tasks we binned the detected HFO into 100ms windows from the word presentation to one second after presentation and identified the channels where the number of detections was higher than mean+3*std of all channels. Channels where at least one time bin was identified were marked as active. To investigate whether the same brain sites are activated during WS task and the encoding phase of the FR task we compared the number of active channels in each anatomical structure between the two tasks. To further determine whether there are specific word responses in the individual channels and structures we compared individual time bins to the mean+3*std of the average HFO rate across all words. We then calculated the percentage of the presented words that showed HFO activation. To assess whether any brain structures are connected to memory encoding we calculated the proportion of recalled words during FR task that showed HFO response during the FR encoding phase. The most active channels during WS were found in the lingual gyrus (N=10), precuneus (N=9) and middle temporal gyrus (N=8). The same structures were active during FR encoding with additional increased activation in orbital gyrus (N=5). The highest proportion of HFO word responses during WS task was observed in supramarginal gyrus (61% of the presented words) and superior frontal gyrus (54% of the presented words). The same structures showed a high percentage of word HFO activation of words that were subsequently recalled during the FR task. Word presentation and encoding show ripple activation in brain areas responsible for visual and word processing, language and memory.

Disclosures: J. Cimbalnik: None. **L. Jurkovicova:** None. **J. Dolezal:** A. Employment/Salary (full or part-time):; Medicton Group Ltd. (Prague, Czech Republic). **M. Lech:** None. **M. Pail:** None. **R. Roman:** None. **M.T. Kucewicz:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.20

Topic: H.05. Working Memory

Support: NIH-NEI R00 EY025768
Brain & Behavior Research Foundation #28174
Alfred P. Sloan Foundation FG-2020-13415

Title: Delta/theta band of non-human primate EEG reflects memory-dependent responses in anticipation of distractors during a visual working memory task

Authors: *D. Y. JUNG[1], A. C. SNYDER[2];
[1]Neurosci. Grad. Program, [2]Neurosci., Univ. of Rochester, Rochester, NY

Abstract: Visual working memory allows maintenance of relevant visual information for near-term goals. Successful maintenance depends not only on accurate memory representation but also on controlled suppression of goal-irrelevant external events, such as distracting perceptual inputs (i.e., distractors). Distractors may be effectively suppressed if they can be anticipated. Some human electroencephalogram (EEG) studies showed changes in specific EEG band activity (e.g., delta (2-4Hz) or alpha band (8-12Hz)) pertaining to anticipation, but whether the same changes occur in animal models remains largely unexplored, especially in a non-human primate (NHP) model. An NHP model gives a significant advantage in studying whole-brain activity (e.g., EEG) and neuronal signals (e.g., spike) simultaneously, thereby providing a key translational approach to understanding human cognitive processes. In this study, we tested how NHP EEG activity changes in anticipation of distractors during memory maintenance. We had three questions to address. First, how does a distractor(s) influence the memory at the varying time points during maintenance? Secondly, is there a specific EEG band(s) reflecting distractor anticipation/suppression? Lastly, can we infer the memory from the anticipatory EEG activity? We recorded EEG from NHPs performing a memory-guided saccade task: remember the location of a briefly presented visual stimulus then saccade to the remembered location after a delay. During the delay, a distractor (a full-field checkerboard pattern visual stimulus) was flashed. We varied the task in two ways: a distractor(s) appeared either (1) at a single time point on half of the trials or (2) at different time points with equal probability.
We found significant EEG band activity changes in the delta/theta band (1-6Hz) in anticipation of distractors in both variations of the task. We wanted to test whether the delta/theta EEG band also reflects memory-related information through decoding approaches. We found significant memory decoding performance when distractors were not presented. This finding suggests that the anticipatory EEG signals reflect suppression of distracting perceptual inputs during memory maintenance as a potential mechanism to protect memory.

Disclosures: D.Y. Jung: None. **A.C. Snyder:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.21

Topic: H.05. Working Memory

Support: NIH Grant R01 AG047972-01A1

Title: Age-related changes to neurovascular coupling dynamics in a Sternberg task with parametrically varying task demand

Authors: Y. ZHAO[1], D. H. ABDELKARIM[2], M. P. TURNER[3], P. LIU[4], K. HAZEL[5], H. LU[6], *B. RYPMA[1];
[1]Univ. of Texas At Dallas, Dallas, TX; [2]UT Dallas, Dallas, TX; [3]Univ. of Texas at Dallas, Dallas, TX; [4]Univ. of Maryland, Baltimore, MD; [5]Johns Hopkins, Baltimore, MD; [6]Johns Hopkins Univ., Baltimore, MD

Abstract: Age-related decline in the modulation of neural recruitment with increasing cognitive task demand has been observed in many studies using blood-oxygen-level-dependent (BOLD) signal. We measured age-changes in demand-related neural modulation in physiological factors underlying BOLD signal while participants performed a Sternberg working memory task (SWMT). In the SWMT, participants were presented with letters that they were instructed to hold in their memory. After a delay period, a probe letter appeared and participants determined if the probe letter was among the original set of letters they had been asked to remember. They responded by pressing a right or left thumb-button. The letter set-size varied parametrically between 2-, 4-, and 6 letters. Utilizing a dual-echo fMRI sequence, participants' BOLD signal and cerebral blood flow (CBF) during the task were simultaneously measured. A CO_2 ingestion procedure enabled the calculation of cerebral metabolic rate of oxygen ($CMRO_2$). After pre-processing, general-linear-modeling was conducted independently for BOLD and CBF to model the task-evoked signal changes. The resulting BOLD and CBF parameter estimates were used to calculate voxel-wise percent signal change (PSC). $CMRO_2$ changes were then calculated using the deoxyhemoglobin dilution model. Accuracy showed significant main effect of set-size, but neither the main effect of age-group or the age-group by set-size interaction was significant. The main effect of age-group and set-size were both significant in reaction time, with no age by load interaction. In both age groups, reaction time (RT) increased monotonically with set-size increases. Task positive ΔBOLD and ΔCBF signals were observed in prefrontal and parietal regions in both groups. As set-size increased, the focus of the signals shifted from parietal to prefrontal regions. Age-increases in signal strength and extent were also observed. Between ΔBOLD and ΔCBF measures, there were considerable differences in the pattern of task positive signal. Especially, ΔCBF signal extent were more spatially restricted than ΔBOLD in younger adults but not in older adults. This result suggests that there were age-differences in CBF-BOLD dynamics during SWMT performance, possibly due to age-related changes in neurovascular coupling.

Disclosures: Y. Zhao: None. **D.H. Abdelkarim:** None. **M.P. Turner:** None. **P. Liu:** None. **K. Hazel:** None. **H. Lu:** None. **B. Rypma:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.22

Topic: H.05. Working Memory

Support: Vidi grant 016.Vidi.185.137

Title: Oscillatory building blocks underlying perceptual decision making

Authors: *H. ELSHAFEI[1], J. ZHOU[1], S. HAEGENS[1,2,3];
[1]Donders Inst. For Brain, Cognition and Behavior, Nijmegen, Netherlands; [2]Dept. of Psychiatry, Columbia Univ., New York, NY; [3]Div. of Systems Neurosci., New York State Psychiatric Inst., New York, NY

Abstract: Neural oscillations in different frequency bands are currently being linked to various cognitive functions e.g., theta with memory and alpha with attention. Instead, we propose that the role of neural oscillations is to control communication by providing building blocks, or low-level operations, that can be employed for these higher-level cognitive functions. In this study, we targeted three rhythms: delta/theta, alpha and beta. We recorded magnetoencephalography (MEG) in human participants performing a visual delayed match-to-sample paradigm in which orientation or spatial frequency of sample and probe gratings had to be matched. A cue occurring before or after sample presentation indicated the to-be-matched feature. We demonstrate that alpha/beta power decrease tracks the presentation of the informative cue and indexes faster responses. Moreover, these faster responses coincided with an augmented phase alignment of slow oscillations, as well as phase-amplitude coupling between slow and fast oscillations. Importantly, stimulus decodability was boosted by both low alpha power and high beta power. In summary, we provide support for a comprehensive framework in which different rhythms play specific roles: slow rhythms control input sampling, while alpha (and beta) gates the information flow, beta recruits task-relevant circuits, and the timing of faster oscillations is controlled by slower ones.

Disclosures: H. Elshafei: None. **J. Zhou:** None. **S. Haegens:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.23

Topic: H.05. Working Memory

Support: NIH Grant R01EY028746

Title: Removal of information from working memory via suppression leads to forgetting

Authors: *Z. BRETTON-GRANATOOR[1], H. KIM[2], M. T. BANICH[2], J. A. LEWIS-PEACOCK[1];

[1]Inst. for Neurosci., Univ. Of Texas at Austin, Austin, TX; [2]Inst. of Cognitive Sci., Univ. of Colorado Boulder, Boulder, CO

Abstract: A thought can be intentionally removed from working memory by, for example, replacing it with another thought, suppressing that particular thought, or clearing the mind of all thoughts. These removal operations are associated with distinct neural patterns of activity in cognitive control regions and have differential impacts on working memory. The long-term consequences of these removal operations, however, are currently unknown. Based on the sensory recruitment model of working memory, we hypothesized that intentionally removing items from working memory, in some cases, could lead to the subsequent forgetting of those items. In this study, we investigated how working memory removal operations can induce forgetting in long-term memory through changes to the representation of the information during its removal. We collected fMRI data (N = 25) while participants performed a working memory removal task with pictures of natural and manmade landscapes, followed by a surprise long-term memory test of the studied items. We used category-level multivariate pattern classification and item-level representational similarity analysis to assess the neural representation of memory items before, during, and after being removed from working memory. Then we linked the neural measurements to the subsequent recognition memory outcomes for these items. Our results show that suppressing an item in working memory produced lasting changes to the representation of that item in long-term memory. Simply replacing an item in working memory did not. This neural result was also behaviorally relevant – items that were forgotten showed greater representational changes after suppression compared to items that were later remembered. These findings indicate that when an item is intentionally removed from working memory via suppression, this degrades its representation in long-term memory, which can lead to subsequent forgetting of that item. This research allows us to better understand the longer-term behavioral and neural consequences of removing information from working memory, thus providing vital insights into the impacts of cognitive control on memory.

Disclosures: Z. Bretton-Granatoor: None. **H. Kim:** None. **M.T. Banich:** None. **J.A. Lewis-Peacock:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.24

Topic: H.05. Working Memory

Support: KL2TR003097

Title: Assessing neural correlates of working memory deficits in pediatric patients with sickle cell disease

Authors: *K. REDDY[1], T. D. TUCKER[2], V. C. SEGHATOL-ESLAMI[2], E. C. LEE[1], J. A. TRAPANI[2], S. M. CARTAGENA[3], A. M. GRIMM[3], D. C. TUCKER[2], K. TRIEBEL[2], D. L. MURDAUGH[3];
[1]Col. of Arts and Sci., [2]Dept. of Psychology, [3]Dept. of Pediatrics, The Univ. of Alabama at Birmingham, Birmingham, AL

Abstract: Sickle cell disease (SCD) is the most common genetic red blood cell disorder in the African American population. Neurocognitive impairment is a pervasive debilitating feature of SCD across the lifespan. In pediatric patients with SCD, a gap in the research is the neural correlates of working memory (WM)—a neurocognitive domain that is consistently identified as being impaired in these patients. This study investigated differences in resting-state functional connectivity (RS-FC) in WM brain regions between SCD patients and age-matched community controls. Twenty SCD patients (12-16y) and twelve controls (10-18y) completed both objective and subjective measures of WM, in addition to resting-state fMRI. Two SCD participants were excluded from the final analyses due to excessive head motion during the scan. Linear regressions were conducted to assess between-group differences in objective and subjective measures of WM, while accounting for demographic-related factors. Resting-state fMRI data was pre-processed using standard approach (SPM12). The WM network, including the medial prefrontal cortex (MPFC), anterior cingulate cortex (ACC), superior (SPL) and inferior parietal lobule (IPL), and middle temporal lobule (MTL), and the salience network (SN) were explored. Whole-brain, seed-to-voxel, between-group analyses of the WM and SN networks were conducted in order to assess differences in RS-FC within and outside of these networks between SCD patients and controls. Results revealed that subjective, parent-reported WM concerns were significantly higher in SCD participants compared to controls ($t=2.109$, $p=.046$). Self-reported WM and objective measures of WM were not significantly different between groups. Neuroimaging results revealed significant underconnectivity (pFDR-corrected=.012) of the WM network to hippocampal regions in SCD patients compared to controls. Within the SN, there was a significant underconnectivity of the SN to regions of the cingulate gyrus and cuneus (pFDR-corrected=.019; pFDR-corrected=.043) among SCD patients compared to controls. Overall, these results demonstrate alterations of connectivity in regions associated with WM among SCD patients, which is consistent with parent-reported concerns of everyday WM functioning, but was not related to objective measures of WM. These findings identify neural correlates to WM deficits and suggest that multimodal assessments may be necessary to fully understand the physiological and neurocognitive impacts of SCD mechanisms on WM.

Disclosures: K. Reddy: None. **T.D. Tucker:** None. **V.C. Seghatol-Eslami:** None. **E.C. Lee:** None. **J.A. Trapani:** None. **S.M. Cartagena:** None. **A.M. Grimm:** None. **D.C. Tucker:** None. **K. Triebel:** None. **D.L. Murdaugh:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.25

Topic: H.05. Working Memory

Support: Swiss National Science Foundation grant P2FRP3_195083

Title: Alpha phase-coding supports feature binding in working memory

Authors: *M. F. PAGNOTTA[1], A. SANTO ANGLES[2], M. D'ESPOSITO[1], K. K. SREENIVASAN[2];
[1]Helen Wills Neurosci. Inst., Univ. of California, Berkeley, Berkeley, CA; [2]Div. of Sci. and Mathematics, New York University, Abu Dhabi, Abu Dhabi, United Arab Emirates

Abstract: Successful working memory (WM) for complex objects requires not only that each objects' individual features are temporarily held in mind, but also that these are bound together into coherent representations. The mechanisms supporting feature binding in WM remain unclear. When subjects are asked to hold multiple items in WM and report a feature of one item, 'swap' errors occur when an inaccurate response to the target item is accurate relative to a non-target item - e.g., if a subject shown a red square and a blue circle mistakenly reports the color of the circle as red. Swaps reflect the failure to maintain the correct feature binding, and can thus offer important insights into its underlying neural mechanisms. A recent biophysical network model (Barbosa et al., 2021) proposes that features in WM are bound through low-frequency synchrony, and swaps result from phase synchrony perturbations. We tested the neurophysiological prediction of this model by using MEG data collected from 26 human subjects in a task designed to induce swaps. Subjects were briefly shown 3 circles (0.2 s) and held their colors and locations in WM. After memory delay (2 s), subjects reported the location of each of the circles, sequentially cued by their color. We used a maximum likelihood approach to distinguish swaps (location of one of the non-cued circles mistakenly reported) from high-precision (HP) trials (location of all circles reported accurately). We estimated the Phase Preservation Index (PPI), which captures the consistency in phase differences across trials with respect to memory delay onset, separately for trial type. PPI was significantly higher in HP trials than swaps in alpha-band (~10 Hz) over parieto-occipital sensors. Importantly, PPI differences did not generalize to low-precision reports of cued location, suggesting that phase inconsistencies are a hallmark of swaps. To understand why the phase preservation may be compromised, we compared the standard deviation of frequency sliding (FS), which measures single-trial temporal fluctuations in oscillation peak frequency, between trial types. Swaps were characterized by increased variability in FS compared to HP trials in the same sensors that exhibited PPI differences. Our findings confirm the central prediction that swaps are characterized by less phase preservation during delay. Moreover, they indicate that phase inconsistencies arise through increased temporal coding variability. These results support the notion that feature binding in WM is accomplished through phase-coding dynamics that emerge from the competition between different memories, which may result from lateral inhibition between neural networks.

Disclosures: M.F. Pagnotta: None. **A. Santo Angles:** None. **M. D'Esposito:** None. **K.K. Sreenivasan:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.26

Topic: H.05. Working Memory

Support: National Institute of Health (MH064498)

Title: When are task-irrelevant features actively encoded into visual working memory?

Authors: *Y. DING[1], J. FULVIO[1], M. PIETRELLI[1], B. R. POSTLE[2];
[1]university of wisconsin-madison, Madison, WI; [2]Psychology and Psychiatry, Univ. Of Wisconsin–Madison, Madison, WI

Abstract: Maintaining an object composed of multiple features in visual working memory (VWM) internally represents these features as a conjunction, as well as binding the object to a representation of the context in which it was encountered (e.g., its location in space, or the order in which it appeared within a series of presented objects). Recent studies observed larger ERPs at contralateral parietal-occipital electrodes during stimulus encoding and at bilateral sites during delay period on trials in which location context was necessary for memory retrieval. Additionally, several studies have demonstrated that stimulus location can be decoded from EEG and fMRI signals even when it is not required for task performance. This suggests that the binding of item location information is automatic, perhaps obligatory. Our current study aims to 1) find a neural marker of binding multiple features in VWM by exploring the time course from encoding and maintaining to retrieving; 2) investigate whether only spatial positions or generally a visual feature can be spontaneously represented when encoding another feature. Each trial begins with a cue (200 msec), followed by the bilateral presentation of arrays of one or two sample items (750 msec), then an unfilled delay (900 msec), and finally a recognition probe. Samples are circular oriented-grating patches that can appear in any of 5 colors. To manipulate the importance of feature binding, trials are blocked according to the dimensions along which invalid probes can differ from the samples: only color; either color or location; or either color or orientation. Preliminary behavioral data have revealed a load effect (i.e., higher accuracy and faster reaction times (RT) for 1 vs. 2 items) and an effect of binding demands (i.e., higher accuracy and faster RTs for color-only vs. color + location and color + orientation). Note that these results argue against the “strong object hypothesis,” in a manner insensitive to the number of features that define an object. In our main experiment, we will record EEG with these same behavioral procedures, to test the following predictions: in addition to replicating the preliminary behavioral results, we will observe 1) the ERP effects mentioned above; 2) elevated power in mid-frontal theta-band oscillations during high-binding conditions; 3) successful decoding of stimulus location and orientation in all three memory conditions (consistent with the automaticity of this operation). Confirmation of these hypotheses will identify a neural marker of binding multiple features in VWM, and establish boundary conditions regarding whether and when a task-irrelevant visual feature is actively represented in VWM.

Disclosures: Y. Ding: None. **J. Fulvio:** None. **M. Pietrelli:** None. **B.R. Postle:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.27

Topic: H.05. Working Memory

Support: ONR Grant N00014-17-1-2041
Swartz Foundation
NIH Grant R01MH062349
Simons Collaboration on the Global Brain program grants 543057SPI
National Key R&D Program of China (Grant No. 2021ZD0201300)
National Natural Science Foundation of China (Grant No. 11975178)

Title: Emergence of distributed working memory in a large-scale macaque neocortex: bifurcation in hierarchical space

Authors: *U. PEREIRA OBILINOVIC[1], J. JIANG[2], X.-J. WANG[1];
[1]Ctr. For Neural Sci., New York Univ., New York, NY; [2]Sch. of Life Sci. and Technol., Xi'an Jiaotong Univ., Xi'an, China

Abstract: Numerous electrophysiological and imaging studies in non-human primates and humans demonstrate the distributed nature of working memory. Working memory-related sustained activity is rarely observed in the sensory areas but robustly displayed in the prefrontal, parietal, and inferotemporal association cortices. What are the organizing principles that determine the pattern of distributed working memory activity across the cortex? To address this question, we use a modeling framework that integrates a generative model for the mesoscopic connectivity of the macaque neocortex (Song et al., 2014) with biophysically realistic neuronal dynamics. First, we use the diffusion map method on the generated connectivity data to estimate the hierarchical position of each cortical region. Using this hierarchy, we define a macroscopic gradient for the strength of excitation across the cortical mantle, consistent with experimental observations. Then, we use the resulting connectivity and gradient of excitation to construct a large-scale model of the macaque neocortex. Our model naturally displays distributed working memory-related persistent activity patterns, consistent with experimental observations. Strikingly, there is a sudden transition in firing rate activity along the hierarchy, determining which brain region will present working memory-related persistent activity. This phenomenon corresponds to a bifurcation in the hierarchy space. The critical hierarchical position when this transition occurs in a distributed working memory state corresponds to the bifurcation point. Around this point in the hierarchy, slow fluctuations or switching between background and persistent activity are present. Thus, during distributed working memory states, the timescale of cortical areas around this bifurcation point increases dramatically from milliseconds to seconds, displaying a non-monotonic timescale profile in the hierarchical space. On the other hand, for background activity, the timescale for cortical areas increases monotonically with the hierarchy from sensory to association areas, as previously reported. We show this bifurcation in space is organized by a cusp bifurcation in parameter space, providing with this a simple geometrical explanation for the observed phenomenon. Together, our work suggests that working memory

states are organized around a bifurcation in cortical hierarchical space, and the signatures of this bifurcation are predicted to be present in cortical timescales.

Disclosures: U. Pereira Obilinovic: None. **J. Jiang:** None. **X. Wang:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.28

Topic: H.05. Working Memory

Title: Visual working memory recruits two functionally distinct alpha rhythms in posterior cortex

Authors: *J. RODRIGUEZ-LARIOS[1], A. ELSHAFEI[2], M. WIEHE[2], S. HAEGENS[3]; [1]Columbia Univ., New York, NY; [2]Donders Inst., Nijmegen, Netherlands; [3]Dept. of Psychiatry, Div. of Systems Neurosci., Donders Inst. For Brain, Cognition & Behaviour, New York, NY

Abstract: Oscillatory activity in the human brain is dominated by posterior alpha oscillations (8-14 Hz), which have been shown to be functionally relevant in a wide variety of cognitive tasks. Although posterior alpha oscillations are commonly considered a single oscillator anchored at an individual alpha frequency (IAF; ~10 Hz), previous work suggests that IAF reflects a spatial mixture of different brain rhythms. In this study, we assess whether Independent Component Analysis (ICA) can disentangle functionally distinct posterior alpha rhythms in the context of visual short-term memory retention. Magnetoencephalography (MEG) was recorded in 33 subjects while performing a visual working memory task. Group analysis at sensor level suggested the existence of a single posterior alpha oscillator that increases in power and decreases in frequency during memory retention. Conversely, single-subject analysis of independent components revealed the existence of two dissociable alpha rhythms: one that increases in power during memory retention (Alpha1) and another one that decreases in power (Alpha2). Alpha1 and Alpha2 rhythms were differentially modulated by the presence of visual distractors (Alpha1 increased in power while Alpha2 decreased) and had an opposite relationship with accuracy (positive for Alpha1 and negative for Alpha2). In addition, Alpha1 rhythms showed a lower peak frequency, a narrower peak width, a greater relative peak amplitude and a more central source than Alpha2 rhythms. Together, our results demonstrate that modulations in posterior alpha oscillations during short-term memory retention reflect the dynamics of at least two distinct brain rhythms with different functions and spatiospectral characteristics.

Disclosures: J. Rodriguez-Larios: None. **A. Elshafei:** None. **M. Wiehe:** None. **S. Haegens:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.29

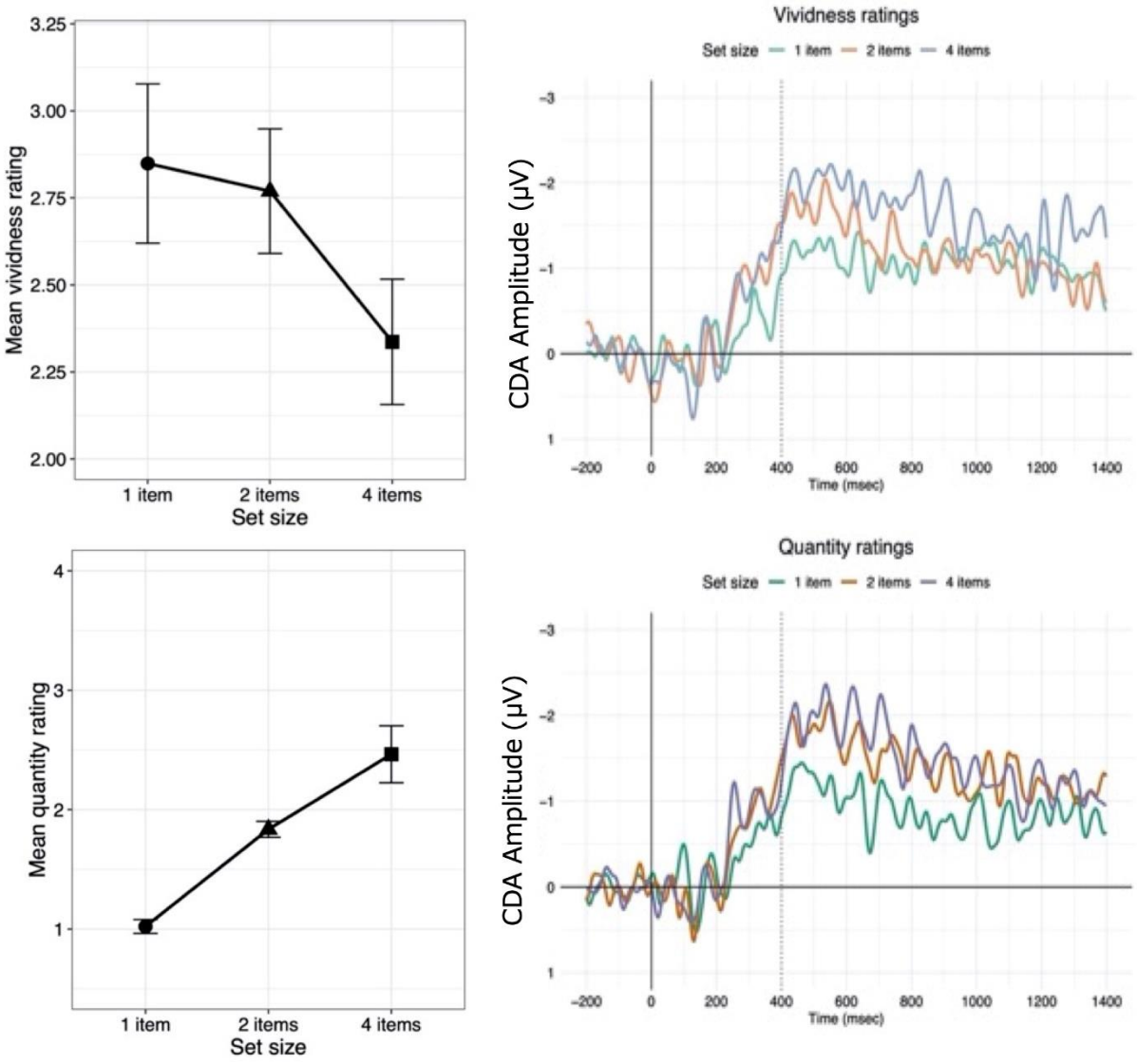
Topic: H.05. Working Memory

Support: JPMJCA2208
JPMJCE1311
JPMJMS2297
1+3 ESRC PhD studentship, reference number: 1788622

Title: How visual is visual working memory? Neural evidence for a distinction between the subjective experience of quality and quantity of mental imagery

Authors: K. E. BATES[1], M. L. SMITH[2], E. K. FARRAN[3], *M. G. MACHIZAWA[4];
[1]Dept. of Psychology, King's Col. London, London, United Kingdom; [2]Dept. of Psychological Sci., Birkbeck, Univ. of London, London, United Kingdom; [3]Dept. of Psychology, Univ. of Surrey, Guildford, United Kingdom; [4]Ctr. for Brain, Mind & KANSEI Sci. Res., Hiroshima Univ., Hiroshima-Shi, Japan

Abstract: Our ability to generate perceptual phenomena in mind allows us to contemplate the future and remember the past. Mental imagery (MI) is defined as the ability to *generate* visual mental images in mind in the *absence* of sensory input. MI is often likened to visual working memory (VWM): the ability to maintain and manipulate visual representations. However, the evidence does not yet warrant this conclusion. In a modified orientation change-discrimination task, the relations between meta-cognitive performance on MI, the focus of attention, and neural substrates of VWM were investigated. During the maintenance period, 23 participants guessed the quality (vividness) and quantity (number) of their MI while they were instructed to focus on either precision or capacity of representation to retain at set-sizes (1, 2 and 4). As a neural marker for visual working memory, the contralateral delay activity (CDA) and their behavioural performance were assessed. As was expected, both vividness and quantity ratings varied over set-sizes; however, those subjective ratings and behavioral performance was not correlated (*p*s > 05, *n.s.*) except for vividness rating at set-size 1 ($r_{Spearman}$ = .578, p = .015). As for the neural responses, CDA mimicked the lack of correlation such that there were no statistical differences between reported 'high and low' vividness and 'many and less' capacity conditions. Contrary to our expectations, individuals appear to have poor insight into the precision and capacity of their MI. These sets of evidence feature that the subjective sensory experience of MI is distinct from the visual quality and quantity of VWM. This study has methodological implications for examining how individual differences in MI support VWM and contribute to the theoretical interpretations of the role of consciousness in VWM. Ultimately, this study opens debate for the theories on VWM and MI supported by the intriguing interaction between meta-cognition of MI and the behavioural and neural correlates of VWM.



Disclosures: **K.E. Bates:** None. **M.L. Smith:** None. **E.K. Farran:** None. **M.G. Machizawa:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Xiberlinc Inc..

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.30

Topic: H.05. Working Memory

Support: TÜBİTAK Grant 120K924

Title: Attended and conscious task relevant WM information may not be decodable from frontoparietal regions if they are not linked to different goals

Authors: *G. SENGIL[1], A. A. FAROOQUI[2];
[1]Bilkent Univ., Ankara, Turkey; [2]Psychology, Bilkent Univ., ankara, Turkey

Abstract: A set of FrontoParietal regions (FP) activates during all kinds of demanding tasks. How these regions control cognition is a key outstanding question. Many accounts purport that these regions represent contents of working-memory/attention/conscious-awareness, and thus are the basis of attention/WM/consciousness undergirding any cognitive control. Indeed, MVPA studies have shown the decodability of working memory/ conscious representations from FP regions. However, in the design of these studies the different representations being decoded were linked to different goals. Their decodability is thus evidence of the decodability of different goal-directed programs and not of contents of WM/consciousness. Indeed, previous work (Farooqui & Manly, 2018) has raised the possibility that these regions activate primarily to goal completions and not to working memory/attention/consciousness. In fact, the latter when dissociated from goal completions, had deactivated these regions. Can attended, conscious, WM representations that are not linked to different goals be decoded from the pattern of activity of control related FP regions? We created an extended task in which picture identities to be decoded were not linked to different tasks or goals. In a modified visual n-back experiment, participants saw a series of pictures and covertly monitored them to see if any picture repeated after n steps and kept a count of such repeats. At the end of the series, they answered how many repeats had occurred. The identity of a picture, especially when repeated, was thus unambiguously attended, conscious and present in WM. At the same time, different repeating pictures were not linked to different tasks or goals. Both in regions of interest as well as wholebrain searchlight analysis, we found that the identities of such pictures were only decodable from visual regions (three local maxima extended along bilateral calcarine fissure, bilateral lingual gyrus, bilateral middle occipital gyrus, left middle occipital gyrus and left supplementary occipital gyrus, $p(FDR)<0.05$) and crucially not from any of the control-related FP regions. We suggest that attended and conscious information present in WM, even when task-relevant, may not be decodable from the activity patterns of FP regions if they are not part of different goal-directed programs. (This work is supported by The Scientific and Technological Research Council of Turkey, grant number: 120K924)

Disclosures: G. Sengil: None. **A.A. Farooqui:** None.

Poster

568. Working Memory: Neural Mechanisms and Oscillations

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 568.31

Topic: H.05. Working Memory

Support: The Scientific and Technological Research Council of Turkey, grant no: 120K924

Title: Not all rests are the same: Differential Fronto-Parietal Activity during Rests at Different Levels of Task Hierarchy

Authors: *I. GIRAY, I. ÇIFTÇI, G. ŞENGIL, A. FAROOQUI;
Bilkent Univ., Ankara, Turkey

Abstract: Our tasks are hierarchical. Tea may be prepared as part of a larger task of preparing breakfast and may in turn consist of smaller acts (e.g., boiling water). Rest are periods when cognition is not active with any task. Hierarchical nature of our tasks creates different kinds of rests. During some (e.g., resting after preparing tea but before preparing sandwich) cognition is still *within* the overarching task of preparing breakfast. During others (e.g., resting after preparing breakfast), the overarching task is over and the rest is *outside* it. We investigated which brain regions distinguished between these different kinds of rests. Crucially, this would be a way of knowing if and which brain regions represent long overarching tasks of durations extending into tens of minutes. The set of brain regions that remain active during rest periods within a long overarching task must be representing that task. Participants (n = 61) did a series of 15-minute functional magnetic resonance (fMRI) runs made of demanding N-back tasks. There were smaller rests (5-19 s) in between the trial blocks and longer rests (15-60 s) before starting and after completing the 15-minute run. We conducted whole-brain and region of interest (ROI) analyses and found that a set of fronto-parietal network (FPN) regions, previously associated with various task demands, remained active during rests that occurred within the experiment run. These included anterior cingulate cortex (Cohen's d = 0.70), supplementary motor area (d = 0.30), right inferior frontal sulcus (d = 0.45), right intraparietal sulcus (d = 0.59). These regions have previously been seen to be active during all kinds of task executions. It has been unclear if their activity was related to ubiquitous control demands, difficulty associated with such executions, or was related to task engagement per se. Our finding suggests that these regions are active during any kind of task engagement including those that involve no active control demands. Representation of task hierarchy has also been unclear. Different accounts have suggested the anterior prefrontal regions or the default mode network (DMN) regions as representing the overarching task. Current finding suggests that control-related FPN regions, purported to represent the lower-level aspects of task execution, are active in relation to the overarching and higher-level task. This work is supported by The Scientific and Technological Research Council of Turkey, grant no: 120K924

Disclosures: I. Giray: None. **I. Çiftçi:** None. **G. Şengil:** None. **A. Farooqui:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.01

Topic: H.06. Social Cognition

Support: NIH Grant R01 NS045193
NIH Grant R01 MH115750
NSF Grant PHY-1734030

Title: Effects of social context on free exploration in a mouse model of autism

Authors: *R. TAM, M. KISLIN, S. S.-H. WANG, J. W. SHAEVITZ;
Princeton Univ., Princeton, NJ

Abstract: Autism spectrum disorder (ASD) is associated with cerebellar malformation, marked by social, communicative, and behavioral challenges. Past studies in mice have shown differences in social preference between animals exhibiting autism-like behaviors and normal mice in a three-chambered environment, but social interactions have not been studied in freely behaving animals in an unobstructed arena. Here, we examine the social behavior of cerebellum-specific Tsc1 mutant and wild-type mice in the open field. We recorded bottom-up, 20-minute videos from 32 pairs of mice, including mutant-mutant, mutant-wild type, and wild type-wild type pairs. We used deep-learning based pose estimation software (SLEAP[1]) to track the animals' position over time and measure body-part dynamics. All mouse pairs exhibited complex chasing and locomotory behaviors, with differences in wildtype-wildtype, wildtype-mutant, and mutant-mutant pairs. In terms of the distance between animals, velocity, and heading, mutant Tsc1 mice avoided their social partners more often than wild-type mice. We next defined individual behaviors from pose dynamics by clustering wavelets derived for each body part[2,3]. This analysis defines a number of non-social behaviors including locomotion, grooming, and climbing, and one social behavior that occurs when mice are close to each other. We find that mutant mice engage in this social behavior less than wild-type mice and that mutant mice sometimes display social-like behavior at large distances from their partners.
1. Pereira, T.D. et al. SLEAP: A deep learning system for multi-animal pose tracking. Nat Methods 19, 486-495 (2022). https://doi.org/10.1038/s41592-022-01426-1.
2. Berman Gordon J., Choi Daniel M., Bialek William and Shaevitz Joshua W. 2014Mapping the stereotyped behaviour of freely moving fruit fliesJ. R. Soc. Interface.112014067220140672. http://doi.org/10.1098/rsif.2014.0672.
3. Klibaite U, Kislin M, Verpeut JL, Bergeler S, Sun X, Shaevitz JW, Wang SS. Deep phenotyping reveals movement phenotypes in mouse neurodevelopmental models. Mol Autism. 2022 Mar 12;13(1):12. http://doi.org/10.1186/s13229-022-00492-8.

Disclosures: R. Tam: None. **M. Kislin:** None. **S.S. Wang:** None. **J.W. Shaevitz:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.02

Topic: H.06. Social Cognition

Support: NIH/NIDCD 04845 (LMR)
Schmitt Program in Integrative Neuroscience
NIH/NIDCD 16419 (LMR)
F30 MH122048 (KKS)

Title: Neural Population Activity in the Primate Prefrontal Cortex During Perception of Audiovisual Expressions Reflects Identity

Authors: K. S. SHARMA, M. D. DILTZ, T. LINCOLN, O. RAHMAN, *L. M. ROMANSKI; Dept. of Neurosci., Univ. of Rochester Sch. of Med. and Dent., Rochester, NY

Abstract: The ventrolateral prefrontal cortex (VLPFC) is consistently active during the perception of faces and vocalizations, suggesting that the region has a critical role in normal social function. While categorical features of social stimuli, like identity and expression, have been shown to drive single unit and population activity in the temporal lobe (Yang and Freiwald, 2021; Gothard et al. 2007) the impact of these features on VLPFC neuronal activity is understudied and the social information encoded by the region is unclear. Additionally, single neurons in the VLPFC exhibit non-linear multisensory integration, suggesting that a single neuron's response to a face or voice in isolation does not scale to the population activity occurring during perception of dynamic communication stimuli. Thus, we tested whether neural responses to naturalistic stimuli were driven by identity or expression in the macaque VLPFC by recording neural activity in this brain region while 2 macaques viewed audiovisual face and vocal expressions from 3 unfamiliar conspecifics making expressions of aggressive, affiliative, and neutral valence. Of the 466 recorded neurons, 285 were responsive to the stimuli. The 285 neuron mean selectivity index for stimulus (0.41 ± 0.13), identity (0.22 ± 0.13), and expression (0.22 ± 0.13) indicated that single units were not particularly selective for single stimuli, identities, or expressions. Although a two-way ANOVA found significant main effects of identity, expression, or their interaction in the firing rates of 111 single neurons ($n=285$, $p < 0.05$), the mean decoding accuracies across the 285 single units for identity (0.38 ± 0.06) and expression (0.38 ± 0.05) were not appreciably higher than chance (0.33). However, when analyzed as a pseudo-population, decoding accuracy increased as a function of population size for both identity (0.80 at n=280) and expression (0.64 at n=280). Principle components analysis of mean population activity across time revealed that population responses to the same identity followed similar trajectories in the response space, facilitating segregation from other identities. Our results suggest that identity is a critical feature of social stimuli that dictates the structure of population activity in the VLPFC during the perception of expressions. These findings enhance our understanding of face processing and social behavior beyond the temporal lobe in macaques and humans.

Disclosures: K.S. Sharma: None. **M.D. Diltz:** None. **T. Lincoln:** None. **O. Rahman:** None. **L.M. Romanski:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.03

Title: WITHDRAWN

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.04

Topic: H.06. Social Cognition

Support: NIH Grant 5R01HD092055-05
Fyssen Foundation

Title: Neural correlates and effect of jealousy on cognitive flexibility in the female titi monkey (Plecturocebus cupreus)

Authors: *L. E. SAVIDGE[1], P. B. ZABLOCKI-THOMAS[2], S. M. FREEMAN[3], R. COTTERMAN[4], E. FERRER[4], K. L. BALES[5], L. R. WITCZAK[4];
[1]Univ. of California Davis, [2]California Natl. Primate Res. Ctr., Davis, CA; [3]Utah State Univ., Utah State Univ., Logan, UT; [4]Univ. of California Davis, Davis, CA; [5]1633 Hampton Dr, California Clin. Trials, Davis, CA

Abstract: Jealousy is a social emotion that elicits behavioral reactions from an individual toward a threat to a valuable relationship. Monogamous species exhibit jealousy-type behaviors as an adaptive response to preserve the relationship. Jealousy is also a complex emotion composed of several basic negative emotions: fear of loss, anxiety, suspiciousness and anger. Negative emotions like anxiety may impair cognitive processes such as cognitive flexibility, an ability important for coping with new situations. However, little is known about how complex social emotions influence cognitive flexibility. To understand the interaction between jealousy and cognitive flexibility, we examined the neural, physiological and behavioral factors involved in jealousy and cognitive flexibility in female titi monkeys. We presented subjects with a jealousy provoking scenario, followed by a reversal learning task and a PET scan. We found that female titi monkeys reacted to a jealousy provoking scenario with increased locomotor behavior and higher glucose uptake in the cerebellum; however, hormonal measures and cognitive flexibility were not affected. Glucose uptake in the orbitofrontal cortex (OFC) was significantly decreased during jealousy scenarios, while uptake in the anterior cingulate cortex (ACC) was decreased during reversal tasks. Our findings suggest that the presence of an intruder produces less visible behavioral reactions in female titis than in males and does not appear to impact cognitive flexibility.

Disclosures: L.E. Savidge: None. **P.B. Zablocki-Thomas:** None. **S.M. Freeman:** None. **R. Cotterman:** None. **E. Ferrer:** None. **K.L. Bales:** None. **L.R. Witczak:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.05

Topic: H.06. Social Cognition

Support: AMEDJPdm0307005
KAKENHI19K07810
KAKENHI22K07337
THE HORI SCIENCE AND ARTS FOUNDATION

Title: Enhancement of rhythmic motion entrainment by social contexts in the macaque

Authors: *S. TOMATSU, M. ISODA;
Natl. Inst. for Physiological Sci., NINS, Aichi, Japan

Abstract: Body movements are influenced by external factors including visual and auditory stimuli and physical constraints. When we move in a rhythmic manner in social contexts, e.g., clapping hands, one's bodily rhythm is easily affected by external rhythms made by other individuals. This phenomenon is called rhythm entrainment. Although rhythm entrainment is ubiquitous and thought to underlie social communication, little is known about its neural mechanism. This is partly because experimental animal models to test rhythm entrainment have not been established. Here, we trained Japanese monkeys (Macaca fuscata) to perform a rhythmic forearm movement task under various social and nonsocial conditions. The monkeys were trained to move a lever back and forth along a straight track at two different speeds. Explicit pacemakers, either visual or auditory, were never presented; the monkeys were given an abstract instruction to move 'fast' or 'slow' by a stationary visual cue. The task performance was judged to be correct if the duration of one-way motion was shorter than 500 ms in the fast condition and longer than 400 ms in the slow condition. Once the monkeys had learned these task rules, they performed the task in front of a monitor, in which movies of another monkey performing the same task (monkey-motion condition) or movies of a moving lever without a monkey (object-motion condition) were replayed at different motion speeds (0.3-2.7 Hz). We found that although no extra reward was given for synchronization with the motion stimuli, the monkeys' movement became faster when the movie was replayed at a faster speed in both conditions. This entrainment effect was larger when the monkeys' line of sight was within a range covering the moving lever. Notably, the entrainment effect was greater in the monkey-motion condition than in the object-motion condition. A further test using regularly blinking lights or rhythmic tones showed that the lack of motion stimuli caused no entrainment effect. These findings suggest that motion of external stimuli, in particular in social contexts, facilitates rhythm entrainment. Thus, macaque monkeys can be a good animal model to elucidate the neural mechanisms underlying socially driven rhythm entrainment.

Disclosures: S. Tomatsu: None. **M. Isoda:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.06

Topic: H.06. Social Cognition

Title: Cognitive and emotional effects of continuous or intermittent socialization in young adult rats

Authors: *A. TAPIA DE JESUS[1], J. A. MATA-LUÉVANOS[2], M. I. MATA, Sr.[3], O. C. GALICIA[4], M. H. BUENROSTRO-JAUREGUI[5];
[1]Univ. Iberoamericana, Ciudad DE Mexico, Mexico; [2]Lab. de Neurociencias, Dept. de Psicología, Univ. Iberoamericana, Ciudad de México, Mexico; [3]Univ. Iberoamericana, México, Mexico; [4]Univ. Iberoamericana, Mexico, Mexico; [5]Dept. of Psychology, Univ. Iberoamericana, Mexico City, Mexico

Abstract: Social interaction is known to have numerous neuroprotective, cognitive, and emotional benefits. Several elements are involved in social behavior, as well as underlying mechanisms and manifestations, including social hierarchy, sexual interaction, parenting and living in groups or isolated. Socialization has been linked to stimulating and enhancing cognitive function, mainly memory and learning processes, reducing the impact of stress, decreasing anxiety, and depression-like behaviors. However, the effects depend on several variables, such as age, sex, and duration of the condition. Our study aims to evaluate the cognitive and emotional effects of continuous or intermittent chronic social interaction. Method: We use sixteen 70 days old Wistar male rats that were divided into two groups according with their treatment: the continuous social interaction group (housing together), and the intermittent social interaction group (2 hrs per week). The condition last for 120 days (approx. 4 months old). After this, the evaluation started. We evaluate recognition memory, navigation memory, aversive memory, and anxiety-like behavior. The results are being processed, currently there is no differences between groups. Conclusion: social interaction continuous or intermittent apparently have similar effects.

Disclosures: A. Tapia De Jesus: None. **J.A. Mata-Luévanos:** None. **M.I. Mata:** None. **O.C. Galicia:** None. **M.H. Buenrostro-jauregui:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.07

Topic: H.06. Social Cognition

Support: MSCA-IF 840562
Gatsby Charitable Foundation and Wellcome Trust Core Grant (090843/F/09/Z)
Wellcome Trust Principal Research Fellowship (Wt203020/z/16/z)

Title: Representation of Ethological Events by Basolateral Amygdala Neurons

Authors: *C. MAZUSKI, J. O'KEEFE;
Univ. Col. London, Univ. Col. London, London, United Kingdom

Abstract: Accurately interpreting and remembering ethologically-relevant events is crucial for the survival of an organism. To understand the role of the basolateral amygdala (BLA) during ethological events, we recorded from large populations of BLA neurons using Neuropixels probes in freely-moving rats during different events - including interaction with social conspecifics, moving toys or food. We identified two main groups of BLA neurons, those that responded globally to several events (panresponsive) and those that were highly tuned to a single type of events (event-specific). These two classes of BLA neurons differed in anatomical spread, response onset and neuronal connectivity. A portion of these neurons continued to fire for minutes after the end of the eliciting events, potentially acting as an active memory trace of the event. Future experiments will further probe the role of BLA neurons in memory.

Disclosures: C. Mazuski: None. **J. O'Keefe:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.08

Topic: H.06. Social Cognition

Support: CIHR MOP119298
CureGrin research foundation

Title: Effect of ketogenic diet and beta-hydroxybutyrate on socio-cognitive deficits in a mouse model of GRIN disorder

Authors: *Y. YAN, T. LIPINA, D. J. STEVENS, L. PEPERA, W. HORSFALL, A. SALAHPOUR, A. J. RAMSEY;
Pharmacol. and Toxicology, Univ. of Toronto, Toronto, ON, Canada

Abstract: NMDA receptors are encoded by *GRIN* genes *GRIN1*, *GRIN2A-D*, and *GRIN3A-B*. Rare *de novo* mutations in the *GRIN* genes cause GRIN disorder, a neurodevelopmental disorder with symptoms of profound intellectual disability, epilepsy, and visual cortical impairment. GRIN disorder can be modeled with $Grin1^{KD}$ mice, which have a 90% reduction in GluN1 expression leading to NMDAR hypofunction. Cognitive deficits, reduced social motivation, impaired sensorimotor gating and seizures has been reported in $Grin1^{KD}$ mouse, making it a

promising model for GRIN disorder. Bioenergetic deficits in *Grin1*^{KD} mouse have been reported, suggesting deficient glucose uptake to meet high energy demand. We asked whether ketone bodies could serve as an alternative energy source in *Grin1*^{KD} mice, particularly since a ketogenic diet is an effective therapy for childhood-onset epilepsy. An emerging hypothesis suggests that elevation in beta-hydroxybutyrate (BHB) is key to the therapeutic effect of a ketogenic diet. BHB can be consumed as a supplement in water, making it a preferred substitute. We hypothesized that both a ketogenic diet and BHB can improve the behavioural consequences in *Grin1*^{KD} mice. Long-term exposure to BHB-supplemented water (NPN 80085843; Natural Health Product, USA; 6mg/ml; from 3 to 14 weeks of age) improved the hyperlocomotion and sensorimotor gating activity in *Grin1*^{KD} mice. However, deficits of social behaviour and spatial working memory (assessed by Y-maze) in *Grin1*^{KD} mice were not improved by BHB. We also tested the effect of a ketogenic diet (TD.07797, Envigo Canada; from 5 to 14 weeks of age) on behavioural impairments of *Grin1*^{KD} mice. Ketogenic diet improved hyperlocomotion but showed minimal effect on social behaviour and spatial working memory function in *Grin1*^{KD} mice. Our results suggested modest effect of BHB and ketogenic diet on socio-cognitive function in a mouse model of GRIN disorder. Ketogenic diet is not superior to BHB. In fact, it showed unwanted side effects including inadequate weight gain. Our study supports BHB as a preferred substitute over a ketogenic diet because BHB produces comparable beneficial effects with fewer adverse effects. We are currently investigating myelination morphology in *Grin1*^{KD} mice which is a potential target improved by both treatment interventions.

Disclosures: Y. Yan: None. **T. Lipina:** None. **D.J. Stevens:** None. **L. Pepera:** None. **W. Horsfall:** None. **A. Salahpour:** None. **A.J. Ramsey:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.09

Topic: H.06. Social Cognition

Title: Effects of time-dependent interpersonal cues on rat social dynamics

Authors: *S. C. PICKARD[1], J. R. HINMAN[2];
[1]Carle Illinois Col. of Med., [2]Dept. of Psychology, Univ. of Illinois Urbana-Champaign, Champaign, IL

Abstract: Conspecifics carry great influence over an animal's behavior. The location, actions and relative social status of conspecifics must constantly be tracked and integrated with the individual's own internal state. The complexity of the world and the behavioral affordances available to an individual will influence how it behaves. Rodent behavioral assays for social interaction, such as the three-chamber test, drastically restrict the behavioral affordances of the animal in order to focus on specific operationally defined behaviors that are easily analyzed. Such approaches eclipse the second-by-second decision making that is found in naturalistic

behavior that animals express in the wild - and in the laboratory when given the opportunity. This work elucidates how individual rats interact and engage with one another in a large open field environment over time, where the evolution of social hierarchy development and inter-personal influence is quantified. Time-dependent modeling is then used to describe how individuals influence the behavior of conspecifics and to assess if increased influence is correlated to specific behavioral phenotypes.
To this end, we collected data from each individual animal (N = 16 males, N = 16 females) by themselves in an open field (1m x 1m), on a plus-maze and in the three chamber sociability test in order to obtain a general phenotypic description of each animal's behavior as it relates to anxiety and social interest. We then introduced groups of four rats of the same sex (N = 4 male groups, N = 4 female groups) into an open field (1m x 1.75m) containing several enrichment objects for four hours a day for ten days. Video was collected and DeepLabCut was used to extract kinematic data on each individual over time. Results showed a large distribution in how time was allocated to exploratory or to anxiety-typed behavior; some individuals generated behavior sequences that exhibited primarily edge seeking, while others spent considerable time traversing the open space. Next steps aim to correlate the level of anxiety expressed in both the isolation and group settings to the degree of influence an individual has on conspecifics. This will be done through time-dependent analysis aimed to perform weight assignments of behavioral cues of an individual. It is hypothesized that rats that exhibit more exploratory behavior will have greater influence on conspecifics. This work will give insight into the recursive feedback between individuals and the group; that is, how individuals contribute to the formation and evolution of a group dynamic while also assessing how the group dynamic induces change in the behavior of the individual.

Disclosures: S.C. Pickard: None. **J.R. Hinman:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.10

Topic: H.06. Social Cognition

Support: Swiss National Science Foundation P2BEP3_200212

Title: Probing observational learning of an abstract rule in the mouse

Authors: *L. T. OESCH, J. COUTO, A. K. CHURCHLAND;
Dept. of Neurobio., UCLA, Los Angeles, CA

Abstract: Observational learning occurs in many species. In humans, learning from another's actions represents a major source of information for updating one's beliefs about the environment. Recent efforts have started uncovering the neural correlates for observational fear learning and motor imitation in mice. However, current paradigms have two shortfalls. First,

current paradigms lack methods to ensure or measure the extent to which observer mice engage in observation of the demonstrator. These paradigms generally rely on either un-tracked observations in freely behaving animals or passive viewing of head-fixed animals. Second, existing paradigms in mice have probed fast learning processes of concrete stimulus-outcome associations, such as learning about fearful contexts or motor sequences. As a result, the ability to gradually discover abstract rules, a hallmark of higher cognitive function, is largely unstudied in mice. Here, we introduce a novel paradigm that surmounts these obstacles to investigate observational learning in freely behaving mice. We built an automated apparatus with separate compartments for observer and demonstrator mice. In one compartment, the demonstrator mouse judges auditory stimuli as high- or low-rate and reports its choice by interacting with a port on the left or the right side of the compartment, respectively. In the other compartment, the observer mouse hears the stimuli and watches the demonstrator's behavior. Taking advantage of the multisensory nature of our rate discrimination task, we first train future observer mice to be experts in visual rate discrimination, such that they are proficient in the task procedure and learn to trigger stimuli at the center port and then choose the rewarded side associated with the visual stimulus rate, but are naïve to the abstract auditory rate rule. We then teach these observer mice to synchronize self-initiated and -terminated observations with a demonstrator. This ensures that the observer remains highly engaged and visually attentive. Task observation can also be performed with a virtual demonstrator as a control. Observers paired with demonstrators initiate 91.8 ± 58.6 (mean ± std) trials per session, of which 51.4 ± 34.4 contain information about the task rule, while the rest are incompletely watched trials or demonstrations without choice. Observer mice reliably respond to visual cues that indicate the end of a trial observation (median reaction times of 159 ± 60 ms). These findings suggest that our behavioral paradigm can overcome earlier limitations and may provide a tightly controlled framework to study observational learning of an abstract rule in the mouse.

Disclosures: L.T. Oesch: None. **J. Couto:** None. **A.K. Churchland:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.11

Topic: H.06. Social Cognition

Support: NSERC

Title: Interplay between estrogens in the supraoptic nucleus and the oxytocin receptor in the medial amygdala on the rapid mediation of social recognition

Authors: *P. PALETTA, A. PALMATEER, E. CHOLERIS;
Psychology, Univ. of Guelph, Guelph, ON, Canada

Abstract: Social recognition (SR) is mediated by both estrogens and oxytocin (OT), as shown in studies in which knocking out the genes for the estrogen receptors, OT, or the OT receptor (OTR) impaired SR. Also, studies in which 17β-estradiol (E2), estrogen receptor agonists, or OT are administered facilitated SR. Since estrogens and OT are needed for SR, it has been hypothesized they may interact. A model has been suggested that estrogens will bind to their receptors in the paraventricular and/or the supraoptic nuclei (PVN and SON) of the hypothalamus, where the majority of OT production occurs, and facilitate the production and release of OT. The OT will then reach the medial amygdala (MeA) and bind to the OTR. The MeA also receives direct projections from the olfactory bulbs, so olfactory information about individuals that are encountered is also sent to the MeA. The model suggests that it is the OT facilitated by estrogens, binding to the OTR in the MeA that allows the incoming olfactory information to be used to recognize a familiar individual. We have previously shown evidence for this interplay through estrogens' rapid mechanisms by finding that E2 infused into the PVN of female mice rapidly facilitates SR and this facilitation can be blocked by simultaneously infusing a subeffective dose of an OTR antagonist (OTRA) into the MeA. These findings showed support for the proposed interaction between estrogens and OT as well as the PVN and MeA in the rapid facilitation of SR. Currently, we are investigating whether this interaction also occurs with estrogens in the SON. We have shown that E2 infused into the SON can rapidly facilitate SR, and the current experiment is investigating whether the same subeffective dose of the OTRA, a dose that doesn't block SR by itself, can block this facilitation by E2 in the SON. Following the infusions, the mice are run through the "difficult" rapid SR paradigm. In this paradigm 2 ovariectomized female stimulus mice are presented to the experimental mouse in 2 sample phases, followed by a test phase where 2 stimulus mice are presented again, however one is a novel mouse. Since mice prefer novelty, if they investigate the novel mouse more, it suggests they recognize the other stimulus mouse. The paradigm takes place within 40 minutes to test the rapid mechanisms of estrogens and was designed to be difficult, meaning the vehicle group will not show SR and therefore facilitating effects of the treatments can be observed. If it is found that the OTRA in the MeA blocks the rapid facilitation of SR by E2 in the SON, it would show support for the idea that estrogens in the SON also interact with OT to rapidly facilitate SR in the MeA, similar to the PVN results.

Disclosures: **P. Paletta:** None. **A. Palmateer:** None. **E. Choleris:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.12

Topic: H.06. Social Cognition

Support: Grant no POIR.04.04.00-00-14DE/18-00 from the Foundation for Polish Science carried out within the Team-Net program co-financed by the European Union under the European Regional Development Fund
Grant OPUS 2019/35/B/NZ7/03477 from the National Science Centre Poland

Statutory funds of the Maj Institute of Pharmacology of the Polish Academy of Sciences

Title: Modeling social effects in mice learning in intelligent cages

Authors: *B. JURA[1], M. LENARCZYK[1], Z. HARDA[2], M. ZIEMIAŃSKA[2], &. SZUMIEC[2], J. RODRIGUEZ PARKITNA[2], D. K. WÓJCIK[1,3];
[1]Fac. of Mgmt. and Social Communication, Jagiellonian Univ., Kraków, Poland; [2]Dept. of Mol. Neuropharm., Maj Inst. of Pharmacol. of the Polish Acad. of Sci., Kraków, Poland; [3]Lab. of Neuroinformatics, Nencki Inst. of Exptl. Biol. of the Polish Acad. of Sci., Warszawa, Poland

Abstract: The ability to learn by observing others allows individuals to learn about the outcomes of actions without directly experiencing their consequences, and may also allow for the formation of behaviors that benefit the group as a whole. Here, we study behavior and learning of mice in the Intellicage system, which automatically tracks the access to drinking bottles by group-housed animals. We designed a paradigm where rewards (access to sweetened water) are offered depending on an arbitrary assignment of an animal to one of two groups, either "majority" or "minority". The two groups were assigned different locations with reward availability, changing in consecutive phases of the experiment. To assess the effect of following others in choosing between locations, we developed a novel model-based approach to analysis of the Intellicage data. We combine point-process description of mice activity with reinforcement learning of the reward, and validate this approach with simulated data. Using data from the Intellicage experiments with different expected social effects and different reward protocols (rewards in the same/different corners; reversal learning tasks) we show a range of analytical approaches and present their effectiveness. We show that the proposed stochastic models capture animals' behavior well and can be used to estimate the social effects of learning in different situations adding extra information for quantitative description of behavior in mice of different strains, age, treatment, or genetic modifications. In particular, we compared standard reinforcement learning models (like Q-learning and its extensions) where animal is assumed to learn based solely on its own experience, with models including also the social effects and learning of value through the observation of others, modeling strength and time span of the social influence. We show that including the social effects in this experimental context allows to account better for the actions taken by animals. Corresponding generative models, through simulations, can be used for prediction of mice behavior, for example at early planning stages of experiments in which Intellicages are employed. Our approach can easily generalize to other type of intelligent cages.

Disclosures: B. Jura: None. **M. Lenarczyk:** None. **Z. Harda:** None. **M. ZiemiaŃska:** None. **&. Szumiec:** None. **J. Rodriguez Parkitna:** None. **D.K. Wójcik:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.13

Topic: H.06. Social Cognition

Support: NSERC Grant

Title: The role of Estrogen Receptor alpha and beta in the BNST in social recognition and aggression in male mice

Authors: *D. ASPESI[1], S. MATTA[2], S. SETHURAMAN[2], T. MANNING[2], E. CHOLERIS[1]; [1]Psychology, [2]Biomed. Sci., Univ. of Guelph, Guelph, ON, Canada

Abstract: Social recognition (SR) allows to identify previously investigated conspecifics and to emit appropriate pro-social or -aggressive behaviours. Both androgens and estrogens in the bed nucleus of the stria terminalis (BNST) can rapidly facilitate social recognition and increase the emission of dominant behaviour in male mice. In particular, testosterone (T) and 17β-estradiol (E2) regulate SR by rapidly interacting with arginine-vasopressin (AVP) in the BNST and inducing increased AVP availability in the lateral septum (LS). The involvement of E2 in this modulation suggests that estrogen receptors (ERs) may be the main regulators of SR and dominant behaviours. Although the rapid interplay between sex steroids and AVP in social behaviour is known, the specific ERs responsible for the behavioural outcomes are yet to be established. To elucidate the role of ERα and ERß in SR and aggression, adult castrated (CX) male mice were intracerebrally infused with one of four different doses of the specific ERα agonist PPT (50, 100, 150, 200nM of PPT in 0.5µl of aCSF/alcohol) or with one of three different doses of the specific ERß agonist DPN (50, 100, 150nM of DPN in 0.5µl of aCSF/alcohol). Mice were then exposed to a 'difficult' SR paradigm, in which CX mice are impaired, consisting of two 5-min sample phases with 2 familiar conspecifics, and one test phase with the choice between a familiar or a novel mouse. To assess aggression, mice were tested in a resident-intruder (RI) paradigm at either 35- or 120-min post-infusion to evaluate rapid and long-lasting effects. Results revealed that infusing PPT or DPN in the BNST rapidly facilitated SR, with treated CX mice spending more time investigating a novel over a familiar CX mouse, with an inverted U-shaped dose-response. In addition, PPT increased the dominance score at 35-min, but not at 120-min. The expected results on DPN will reveal a similar behavioural outcome to that of PPT with a facilitation of SR and increased dominance. These results will confirm the ERs mediating E2 rapid facilitation of SR and aggression in male mice, by probably rapidly interacting with the AVP system in the BNST. Next step of this project will be to investigate the role of the G-coupled protein estrogen receptor 1 (GPER1) and androgen receptor (AR) in the BNST in the modulation of social recognition and aggression. Elucidating the AVP-sex hormones interplay may lead to new therapeutic approaches for psychopathologies of social behavior with marked sex differences.

Disclosures: D. Aspesi: None. **S. Matta:** None. **S. Sethuraman:** None. **T. Manning:** None. **E. Choleris:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.14

Title: WITHDRAWN

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.15

Topic: H.06. Social Cognition

Support: NWO VIDI/917.18.380,2018/ZonMw

Title: Identifying environment-dependent behavioral domains predictive of autism-like phenotype

Authors: *L. WAHL, A. HASSETT, A. KARIM, M. VAN DER DOE, I. SMAL, C. VAN DER ZALM, A. BADURA;
Erasmus Med. Ctr. Rotterdam, Rotterdam, Netherlands

Abstract: Genetic and environmental factors alike can contribute to the development of Autism Spectrum Disorder (ASD), making the studies on the underlying neural substrates highly complex. In our study, we aimed to develop a "behavioral fingerprint" for the *Shank2*$^{-/-}$ ASD mouse model raised in standard and enriched environments. To this end we employed a wide range of both discrete, standardized tests such as: elevated-plus-maze, social chamber, marble burying test, Y-maze assay, and open field test; as well as two multi-parametric behavioral assays: the Live Mouse Tracker (described in de Chaumont et al., 2019) and Motion Sequencing (MoSeq) developed by the Datta Lab. Our aim was to integrate our high dimensional data into one single platform, to classify differences in experimental groups along dimensions with maximum consistency and discriminative power. We have found that some behavioral phenotypes of *Shank2*$^{-/-}$ mice, such as impaired burying behavior and behavioral flexibility, as well as hyperactivity and decreased anxiety-like behavior, were consistently altered whereas other features were environment-dependent. Groups raised in enriched housing showed a distinct effect on the behavioral phenotype in both the discrete and multi-parametric assays. *Shank2*$^{-/-}$ mice showed a unique phenotypic profile based on housing conditions using both the Live Mouse tracker and the MoSeq analysis. The described identity domains captured variability in *Shank2*$^{-/-}$ mice reared in different housing conditions, suggesting the feasibility of reducing behavioral test batteries to multi-parametric assays. Together, our results provide a "behavioral fingerprint" of *Shank2*$^{-/-}$ mice, which is shown to be altered by different housing conditions during development.

Disclosures: L. Wahl: None. **A. Hassett:** None. **A. Karim:** None. **M. van der Doe:** None. **I. Smal:** None. **C. van der Zalm:** None. **A. Badura:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.16

Topic: H.06. Social Cognition

Support: Intramural Research Program of the NIH
Center on Compulsive Behaviors to R.O.G.

Title: Role of GABA co-transmission from cholinergic neurons in brain function

Authors: *R. O. GORAL[1,2], K. M. HARPER[3], B. J. BERNSTEIN[1], S. A. FRY[1], P. W. LAMB[1], J. D. CUSHMAN[1], S. S. MOY[3], J. L. YAKEL[1];
[1]Neurobio. Lab., Natl. Inst. of Envrn. Hlth. Sci., Durham, NC; [2]Ctr. on Compulsive Behaviors, NIH, Bethesda, MD; [3]Mouse Behavioral Phenotyping Lab., Univ. of North Carolina, Chapel Hill, NC

Abstract: Several acetylcholine (ACh) neuron populations co-transmit the neurotransmitter gamma-aminobutyrate (GABA). ACh/GABA co-transmission occurs at synapses in the hippocampus, striatum, and medial prefrontal cortex (mPFC). Altered ACh signaling or ACh neuron function through disease or substance exposure has been found in or increased the risk for many brain disorders. While ACh transmission is known to have a substantial impact on higher brain functions such as learning and memory, the role of co-transmitted GABA from ACh neurons in brain function remains unknown. The overarching goal of this study was to assess how a systemic loss of GABA co-transmission from ACh neurons affected the behavioral performance of mice. In the absence of the vesicular GABA transporter (vGAT), neurons are unable to pump GABA into synaptic vesicles and, therefore, unable to release GABA at their synapses. All ACh neurons express choline acetyl transferase (ChAT), so we selectively knocked out vGAT in ACh neurons by crossing vGAT-flox with ChAT-Cre mice. In a comprehensive series of standardized behavioral assays, we compared Cre-negative control with Cre-positive vGAT knock-out mice of either sex. Loss of GABA co-transmission from ACh neurons did not disrupt the animal's motor skills and sensation, sociability, or contextual learning. However, in the absence of GABA co-transmission, we found significant alterations in memory, problem-solving and other higher brain functions independent of sex. In addition, male mice showed obsessive compulsive-like behaviors previously described in the literature for models of drug abuse. Taken together, the loss of GABA co-transmission leads to deficits in higher brain functions and behaviors which are associated with hippocampus, striatum, as well as the mPFC. Therefore, we conclude that ACh/GABA co-transmission is required to modulate neural circuitry involved in the affected behaviors. Future studies will investigate how ACh/GABA co-transmission shapes neural circuit function in specific brain regions and whether ACh/GABA co-transmission is disrupted in brain disorders.

Disclosures: R.O. Goral: None. **K.M. Harper:** None. **B.J. Bernstein:** None. **S.A. Fry:** None. **P.W. Lamb:** None. **J.D. Cushman:** None. **S.S. Moy:** None. **J.L. Yakel:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.17

Topic: H.06. Social Cognition

Support: Scholarship to JP 1105815 from CONACyT (Mexico)
CONACyT INFR-281265

Title: Problem solving in wistar rats using a conspecific as goal

Authors: *J. C. PARRA-CRUZ[1], A. LONGÁN[1], P. S. DILLON SOARES-FILHO[2], D. M. CORTÉS-PATIÑO[3], J. BURITICÁ[1];
[1]Lab. of Cognition and Comparative Learning, CEIC, Univ. of Guadalajara, Guadalajara, Mexico; [2]Lab. of Complex Behavioral Processes: aversive contingencies and ontogenesis of creativity, Univ. Federal of Pará, Belém, Brazil; [3]Lab. of Neurosci., Natl. Univ. of Colombia, Bogotá, Colombia

Abstract: Innovation is related to the generation of new behavior and refers both to a novel behavioral pattern and to the process by which an animal generates or modifies a behavioral pattern. Innovation helps animals to cope with environmental challenges such as exploring environments, foraging food or dealing with predators. In the laboratory, we use the problem-solving paradigm to study innovative behavior, in which the animals solve problems to achieve a goal; frequently, researches use food as the goal of the problem-solving task, however for a better understanding of innovation, it is essential to explore the rats' problem-solving behavior using unconventional goals. We explored in wistar rats the effects of a conspecific as goal in a problem-solving task. The problem-solving test required the animals to climb a ramp, to cross a bridge and to open a container where the conspecific was restricted (Bartal et al., 2011). The research used 48 males naïve wistar rats assembled into three groups of order of exposure to the test: the trapped rat first group, the empty restrainer first group and the control group (empty restrainer). The order of exposure to the test had two moments of four sessions; depending on the order of exposure, the animals encountered the test first in presence or in absence of the conspecific and then at the second moment they experienced the task with the remaining condition. Subjects were ~63 days old and they were housed (2 rats/cage) under standard colony room conditions. The procedure had five phases: time-out test, open field test-habituation to the arena and movement restrainer, trapped rat test, training of behavioral repertoires and the problem-solving test. We found that rats learned to open the restrainer door without training, they climbed the ramp and crossed the bridge after training, and finally animals solved the task and liberated the conspecific trapped into the movement restrainer. Preliminary analysis suggests that order of exposure to the problem may affect the latency and the frequency of the solution of the problem. These results show that rats solve problems in the presence of a goal different to food, also the results point out that rats may direct a complex action such as the problem-solving behavior toward a conspecific.

Disclosures: **J.C. Parra-Cruz:** None. **A. Longán:** None. **P.S. Dillon Soares-Filho:** None. **D.M. Cortés-Patiño:** None. **J. Buriticá:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.18

Topic: H.06. Social Cognition

Title: Frontopolar mechanisms for driving social and economic decisions in primate groups

Authors: E. P. MASTROBATTISTA[1], A. J. WANG[1], Z. WILLIAMS[1], **R. BÁEZ-MENDOZA**[2];
[1]MGH, Boston, MA; [2]German Primate Ctr., Göttingen, Germany

Abstract: Primate group behavior allows individuals to build affiliations and benefit from the reciprocation with others but also poses the unique challenge of tracking others' behavior across multiple distinct interactions. These interactions can often also be highly dynamical and change rapidly based on the reputation or wealth distribution of others. The single-cellular mechanisms that precisely underlie these decisions or that drive the social-economic behavior of groups, however, remain poorly understood. Here, we obtained multiple-neuronal recordings from the dorsomedial prefrontal cortex (dmPFC) and frontopolar (FP) cortex of Rhesus macaques as they performed a structured reciprocity-based social task. In this task three individuals interacted with each other over multiple rounds by offering each other reward and which could allow us to dissociate computations associated with interactive behavior, social preference, and group dynamics. Behaviorally, we find that the monkeys demonstrated a strategic preference for other individuals and favored rewarding those who reciprocated. The rate at which individuals reciprocated within and across sessions was reflected in distinct levels of reputation. At the single-cellular level, we have previously shown that different subpopulations of dmPFC neurons tracked the identity of the current actor and reward recipient. Here, we show that the activity of a subpopulation of FP neurons correlated with the current actor's own reputation for reciprocity. These findings reveal neurons in the primate prefrontal cortex that encode information about specific individuals within social groups and which could help optimize economic benefit during interactive group dynamics. Future work in Macaques and Marmosets will expand on how social ties impinge on these economic behaviors and their neuronal mechanisms.

Disclosures: **E.P. Mastrobattista:** None. **A.J. Wang:** None. **Z. Williams:** None. **R. Báez-Mendoza:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.19

Topic: H.06. Social Cognition

Support: NIH Grant AA013983

Title: To Fight or not to Fight: Activation of the mPFC during decision to engage in aggressive behavior after alcohol consumption in a novel murine model

Authors: *K. MICZEK[1], M. Z. LEONARD[1], N. AKDILEK[1], V. M. FERREIRA[2], L. MARINELLI[1], H. E. COVINGTON, III[1];
[1]Tufts Univ., Medford, MA; [2]Univ. Federal De Santa Catarina, Florianopolis SC 88.015-420, Brazil

Abstract: Alcohol consumption is a common antecedent of aggressive behavior. The effects of alcohol on the decision to engage in aggression in preference over pro-social interaction are hypothesized to arise from augmented function within the medial prefrontal cortex (mPFC). *Objective:* In a newly developed procedure we studied social decision-making in male C57BL/6J mice based on preferentially seeking access to either sociosexual interactions with a female partner or the opportunity to attack an intruder male. While deciding to engage in aggressive vs. sociosexual behavior, corresponding neural activation was assessed via c-Fos immunoreactivity in cortical, amygdaloid and tegmental regions of interest. A further objective was to investigate how self-administered alcohol impacted social choice. *Methods:* During repeated confrontations with an intruder male in their home cage, experimental B6 mice began to engage in species-specific sequence of pursuit, threat and attack behavior within < 2min. Mice were then conditioned to respond at one of two separate illuminated operanda in an experimental chamber (octagon) attached to their home cage; completion of 10 responses (Fixed Ratio 10; FR10) was reinforced by access to either a female or a male intruder which were presented in the resident's home cage. Brains were harvested following choice between the concurrently available aggressive and sociosexual options and processed for c-Fos immunoreactivity across 10 separate brain regions. In two separate groups, mice were trained to rapidly self-administer ethanol prior to social choice in order to examine the effects of alcohol on social choice, sociosexual, aggressive acts and postures, and corresponding c-Fos activity in the mPFC and limbic regions. *Results and Discussion:* Eight out of 57 mice consistently choose to engage in aggressive behavior in preference to sociosexual contact with a female when each outcome was concurrently available. Self-administered alcohol (alcohol experiment 1: 1.2 + 0.02g/kg, alcohol experiment 2: 0.5, 1.0, 1.5 and 1.8 g/kg) increased responding for the aggressive option in mice that previously opted exclusively for access to sociosexual interactions with the female. When choosing the aggressive, but not the sociosexual option, the prelimbic area of the PFC revealed increased c-Fos activity, guiding future detailed inquiry into the neural mechanisms for aggressive choice**.**

Disclosures: K. Miczek: None. **M.Z. Leonard:** None. **N. Akdilek:** None. **V.M. Ferreira:** None. **L. Marinelli:** None. **H.E. Covington:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.20

Topic: H.06. Social Cognition

Support: NIH Grant R01MH127423
SUNY Upstate Institutional Funds
NHMRC (Australia) Grant APP1145770
SAMHRI Institutional Funds
Flinders University bridge Grant

Title: Characterization of a PHF21B knockdown mouse model

Authors: *J. LICINIO, E. CHIN, Q. MA, H. RUAN, C. CHIN, M.-L. WONG;
Psychiatry, SUNY Upstate Med. Univ., Syracuse, NY

Abstract: The *PHF21B* gene belongs to the PHD finger protein family and encodes a histone modification reader. Despite being widely expressed in the brain, its functional role in the adult brain is unknown. We have recently reported that chronic stress modulates its hippocampal expression. Rare variants of this gene are associated with major depressive disorder risk and this gene controls an epigenetic program for neural stem cells during murine cortical development. Hence, we produced a *Phf21b* knockdown mouse model to characterize behavior and the functional role of this gene.We generated a *Phf21b* knockdown mouse model lacking exon 4 using CRISPR/Cas9. We used a battery of behavioral assays to ascertain the phenotypes of PHF21B deficiency using male and female wildtype (WT) and homozygous (Phf21bΔ4/Δ4) littermates. The functional role of this gene was assessed using analytical tools, including histology, immunoblotting, qRT-PCR, RNA sequencing, and electrophysiology approaches.Mice with PHF21B deficiency exhibit normal locomotor activity in the open field test compared to WT mice. Compared to WT mice, Phf21bΔ4/Δ4 mice did not display anxiety-like behaviors, behavioral despair, anhedonia, or spatial memory impairments. These mice had increased preference for social novelty in the three-chamber social test ($P<0.01$) that suggested social memory impairment, which was confirmed by the 5-trial social memory test ($P<0.05$). Brain analytical techniques revealed that PHF21B deficiency resulted in thinner cortices ($P<0.001$), reduced neurogenesis (DCX + cells; $P<0.0001$) and astrocyte numbers (GFAP + cells; $P<0.0001$). Phf21bΔ4/Δ4 mice also had decreased hippocampal synaptic protein expression (PSD95 + puncta numbers; $P<0.001$, and GLUR1 + puncta numbers; $P<0.0001$), glutamatergic neurotransmission (decreased EPSC amplitude in CA1; $P<0.001$), and GluN2B/*Grin2b* levels ($P<0.05$ and $P<0.01$, respectively). RNA seq analyses revealed that PHF21B modulates genes involved in neurotransmission in the hippocampus. PHF21B regulates transcription by interacting with H3K9ac, H3K9me2, and CREB. It also binds to H3K36me3.In summary, PHF21B has a role in social memory, its deficiency impairs social memory. It modulates genes involved in neurotransmission in the hippocampus by regulating transcription through H3K9ac, H3K9me2 and CREB. We described novel interactions of PHF21B and H3K36me3 and CREB.

Disclosures: **J. Licinio:** Other; Springer Nature. **E. Chin:** None. **Q. Ma:** None. **H. Ruan:** None. **C. Chin:** None. **M. Wong:** Other; eLife Sciences Publications, Ltd.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.21

Topic: H.06. Social Cognition

Support: Harris Research Endowment
Drake Undergraduate Science Collaborative Institute

Title: Relationship between barbering and dominance hierarchy in B6129SF2/J male mice

Authors: A. K. KLEIN, K. R. REIMAN, N. J. ANDERT, *C. C. WRENN;
Drake Univ., Des Moines, IA

Abstract: Mice that exist in social groups will naturally form dominance hierarchies which can have a significant impact on animals' health, physiology, and general behavior. An observed behavior hypothesized to influence and maintain these hierarchies is barbering, which is when one mouse plucks, trims, or removes the fur or whiskers of another mouse. We hypothesized that barbering extent would reflect social rank. This study started with one observer taking notes and photographs of aged B6129SF2/J male mice and their fur loss. Barbering patterns ranged from completely unbarbered to some mice exhibiting significant fur loss. Based on these observations, barbering extent of each mouse was scored from 0-5, and cage mates were ranked as a predicted hierarchy in which social rank decreased as barbering extent increased. To determine the actual dominance hierarchy, cage mates were run for 8 bouts in the tube test in a round robin fashion. The wins and losses for each bout were recorded, and David's Score was used to determine the hierarchy for each cage. This hierarchy was then compared to our barbering-based, predicted hierarchy. We identified a pattern in which the most barbered mice were the most dominant, counter to our prediction and studies of other strains. In 7 cages out of 10, our predicted most dominant mouse was found to be the least dominant based on David's Score and vice versa. Results from this study will guide future experiments, which will be conducted with a larger population of younger mice to further explore the validity of these preliminary findings.

Disclosures: **A.K. Klein:** None. **K.R. Reiman:** None. **N.J. Andert:** None. **C.C. Wrenn:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.22

Topic: H.06. Social Cognition

Title: The impact of the LC-NAergic system in controlling empathic fear.

Authors: *J.-H. KIM[1], M. KIM[1], H.-S. SHIN[1,2];
[1]Ctr. for Cognition and Sociality, Inst. for Basic Sci. (IBS), Daejeon, Korea, Republic of; [2]SL Bigen, Incheon, Korea, Republic of

Abstract: Observational fear is a useful behavioral paradigm for assessing affective empathy in rodents. The anterior cingulate cortex (ACC) is involved in empathic responses to pain or fear in a conspecific animal. The ACC receives various neuromodulatory inputs from subcortical structures, including the locus coeruleus-noradrenaline (LC-NA) neurons. The LC is the main origin of noradrenergic neurons, and NA is a key neuromodulator playing critical roles in various higher brain functions in the central nervous system (CNS) including arousal, attention, cognition, and memory. However, the relevance of the LC-NA system to empathic behavior is not known. Here, we show that the LC-NA system is crucial for maintaining freezing behavior during observational fear. We found increasing LC-NA neurons' terminal activity during observational fear in the ACC. Additionally, the optogenetic inhibition of LC-NA input in the ACC affected only the maintenance of observational fear responses but not the initial response in observer mice, indicating that the LC-NA system in the ACC controls the degree of socially transmitted fear. By monosynaptic rabies tracing we identified the bed nucleus of the stria terminalis (BNST) as the upstream input to the LC- NAergic projections to the ACC. Notably, we found that there is no anatomical connection between the ACC and the BNST. Abolishing the LC-projecting BNST neurons elicited a failure of contiguous freezing response during observational fear, suggesting that the BNST is involved in controlling empathy through the LC-NA system. This work expands our understanding of the neural circuits involved in observational fear beyond the amygdala and the ACC, revealing the role of a novel neural pathway centered on the LC-NAergic system in affective empathy.

Disclosures: **J. Kim:** None. **M. Kim:** None. **H. Shin:** None.

Poster

569. Social Cognition: Animal Behavior I

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 569.23

Topic: H.06. Social Cognition

Support: NIH R01HD059852
NIH R01NS091390
NIH U01NS123130
Autism Science Foundation

Title: Insular-prefrontal circuit driving compassionate social behavior

Authors: *S. W. LI, P. B. GABRIELI, M. SUZUKI, O. ZELIGER, J. DEMAREE, R. CAUCHON, N. OCCIDENTAL, Z. WILLIAMS;
Massachusetts Gen. Hosp., Boston, MA

Abstract: Compassionate behavior, or the ability to help others in need, is a cornerstone of prosocial interaction in mammals and humans. To benefit others, it is necessary for individuals not only to perceive the internal states or emotions of others but also to take appropriate actions. For example, one must identify the distress of another and perform specific actions that will relieve their discomfort. Yet, how mammalian neurons precisely link social-specific information with such complex adaptive behavior has been a major challenge to understand. Here, we developed a place-preference assay that allowed mice to directly control in real-time the experience of a nearby conspecific while also allowing the animal's own actions to be dissociated from the other's identity and experiences. Behaviorally, we found that wild-type male mice consistently chose to reduce the aversive experience of familiar but not unfamiliar partners, actions that were not observed when visual and olfactory cues were blocked. Their decisions were uncorrelated with hierarchical rank, baseline sociability or vocalizations; together suggesting that the mice displayed consistent compassionate behavior towards familiar partners. By recording from neurons in the anterior insular (AI), we identified cells that encoded task relevant information, including the social identity of the animal's partners and their specific experience. Cells in the dorsal anterior cingulate cortex (dACC), by contrast, preferentially encoded information about the act of helping their partners, displaying changes in activity prior to making their decisions. Further, whereas information about the experience of others could be predominantly decoded from AI activity, information about the animal's prosocial actions could be predominantly decoded from dACC activity; demonstrating a partitioning of information within the insular-prefrontal circuit. Finally, chemogenetic excitation of AI-to-dACC projectors but not dACC-to-AI projections increased helping behavior when the animals were paired with unfamiliar partners suggesting that the AI transmitted social-specific information to the dACC. Inhibitions of both dACC to AI as well as AI to dACC projectors, on the other hand, decreased helping behavior with familiar partners - further suggesting that the dACC controlled compassionate decisions based on information from the AI. Taken together, these findings identify a putative insular-prefrontal circuit for driving compassionate behavior and a mechanism that could allow insular neurons to instruct social-specific actions through prefrontal control.

Disclosures: S.W. Li: None. **P.B. Gabrieli:** None. **M. Suzuki:** None. **O. Zeliger:** None. **J. Demaree:** None. **R. Cauchon:** None. **N. Occidental:** None. **Z. Williams:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.01

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH R01 MH119384-01

Title: Neurophysiology of the prefrontal-striatal circuitry during extradimensional Set-shifting in rats

Authors: *Z. SONG, A. ALPERS, K. WARNER, E. D. RIJN, K. SINGH, E. SACHSE, E. HOSKINS, A. S. WIDGE;
Univ. of Minnesota, Minneapolis, MN

Abstract: The balance between cognitive flexibility and rigidity is impaired in a wide range of psychiatric disorders such as depression, substance abuse, and autism. Extradimensional set shifting, a commonly used neuropsychological test that evaluates the ability to shift strategies based on continuously changing environments, has proven to be clinically relevant and has been successfully adapted for research in animal models. The present project aimed to examine electrophysiological activity in the prefrontal cortex (PFC) and the striatum, and the connectivity between these two regions, while rats performed an extra-dimensional Set-shift task. It is hypothesized that the PFC and different sub-zones of striatum may have parallel circuits that regulate the process of decision-making through biasing the balance between cognitive flexibility and rigidity. Specifically, flexible behaviors may require involvement of dorsal PFC and its connection to dorsal striatum, and rigid behaviors may require ventral PFC and its connection to ventromedial striatum. We implanted three rats with dual silicon recording probes, covering both the dorsal and ventral sub-regions of the medial PFC and the striatum. Electrophysiological data were obtained while rats shifted between a cue-driven Light rule and a spatial Side rule to obtain food rewards. The task required five consecutive correct responses to proceed to the next rule and one test session had a total of eight rules (alternating between light and side rules). Local field potential and spike data were analyzed and compared while rats were in different behavioral states including task time versus interval time, and correct trials versus incorrect trials. The dorsal PFC to dorsal striatum pathway had an increased coherence in frequencies between 8-14Hz during task versus interval and during correct trials versus incorrect trials. In contrast, the ventral PFC to the ventral striatum pathway did not show such changes. Analyses on spike data revealed that approximately 100 single units could be isolated from each probe, 10-30% of which showed task-related changes in firing rates. Spike-field coupling analyses showed that spike timing of at least half of the units were less locked to theta/alpha oscillations than to gamma oscillations. Taken together, the present study suggests that a dorsal PFC to dorsomedial striatum pathway is important in the regulation of cognitive flexibility that is required in the Set-shift task. Oscillations in theta/alpha bands might be involved.

Disclosures: Z. Song: None. **A. Alpers:** None. **K. Warner:** None. **E.D. Rijn:** None. **K. Singh:** None. **E. Sachse:** None. **E. Hoskins:** None. **A.S. Widge:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.02

Topic: I.08. Methods to Modulate Neural Activity

Support: NIMH grant MH119384

Title: Pre-frontal Cortex and Amygdala Interactions during an approach and avoidance task

Authors: *D. J. TITUS, C. LI, K. DRAHOS, S. J. OLSON, S. S. NAGRALE, A. S. WIDGE; Univ. of Minnesota, Univ. of Minnesota, Minneapolis, MN

Abstract: The ability to discriminate between threatening and non-threatening environmental cues and execute an appropriate behavioral response is crucial for survival and for maintaining a healthy balance between avoidance and reward-seeking. The infralimbic (IL), and prelimbic (PL) medial prefrontal cortices and basolateral nucleus of the amygdala (BLA), have been implicated in reward-seeking and fear-related responses. Their homologues often have altered outputs in psychiatric disorders like anxiety and Post Traumatic Stress Disorder (PTSD). Despite abundant evidence, little is known about the circuit-level connections between these regions that coordinate an appropriate behavioral response. The communication between brain regions may depend on synchronized neuronal oscillations. Theta band oscillations (4-8 Hz) are proposed to be responsible for long-range communications, specifically in the medial prefrontal cortex (mPFC) - BLA circuitry. Changes in theta synchrony between mPFC and BLA are associated with psychiatric disorders. Therefore, the aim of this study is to investigate the interaction between these medial prefrontal structures and the amygdala during platform-mediated avoidance (PMA). PMA is a behavioral paradigm that resembles real-life avoidance scenarios and captures approach-avoidance conflict behavior. In this paradigm, rats learn to avoid a tone-signaled foot shock by stepping onto a platform, with the cost of avoidance being the loss of access to food. Adult Long-Evans rats between 5 and 7 months of age received electrode bundles or tetrode implants in the IL and PL of the mPFC and BLA regions after training in the PMA task. We will analyze the correlation between the oscillatory synchrony in the theta frequency band between IL to BLA, and PL to BLA, and behavioral responses such as time spent in different zones (platform zone, reward zone, and out of platform zone) or bar press rates during PMA high conflict performances. This study is an exploratory study that may provide a neurobehavioral correlation during motivated decision-making. The outcome of this study could allow us to find specific targets that are altered in anxiety disorders like PTSD and serve as a basis to develop simulation-based therapeutic interventions for these disorders in humans. Supported by NIMH grant MH119384.

Disclosures: D.J. Titus: None. **C. Li:** None. **K. Drahos:** None. **S.J. Olson:** None. **S.S. Nagrale:** None. **A.S. Widge:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.03

Topic: I.08. Methods to Modulate Neural Activity

Support: R01 Circuit and Cognitive Mechanisms of DBS

Title: Dbs disrupts reinforcement-based decision making in rats

Authors: *F. IACOBUCCI, D. COOPER, E. DASTIN-VAN RJIN, A. WIDGE;
Univ. of Minnesota, Minneapolis, MN

Abstract: Deep brain stimulation (DBS) has been used to alleviate symptoms in treatment-resistant neuropsychiatric disorders, including obsessive compulsive disorder (OCD) and major depressive disorder (MDD). Despite DBS' therapeutic success, its mechanism of action is not well understood. MDD and OCD can be characterized by impairments in cognitive control—the ability to selectively organize, plan, and schedule mental operations in different environments. Cognitive control depends on distinct subregions of the prefrontal cortex which project to and receive projections from the striatum. Further investigating cortico-striato-thalamo-cortical (CSTC) circuits, which are often implicated in these disorders, may provide insight into better understanding how DBS can aid in the treatment of neuropsychiatric disorders. Therefore, we submitted Long-Evans rats (n=5) to a probabilistic reinforcement learning paradigm (i.e. bandit) to determine the behavioral effects of mid-striatal DBS. Rats received chronic biphasic electrical stimulation (300 µA, 0.05 ms pulse width, 130 Hz), delivered continuously for one hour prior to and during the bandit task. The task consisted of daily alternate sham and stimulation treatments. DBS significantly increased reaction time by 35 ms ($t = -4.55$, $p < 5.26e\text{-}7$) and the number of errors committed ($t = -3.89$, $p < 9.86e\text{-}05$). Furthermore, DBS disrupted the ability of rats to use flexible decision making strategies following high-conflict reversals, marked by reductions in learning rates. Continuing to investigate how DBS alters the decision-making processes associated with CSTC circuitry may help further our understanding of its therapeutic effects.

Disclosures: F. Iacobucci: None. **D. Cooper:** None. **E. Dastin-van Rjin:** None. **A. Widge:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.04

Topic: I.08. Methods to Modulate Neural Activity

Support: UMN CSE Graduate Fellowship
R01 EB026938
R01 MH119384

Title: Online Estimation of Impulse Response Functions for Desynchronization of Pathological Neural Rhythms

Authors: *J. N. NELSON[1], A. S. WIDGE[2], T. I. NETOFF[3];
[1]Dept Biomed Eng, Univerity of Minnesota, Minneapolis, MN, Minneapolis, MN; [2]Psychiatry, Univ. of Minnesota, Minneapolis, MN; [3]Dept Biomed Eng, Univerity of Minnesota, Minneapolis, MN

Abstract: Synchronous oscillatory activity has been shown to correlate with symptom severity for psychiatric disorders (obsessive compulsive disorder, treatment-resistant depression) in the cortical striatal network, for essential tremor in the cerebello-thalamo-cortical network, and for parkinson's disease in the cortico-basal ganglia thalamic network. In particular, greater power within certain frequency bands of a neural population's local field potential (LFP), or increased synchrony within the frequency bands of multiple neural populations' LFP, correlates with the severity of these disorders. Reducing pathological power or synchrony in neural activity can provide a biomarker for relieving behavioral and motor symptoms. The symptoms and underlying neural rhythms of these disorders fluctuate over time, which may render a static, open-loop neuromodulation device ineffective or inefficient for delivering therapeutic symptom relief. Phase-triggered, closed-loop neuromodulation using Phase Response Curves (PRCs) could provide a subtle way to modulate pathological synchrony between populations of neurons in the brain, without disrupting healthy neural oscillations. PRCs are impulse response functions that map the phase at which stimulation is delivered within a target neural population's rhythm to the resulting change in phase or amplitude of that population's neural rhythm. Estimating these response functions could enable rational design of phase-triggered stimulation. However, current approaches for estimating PRCs from electrophysiological data are offline methods, and current PRC-based neural desynchronization algorithms rely on static PRCs. Thus, closed-loop neuromodulation devices would benefit from adaptive algorithms that can calculate PRCs in real time. We have developed an algorithm for robust, online estimation of PRCs, which would enable the development of a phasic adaptive closed-loop controller for neuromodulation. We test this algorithm by fitting various PRC shapes, with convergence to a minimal mean square error within approximately 100 trials (about 1 2/3 minutes with a 1 Hz average stimulation frequency). We use this algorithm to estimate PRCs from computational neural mass models and local field potential data using minibatch stochastic gradient descent, with bayesian optimization hyperparameter tuning. We show that this adaptive estimation algorithm can, in principle, achieve reliable estimates of PRCs on a time scale comparable to an electrophysiological experiment or clinical session. A controller using this PRC estimate could update with changing patient neurophysiology.

Disclosures: **J.N. Nelson:** A. Employment/Salary (full or part-time):; University of Minnesota CSE Fellowship. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Method for Estimating Phasic Alterations in Physiologic Oscillations, Case Number: 2022-273. **A.S. Widge:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; R01 EB026938, R01 MH119384. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Method for Estimating Phasic Alterations in Physiologic Oscillations, Case Number: 2022-273. **T.I. Netoff:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Method for Estimating Phasic Alterations in Physiologic Oscillations, Case Number: 2022-273.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.05

Topic: I.08. Methods to Modulate Neural Activity

Title: Inefficient decision-making processes predict non-response to TMS for depression

Authors: *A. N. MCINNES, D. C. COOPER, S. T. OLSEN, C. R. P. SULLIVAN, A. S. WIDGE;
Univ. of Minnesota, Minneapolis, MN

Abstract: As a treatment for major depressive disorder (MDD), transcranial magnetic stimulation (TMS) is most often targeted at the dorsolateral prefrontal cortex (DLPFC), a key hub in decision-making circuitry. TMS of DLPFC may reduce the severity of MDD by augmenting decision-making related cognitive control and help patients to redirect maladaptive cognitive patterns. Identifiying specific cognitive deficits in psychiatric patients may aid in more personalized targeting in neuromodulation therapies for psychiatric disorders. Hence, here we examined the decision-making processes of individuals experiencing MDD who do and do not respond to TMS treatment. Participants (n = 16) undergoing daily TMS over DLPFC for treatment of MDD were each categorized into one of three groups (high-baseline response; low-baseline response; no-response), based on the severity of their depressive symptomology over the course of treatment. Participants completed weekly sessions of the WebSurf task, which in each trial, involves watching short entertaining videos after a delay period of 3-30 seconds. Participants are given the choice to either accept the delay period offer and wait for the video to play, or reject an offer and move to the next video category.Preliminary analyses indicate that the two response groups used the contextual value of offers to guide their decisions, whereby their decision latencies were shorter for low-value offers. In contrast, the no-response group unnecessarily deliberated on, and were less likely to reject offers regardless of their value, suggesting that they are unable to use offer value to guide their decisional processing. These findings indicate that there may be a sub-group of individuals experiencing MDD who have an impaired ability to use value-based contextual information to heuristically optimize decision-making processes, and that these individuals are unlikely to respond to DLPFC modulation. Given the non-responders showed impaired value-guided decision-making, these individuals may receive more benefit from modulation of cortical windows targeting orbitofrontal cortex, rather than DLPFC.

Disclosures: A.N. McInnes: None. **D.C. Cooper:** None. **S.T. Olsen:** None. **C.R.P. Sullivan:** None. **A.S. Widge:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.06

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH 5R01MH123634-02

Title: Real-time phase-locked stimulation systems for network-based conditions

Authors: *V. M. WOODS[1], J. H. WHEAR[1], S. NAGRALE[1], A. N. LUND[1], U. SHIN[2], M. SHOARAN[2], A. S. WIDGE[1];
[1]Univ. of Minnesota, Twin Cities, Minneapolis, MN; [2]EPFL, Lausanne, Switzerland

Abstract: Neurostimulation locked to the phase of ongoing brain oscillations appears to be a promising treatment strategy for a range of brain disorders, including stroke/trauma, Parkinson disease, and psychiatric illness. However, existing phase-locking algorithms require significant computational resources. They would be difficult to deploy in wearable or implantable systems because the complex number and trigonometric calculations utilized are computationally intensive and expensive. Providing such a system in an implant imparts added limitations on power consumption, and this work addresses the need for accurate phase extraction within an implant feasible power budget. Here, we present a flexible and efficient real-time framework to support novel stimulation strategies in neuroscience research, with a translational pipeline towards clinically viable therapies for network-based disorders.
First, we implemented a power-efficient phase-estimation algorithm in an open-source Open Ephys GUI (OEGUI) plugin for dissemination to the neuroscience research community. The flexibility of the OEGUI will allow the algorithm to be easily deployed for both preclinical and clinical studies of phase-locking stimulation. This software version demonstrated comparable phase accuracy as previously published algorithms (mean ± std): 3.09° ± 86.87° compared to Schatza et al (2022) at 0.42° ± 63.5°. Second, we translated the algorithm to an integrated circuit to enable its integration into an implantable neuromodulation therapy. Phase estimation accuracy, noise performance, and power consumption are reported compared to available systems using both in silico and in vivo neural signals. In particular, we show a power savings of 52.4% compared to other on-chip processing algorithms for real-time phase estimation. To demonstrate the phase-locked stimulation capabilities of the closed-loop system, stimulation was targeted at both a specific phase (180°) and a random-phase of theta-band local field potential (4-8 Hz), which is known to correlate with defense and threat appraisal behavior in the rodent model of disease. Taken together, both the software and hardware versions of this work offer a translational framework for the development of next-generation neuromodulation therapies for network disorders.

Disclosures: V.M. Woods: None. **J.H. Whear:** None. **S. Nagrale:** None. **A.N. Lund:** None. **U. Shin:** None. **M. Shoaran:** None. **A.S. Widge:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.07

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH grant 1R01NS120851-01A1

Title: Effects of Striatal DBS on the Trial-Irrelevant Activity in the Extradimensional Set Shifting Task

Authors: *K. SINGH[1], E. HOSKINS[2], E. DASTIN-VAN RJIN[3], A. E. REIMER[5], A. S. WIDGE[4];
[1]Dept. of Neurosci., Univ. Of Minnesota, Minneapolis, MN; [2]Dept. of Neurosci., [3]Dept. of Biomed. Engin., [4]Dept. of Psychiatry, Univ. of Minnesota, Minneapolis, MN; [5]Federal Univ. of São Carlos, São Carlos, Brazil

Abstract: Cortico-striatal circuits are central to several cognitive processes, including cognitive control-the ability to regulate and refrain from a learned behavior to achieve a specific goal. Deficits in cognitive control are common in many psychiatric disorders including obsessive-compulsive disorder and major depressive disorder. In clinical studies, deep brain stimulation (DBS) of cortico-striatal circuits can benefit patients with treatment-resistant psychiatric disorders through a mechanism involving enhanced cognitive control. Prior results from our lab have replicated this effect in male, Long-Evans rats (n=9) receiving DBS while performing an extradimensional set-shifting task, a task used to assess cognitive control. We observed that rats performed task-irrelevant nosepoke behaviors during periods between trials. To further characterize the effect of DBS on cognitive control, we reanalyzed the prior data with specific focus on these intertrial intervals (ITIs). We found that activity during ITIs was purposeful and patterned, with evidence of corrective responses, compulsivity, and pre-commitment/rehearsal behaviors. Additionally, we identified two distinct phases of ITI activity corresponding to the beginning and last few seconds of the interval. Further analysis revealed that activity in the initial phase correlated with increased corrective responses after the previous trial was incorrect. In contrast, the final phase was predominantly composed of responses after correct trials, with activity predicting a rat's response on the following trial, indicating pre-commitment. Furthermore, we found that stimulation of the mid-striatum specifically increased pre-commitment behavior at the end of the ITI. These findings could be extended in future work to further examine response prediction, compulsivity, and flexibility, thereby maximizing mechanistic understanding and future therapeutic benefit.

Disclosures: K. Singh: None. **E. Hoskins:** None. **E. Dastin-van Rjin:** None. **A.E. Reimer:** None. **A.S. Widge:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.08

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant R21MH109722
NIH Grant R21MH113103
NIH Grant R01MH11938402
MnDRIVE Brain Conditions Initiative
Minnesota Medical Discovery Team on Addictions

Title: State dependence of connectivity alterations from closed loop phase locked stimulation

Authors: *K. DRAHOS, R. L. YOUNK, S. YADAV, S. NAGRALE, A. S. WIDGE;
Univ. of Minnesota, Minneapolis, MN

Abstract: Information flow in brain networks may depend on the oscillatory synchrony of local field potential (LFP) between regions. For fear-related disorders, the connection between the medial prefrontal cortex (mPFC) and basolateral amygdala (BLA) is particularly important. Theta-band (4-8 Hz) PFC-BLA LFP synchrony correlates with the ability to learn and recall safety memories, but acute stressors alter the oscillatory synchrony between these regions. Manipulating that synchrony may thus require strategies that adapt to changes in the circuit biology. We sought to assess an adaptive stimulation method to alter PFC-BLA connectivity based on stimulation locked to the theta oscillation.Electrodes with 8 recording channels and one stimulating channel were implanted into the prelimbic or infralimbic cortex (PL or IL, part of the rodent homologue of mPFC) and BLA of Long-Evans rats. Rats underwent 20 phase-locked stimulation sessions. Single electrical pulses (45 μs and 100 μA) were delivered to BLA when mPFC phase was at 0, 90, 180, 270°, or random at theta band for a total of 4 sessions per phase. Each session consisted of pre recordings followed by 30 minutes of phase-locked stimulation, and 60 minutes of post recordings. Rats then underwent a three-day tone-shock and extinction paradigm with habituation/conditioning, extinction, and recall phases. After the recall phase, rats underwent 20 phase-locked stimulation sessions as previously described. We measured stimulation induced LFP connectivity changes for targeted phase during pre/post conditioning. Prior to conditioning, 180-degree phase-locked stimulation best increased PL-BLA LFP synchrony. Yet, post conditioning the 90-degree PL-BLA phase-locked stimulation was most effective at increasing LFP synchrony. Since IL and PL are both a part of the mPFC we expect IL-BLA LFP synchrony to follow a similar trend. In summary, we have proposed a phase-locked electrical stimulation technique to alter brain oscillatory synchrony. The change in optimal stimulation phase pre and post tone-shock conditioning shows that the response to stimulation is state dependent and altered by emotionally valenced learning (shock conditioning). Identifying methods for manipulating mPFC-BLA connectivity that are viable after acute stressors may be a necessary step towards developing therapies for patients with treatment-resistant psychiatric disorders.

Disclosures: K. Drahos: None. **R.L. Younk:** None. **S. Yadav:** None. **S. Nagrale:** None. **A.S. Widge:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.09

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant R21MH109722
NIH Grant R01MH11938402
NIH Grant R21MH113103
MnDRIVE Brain Conditions Initiative
Minnesota Medical Discovery Team on Addictions

Title: Quantifying defensive behavior and threat response through integrated headstage accelerometry

Authors: *R. L. YOUNK, A. S. WIDGE;
Psychiatry, Univ. of Minnesota, Minneapolis, MN

Abstract: Rodent defensive and threat-related behaviors are common targets of investigation, because they model aspects of human mental illness. These behaviors are typically quantified by video recording and post hoc analysis. Those quantifications can be laborious and computationally intensive. Depending on the analysis method, the resulting measurements can be noisy or inaccurate. Other defensive behaviors, such as suppression of operant reward seeking, require extensive pre-training. We sought to identify a method, using commodity hardware, for quantifying defensive behavior by 3-axis accelerometry integrated with an electrophysiology headstage. Rats were trained to lever press for pellets before surgical implantation of electrodes. With a headstage attached, rats underwent a tone-shock conditioning protocol with three phases: habituation/conditioning, extinction, and extinction recall. Behavior as measured via accelerometry, video based freezing scores, and bar press were assessed for each rat and phase. We processed the accelerometry data through converting the 3-axis voltages to total acceleration, calculating acceleration change, Gaussian smoothing, and normalizing from 0 to 1. We ran a cross-correlation between accelerometry and the other metrics, and then shifted accelerometry by any potential lag. We correlated freezing and bar press against accelerometry as transformed through seven processing methods: raw, raw change, smoothed raw, smoothed change, smoothed shifted, smoothed change shifted, and smoothed normalized change. The best approach to tracking defensive behavior from accelerometry was Gaussian filter smoothing of the first derivative (change score). Behavior scores from this method reproduced canonical conditioning and extinction curves at the group level ($r=0.86$ between accelerometry and bar press). At the individual level, timepoint-to-timepoint correlations between accelerometry, video, and bar press metrics were statistically significant but modest (largest $r=0.52$, between accelerometry and bar press). In this work, we developed an approach to detect rodent defensive behaviors based on continuous motion sensors. Accelerometer measurements of defensive behavior require less specialized software and no additional animal training. The similarities in behavioral tracking and modest correlations between each metric suggest that each measures a distinct aspect of defensive behavior. Accelerometry is a viable alternative to current defensive measurements, and

its non-overlap with other metrics may allow a more sophisticated dissection of threat responses in future experiments.

Disclosures: **R.L. Younk:** None. **A.S. Widge:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.10

Topic: I.08. Methods to Modulate Neural Activity

Support: NSF GRFP CON-75851
NIH Grant 1R01NS120851-01A1

Title: Rodents solve an extradimensional set-shifting task by forgetful, adaptive reinforcement learning

Authors: *E. M. DASTIN-VAN RIJN, A. ALPERS, E. SACHSE, A. WALD, A. REIMER, E. SONG, A. S. WIDGE;
Univ. of Minnesota, Twin Cities, Minneapolis, MN

Abstract: Cortico-striatal circuits are central to several cognitive processes, including attentional set-shifting. Deficits in this process are common in obsessive-compulsive disorder, major depression, and many other disorders. Prior results from our group have demonstrated improvements in rodent performance on an operant set-shifting task when deep brain stimulation was applied. To characterize the cognitive basis of this stimulation effect, we fit a series of computational models to rodent behavior.Long-Evans rats (n=32) were trained on an extradimensional set-shifting task. In this task, rats had to shift between selecting stimuli based on side or light to receive rewards. Using Akaike Information Criterion (AIC) as a measure of model performance, we fit a set of 11 models to rat behavior. We used Bayesian model selection to compare performance and evaluated the ability of each model to replicate rat behavior based on a series of task metrics. A feature-specific, forgetful, adaptive reinforcement learning model fits the behavior best (r=0.85, $\varphi \approx 1$). Model behavior was not significantly different from rat behavior for trials to finish (p=0.04), error rate (p=0.03), and rule-specific error rate (p=0.04 and p=0.04). Stimulation specifically in the mid-striatum significantly improved forgetfulness (p=0.02), a parameter related to the ability to discount the value of options that were no longer being chosen or considered rewarding.Models utilizing only a singular task dimension were unable to effectively describe rat behavior. Instead, rat behavior is well described by a feature-specific, forgetful adaptive reinforcement learning model indicating rats flexibly adapt to different reward structures in the environment. Additionally, the effect on forgetfulness suggests that mid-striatal stimulation enhances behavioral features requiring cognitive flexibility. This model could be used in future work in rats and humans to determine and eventually predict the behavioral effects of psychiatric neuromodulation.

Disclosures: **E.M. Dastin-van Rijn:** None. **A. Alpers:** None. **E. Sachse:** None. **A. Wald:** None. **A. Reimer:** None. **E. Song:** None. **A.S. Widge:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.11

Topic: I.08. Methods to Modulate Neural Activity

Support: NIHR01NS113890
Brigham Research Institute Microgrant

Title: Use two-photon imaging and single-cell optogenetics to dissect cortical circuit connectivity

Authors: *M. YE[1], S. M. SMIRNAKIS[2];
[1]Neurol., [2]Neurology, Room BB-326, Mass Gen. Brigham, Harvard Med. Sch., Boston, MA

Abstract: Sensory perception, memory, emotion, intelligence, creativity and many other brain functions are but the symphonies of electrical signals produced by diverse neurons and their complex recurrent circuits. Deciphering their operating logic is key to understanding information processing in the brain and the emergence of cognitive functions. Here, we use two-photon calcium imaging and a single cell specific SLM (spatial light modulator) based optogenetic strategy to probe the mechanisms by which patterns of activity are generated in cortical neuronal ensembles. Specifically, we use SLM coupled to ChroME expression to manipulate the activity in select groups of neurons and study the effect on the nearby circuit. The ultimate goal is to decipher the operating logic of cortical circuitries underlying sensory perception. Preliminary data showed that, under certain conditions, SLM-guided stimulation of a single SOM+ interneuron in Layer 2 of the primary visual cortex surprisingly excited, but not inhibited, a set of surrounding neurons within a distance of ~100 μm with activity that persisted for longer than 30s. The activity propagated outward from the stimulated neuron in a wave-like fashion, engaging neurons with latencies ranging from 0.2 to 2.4 s, defined as the time difference between the onset of photo-stimulation and the time at 10% peak fluorescence of the engaged neurons. It is likely that this pattern of activity was generated by recurrent cortical networks involving disinhibition of other interneurons, potentially PV+ cells. In future studies we plan to identify the neuronal components mediating the observed phenomenon, as well as to study its potential relation to sensory stimulus processing. It remains to be seen whether the response elicited by the type of stimulation we used reflects a physiological pattern of activation of the underlying circuit, versus an aberrant wave of activity propagating through the cortex.

Disclosures: **M. Ye:** None. **S.M. Smirnakis:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.12

Topic: H.08. Learning and Memory

Support: NRF-2020M3E5D9079908
NRF-2019R1A2C2005213
NRF-2020M3E5D9079913
Creative-Pioneering Researchers Program

Title: Three types of neurons encoding flexible, stable and both values of visual objects in the primate ventral striatum

Authors: *S.-H. HWANG[1], H. F. KIM[2];
[1]SNU, Seoul, Korea, Republic of; [2]Sch. of Biol. Sci., SNU Med. Library, Seoul, Korea, Republic of

Abstract: Ventral striatum (VS) in the brain is a well-known region where changes of reward values are updated, contributing to habit learning. However, a recent study (Kang et al., 2021) reported its new role that the posterior part of the VS maintains stable value memory of visual objects over a long period of time. This data raises a question of how individual neurons in the VS encode flexible and stable value of visual objects. To address this, we recorded visually responsive neurons in the VS and tested individual neurons encode different types of values (flexible value and/or stable value). For testing the flexible value encoding, contingency of visual object and reward value was reversed across each block of 24 trials. Among 79 visually responsive neurons in a monkey, 56 neurons (71%) showed discriminative activity for flexibly changed values of visual objects and these neurons were found in most regions of the VS. To test the stable value encoding in the VS neurons, the monkey first learned the object values in learning task and the learned value memory was tested in passive-viewing task. In the learning task, the object-value contingency was maintained during days of the learning sessions (> 4days of learning). The stable value-guided behavior was tested by habitual gaze of the monkey as previously described: the monkey showed gaze bias to previously learned high-valued objects in free-viewing state where no reward outcome was given (Kang et al., 2021). Next, to test neural representation of the stable value memory, we recorded the neural responses to the learned high- and low-valued objects with no reward outcome while the monkey fixated at a central white dot (passive-viewing task). Total 19 visual neurons (24%) showed a differentiated value response, and these stable value-coding neurons were mainly located in the medial and posterior parts of the VS. Among these neurons, 13 neurons showed higher activities for rewarded objects than non-rewarded ones (positive value coding), while 6 neurons responded more strongly to non-rewarded objects than rewarded ones (negative value coding). Interestingly, among these stable value-coding neurons, 12 out of 19 neurons (63%) also represented to the flexible change in object-value contingency (both value type neurons), but the rest of neurons selectively encoded stable values. Our data show that in the VS, there are heterogeneous types of neurons that encode three different types of values: stable, flexible, and both value-coding neurons.

Disclosures: S. Hwang: None. **H.F. Kim:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.13

Topic: H.08. Learning and Memory

Title: Characterising foraging dynamics during naive learning across many options in time and space

Authors: *L. L. GRIMA, J. T. DUDMAN;
Janelia Res. Campus, Ashburn, VA

Abstract: The ability to rapidly learn about and therefore exploit multiple options simultaneously as a form of foraging is highly advantageous in many situations, and for many species. Studying foraging in the context of multiple (3+) options also allows for greater differentiation between models that would otherwise make very similar predictions when only two alternatives are available. This also affords the opportunity to investigate underexplored strategies relating to the choice of multiple alternatives. With this in mind, we studied freely moving mice as they sampled from six ports in a large (7ft x 1.5ft) arena. Each port was concurrently rewarded at a deterministic interval, ranging from 30s to 2400s, and mice were free to sample ports in any order and frequency across sessions lasting 3 hours. Naive mice quickly (< 20 minutes) learned the relative value of all six ports within a single session and were able to collect the majority of available rewards by the end of the first session. When port quality was subsequently varied across days, mice again rapidly learned the new port values and updated their sampling of ports appropriately. To understand the strategy underlying the mice's ability to learn and update their behaviour in this task context, we explored a range of decision models of varying sophistication. We found that mice used the history of visits at each port to learn its respective quality and thus match their visit rate to the port in question. As ports were also distributed in space, we also investigated the effect of port-to-port distance on sampling decisions and found that mice shaped their behaviour according to the time to travel between ports. Finally, we also recorded dopamine levels in the nucleus accumbens core or dorsal striatum using dLight as mice behaved in the arena, and found differential encoding of port quality across mesolimbic and nigrostriatal dopamine.

Disclosures: L.L. Grima: None. **J.T. Dudman:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.14

Topic: H.08. Learning and Memory

Support: DFG JA 1999/3-1
ERC StG MEMCIRCUIT 758032

Title: Dopamine signaling across striatal subregions during acquisition of instrumental associations

Authors: *T. BERNKLAU[1,2], B. RIGHETTI[1], L. S. MEHRKE[1], S. JACOB[1];
[1]Translational Neurotechnology Laboratory, Dept. of Neurosurg., Klinikum rechts der Isar, Tech. Univ. of Munich, Munich, Germany; [2]Grad. Sch. of Systemic Neurosciences, Ludwig-Maximilians-University Munich, Munich, Germany

Abstract: Phasic dopamine activity plays a key role in learning and decision-making and serves as a teaching signal by encoding reward prediction error (RPE), the difference between predicted and actual reward. Although most dopamine neurons encode RPEs, dopaminergic projections to their main target, the striatum, have been shown to differ across subregions. RPE coding has been extensively studied in Pavlovian tasks or simple probabilistic instrumental tasks, while more complex learning paradigms are less abundant. Predictive dopamine signals during cues and reinforcing dopamine signals during outcomes are strongly coupled to reward expectations, when those are independent of actions (as in Pavlovian tasks) or tied to external reward probabilities and magnitudes during well-trained behavior (as in probabilistic instrumental tasks). How predictive and reinforcing dopamine signals evolve during the acquisition of instrumental associations in a deterministic task without external reward manipulations remains elusive. We found that predictive and reinforcing dopamine signals can be uncoupled during the acquisition of instrumental associations, when rewards are action-dependent. We trained head-fixed mice in an auditory decision-making task with rule switches, while we performed direct fluorescent imaging of dopamine using fiber photometry in the ventral striatum, dorsomedial striatum, and dorsolateral striatum. Depending on the learning state, mice used different strategies to perform the task. During learning, but not in the well-trained state, mice showed a tendency to repeat previous choices, but no other trial history-related biases. When animals were fully trained, they followed only the instruction cue to guide their choice. While dopamine outcome responses were inversely scaled with task performance, and thus reward expectation, dopamine cue responses were not reciprocally scaled. Predictive and reinforcing dopamine signals were uncoupled after rule switches and the RPE signature was intact again when associations were re-established, suggesting a mechanism for learning. The strongest effects were observed in the ventral striatum, while the dorsomedial striatum preferentially encoded predictive signals, and the dorsolateral striatum preferentially encoded reinforcing signals, in line with their assumed roles as associative and sensorimotor striatum, respectively. These findings contribute to the understanding of specialized dopaminergic mechanisms for the acquisition of instrumental associations.

Disclosures: **T. Bernklau:** None. **B. Righetti:** None. **L.S. Mehrke:** None. **S. Jacob:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.15

Topic: H.08. Learning and Memory

Support: NARSAD Young Investigator Award
Rutgers Busch Biomedical Research Award
NIH NS034865

Title: Nicotinic signaling in striatal circuits and striatum-dependent behavior

Authors: *E. B. GUVEN, S. KOCATURK, F. SHAH, J. M. TEPPER, M. SHIFLETT, M. ASSOUS;
Rutgers Univ. Ctr. for Mol. and Behavioral Neurosci., Newark, NJ

Abstract: Striatal cholinergic interneurons (CINs) are the principal source of acetylcholine (ACh) in the striatum. They play an essential role in modulating striatal circuits and behaviors including reward processing, goal-directed behavior and behavioral flexibility. ACh exerts its effects through pre- and postsynaptic muscarinic and nicotinic receptors (NRs). Many studies have highlighted the role of muscarinic signaling due to their widespread expression in striatal projection neurons (SPNs). However, recent findings have indicated that CINs also form a fast synaptic circuitry involving NRs and in particular those containing β2 subunits (β2-NRs), expressed selectively by striatal GABAergic interneurons (GINs). Our previous work has demonstrated that optogenetic activation of CINs evokes large IPSCs, consisting of two components ($GABA_{Afast}$ and $GABA_{Aslow}$), in SPNs due to the direct nicotinic activation of one or more subtypes (likely several) of GINs. Nonetheless, the synaptic mechanisms and the functional roles of these circuits have not been precisely identified. Using multiple double transgenic mouse lines, we found that in addition to NGF interneurons tyrosine hydroxylase interneurons (THINs), low-threshold spiking interneurons (LTSIs), fast-spiking interneurons (FSIs) and spontaneously active bursty interneurons (SABIs) receive suprathreshold cholinergic input involving distinct NR-mediated excitation. Given the striatal connectivity, we suggested the involvement of THINs in the disynaptic inhibition of SPNs following synchronous activation of CINs. Using a double transgenic/ double optogenetics strategy, inhibition of THINs while simultaneously activating CINs produced a significant reduction in both the $GABA_{Afast}$ and the $GABA_{Aslow}$. While the former can be explained by striatal synaptic connectivity of THINs, the change in the $GABA_{Aslow}$ is more puzzling. Our lab previously suggested that NGFs are the primary source of striatal $GABA_{Aslow}$. Thus, we hypothesized and demonstrated a functional heterotypic electrical coupling between striatal NGFs and THINs. Lastly, using transgenic β2-floxed mice, we were able to remove β2-NRs in striatal GINs and test their role in a striatum-dependent behavior, such as cognitive flexibility. Our preliminary results indicate that striatal β2-KO mice exhibit a difficulty to adapt their behavior when the task contingencies are switched in a reversal learning paradigm, hence suggesting a significant impairment in cognitive flexibility. Overall, our results elucidate a significant role for striatal β2-NRs in coordinating striatal output activity and behavior.

Disclosures: **E.B. Guven:** None. **S. Kocaturk:** None. **F. Shah:** None. **J.M. Tepper:** None. **M. Shiflett:** None. **M. Assous:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.16

Topic: H.08. Learning and Memory

Support: NIH Grant R01MH123661

Title: Reward-guided decision making is modulated by sex and 16p11.2 hemideletion in mice

Authors: *M. J. MERFELD[1], C. CHEN[2], E. M. GIGLIO[2], N. M. GRISSOM[2];
[1]Grad. Program in Neurosci., [2]Dept. of Psychology, Univ. of Minnesota, Minneapolis, MN

Abstract: Gene variants associated with neurodevelopmental disorders, including copy number variation at the 16p11.2 region, are known to impact reward learning and corticostriatal function in mouse models. These impacts, however, appear to differ across sexes, raising the question of how sex modulates the influence of autosomal gene variants on reward processing and striatal dopamine neurobiology. We have previously observed robust differences in reinforcement learning strategies across males and females in a reward-guided restless bandit task, with male animals spending longer exploring options. We are using this task to understand how sex modulates the impact of 16p11.2 hemideletion on reward-guided cognition. Training data prior to bandit performance suggests that 16p11.2 hemideletion female animals complete an increased number of trials compared to sex-matched control animals. In the bandit task, we use a Hidden Markov Model to define trials as exploration or exploitation, and preliminary results suggest that strategies are modulated by state in a way that is sex- and genotype-dependent. As the nucleus accumbens is critical in reward-guided learning such as bandit tasks, we will examine dopamine release in the nucleus accumbens core (NAcc) using dLight fiber photometry to determine whether dopaminergic signaling differs across sexes and between genotypes, and how that may contribute to differences seen in task performance. We hypothesize that 16p11.2 hemideletion animals will have increased dopamine release in the NAcc during rewarded trials, and this difference will be more pronounced in females.

Disclosures: **M.J. Merfeld:** None. **C. Chen:** None. **E.M. Giglio:** None. **N.M. Grissom:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.17

Topic: H.08. Learning and Memory

Support: NIH R01 DC018650
NSF CAREER 2145247

Title: Behavioral evidence for offline consolidation in a procedural learning task in mice

Authors: R. MONGA[1], C. DRIEU[2], K. KUCHIBHOTLA[3];
[1]Psychological and Brain Sci., [2]Psychological & Brain Sci., [3]Psychological and Brain Sciences, Neurosci. and Biomed. Engin., Johns Hopkins Univ., Baltimore, MD

Abstract: Most conceptual and analytical models assume that procedural memory formation in animals is dominated by 'online' trial-and-error learning. And, yet, 'offline' processes are known to play a critical role in declarative forms of memory, including episodic-like memory in rodents. To what extent do offline processes contribute to procedural learning in rodents? To address this, we reasoned that if online processes dominate in a procedural learning task, then reducing the number of training trials per session by half would double the number of sessions required to reach expert performance. We trained water-restricted mice on an auditory go/no-go task in which they had to lick to one tone (S+) to obtain a water reward and withhold licking to another (S-) to avoid a timeout. We assayed learning in reinforced and non-reinforced (probe) trials to allow us to dissociate between 'acquisition' of task contingencies (measured in probe trials) and slower behavioral 'expression' (measured in reinforced trials). Mice (n=6) that received 140 reinforced trials per session took 4.1 ± 2.3 sessions to acquire the task contingencies (measured in probe trials) and 13.0 ± 4.9 sessions to express those contingencies (measured in reinforced trials). We then reduced the number of trials per session to 70 (n=8). Surprisingly, these mice acquired (5.0 ± 1.4 sessions) and expressed (11.6 ± 3.0) the task in the same number of sessions ($p=0.6$) despite experiencing only half the trials. These results suggest that offline processes contribute to procedural learning. To further test this possibility, we reduced the number of trials per session to 35. Mice (n=5) acquired the task contingencies in slightly more sessions (7.0 ± 0.8 sessions) but still with significantly fewer trials than predicted from the 140 or 70 trial-per-session tasks ($p<0.01$). Interestingly, these mice took far more sessions to express the task contingencies in reinforced trials with 3 of 5 mice not reaching criterion even after 50 sessions. These data support the model that procedural learning exhibits two, dissociable learning processes and provides new behavioral evidence that acquisition leverages offline processes to enhance learning while expression depends more heavily on trial-and-error practice.

Disclosures: R. Monga: None. **C. Drieu:** None. **K. Kuchibhotla:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.18

Topic: H.08. Learning and Memory

Support: NIH Grant MH002928

Title: Motivational preference for secondary reinforcers during reinforcement learning

Authors: *D. BURK, B. B. AVERBECK;
NIMH/NIH, NIMH/NIH, Bethesda, MD

Abstract: Much of our behavior is motivated by secondary reinforcers, like money. While money cannot directly satisfy needs, it can be exchanged for primary reinforcers, such as food or water. Secondary reinforcers can have powerful motivational value, as many species will work to obtain them. In the present study, we examined how monkeys learn to make choices to obtain tokens in a reinforcement learning task. At the beginning of a block of trials we introduced unfamiliar visual images, each of which was associated with a different outcome. In each trial, after a fixation period, two of the images were presented. The monkey then made a saccade to one of the images to indicate their choice. Each image was associated with tokens, such that choice of that image led to a change in the number of orange (juice) or blue (water) tokens. The images were randomly assigned to token outcomes such that the monkey received either 2 juice tokens, 1 juice token, 2 water tokens or 1 water token after choosing the corresponding image. The monkey had to learn over trials which images delivered the tokens for the larger volume and preferred fluid and to choose those images as often as possible. Tokens were accumulated across trials and cashed out after 4-7 correct trials. During cash-out, the monkey received one drop of water for each blue token and one drop of juice for each orange token. Preliminary data from one monkey shows that he learned to select images that delivered juice tokens, even when the offer pair included an option with more water tokens. Thus, he had a strong preference for juice tokens, that were later traded for primary juice rewards. Data was collected from four variants of this task: (A) juice 1 (orange tokens) vs. water (blue tokens); (B): juice 1 (orange) vs. juice 2 (pink); (C) juice 1 (orange) vs. juice 3 (purple), and (D) juice 1 (orange) vs. water (blue) with an additional null cue. Data from these task variants show that monkeys can learn to associate differently colored tokens with liquid reward outcomes, and they can learn to choose images that maximize tokens for preferred outcomes. In addition, Rescorla-Wagner (RW-RL) models were fit to the behavioral data and show that the monkey valued tokens associated with preferred outcomes more than tokens associated with less preferred outcomes. A Temporal Difference (TD) model was developed to assess the contribution of task factors to choices and to make predictions about neural responses across cortical and subcortical circuits. This task and modeling framework allows us to explore how traditional RL models do and do not capture learning from secondary reinforcers when there is more than one, as is typical in a real-world environment.

Disclosures: D. Burk: None. **B.B. Averbeck:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.19

Topic: H.08. Learning and Memory

Support: NIH Grant ZIA MH002928

Title: Reinforcement-learning in the macaque monkey performing a two-arm bandit task requiring eye and reaching movements

Authors: *F. GIARROCCO, C. J. LEWARNE, R. BARTOLO, B. B. AVERBECK;
Natl. Inst. of Mental Hlth., NIH, Bethesda, MD

Abstract: Learning to maximize reward and avoid punishment is crucial for an effective interaction with the environment. This process relies on the brain's ability to encode a value representation of stimuli and/or actions, and to use this representation to select behaviors that maximize the future rewards. Reinforcement-learning (RL) models have been particularly fruitful in describing behavioral and neural value-related learning processes. These processes are typically studied in RL paradigms where choices are made through a single motor system, usually either the oculomotor or the skeleto-motor. Here, we trained a macaque monkey in a novel version of a two-arm bandit task in which we randomly intermixed blocks of trials that required a choice with either an eye or a reaching movement. On each block the monkey was presented with two new stimuli associated with a different probability of receiving the reward and, by trial and error, the monkey learned to maximize reward by choosing the most frequently rewarded stimulus. We found that monkey's choice behavior was characterized by better performance in reaching blocks compared to eye blocks in terms of how often the most rewarded stimulus was selected. We then used a standard RL model and estimated the learning rate and the inverse temperature parameters during eye and reaching blocks. We found that the RL model fit observed monkey behavior well and the learning rate was higher during reaching compared to eye blocks. We then used the inverse temperature parameter of the RL model as a measure of choice consistency. A high (low) inverse temperature indicates that the monkeys more (less) frequently chose the better option. Consistent with the choice accuracy, we found that the inverse temperature was higher during reaching compared to eye blocks. Next, we analyzed the reaction times (RTs) and the speed-accuracy tradeoff in both eye and reaching blocks. We found that during eye blocks the accuracy peaks near the average RT, while during reaching blocks the highest accuracy occurred during shorter RTs. This is consistent with the further observation that RTs decreased as the learning increased during reaching but not eye blocks. Our results suggest that different value-related processes might drive learning in the eye and reaching blocks. One hypothesis is that there may be a different value representation across brain regions subserving eye and reaching movements. Upcoming neuronal recordings in different cortico-basal ganglia circuits will allow us to investigate whether and how these value representations are encoded in the brain and make hypothesis on how they drive learning.

Disclosures: F. Giarrocco: None. **C.J. LeWarne:** None. **R. Bartolo:** None. **B.B. Averbeck:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.20

Topic: H.08. Learning and Memory

Support: NIMH ZIA MH002928-01

Title: Neural circuit mechanisms underlying learning from gains and losses in monkeys

Authors: *H. TANG, R. BARTOLO, B. B. AVERBECK;
NIMH/NIH, NIMH/NIH, Bethesda, MD

Abstract: Reinforcement learning (RL) is the behavioral process of learning to associate stimuli or responses with gaining and losing positive reinforcers. Recent studies revealed that RL is mediated by a broad set of cortical and subcortical regions, which can be grouped into ventral and dorsal systems. The ventral circuit is critical for specifying behavioral goals by updating and maintaining the value of stimuli, and the dorsal circuit is critical for orchestrating actions to obtain the goals (H. Tang *et al.*, 2022). However, how value-relevant information flows across the ventral circuit during learning of gains and losses has not been revealed. To address this question, we simultaneously collected neural activity in the orbitofrontal cortex (OFC), ventral striatum (VS), amygdala, and medial portion of the mediodorsal (MD) thalamus from rhesus macaques as they performed a two-armed bandit token reward learning task. In the task, the monkeys learned to choose between two images associated with different values (+2, +1, −1, −2 accumulated tokens). The tokens were periodically exchanged for primary reinforcers. The monkeys learned to associate visual stimuli faster for gaining than losing tokens. Among areas, OFC neurons show the best discrimination of gains and losses; VS and amygdala neurons also represent these well, but the activity in these structures is more relevant to the value of the stimuli, not gains vs. losses. The MD thalamus neurons, however, do not encode gains vs. losses as well as the others. When choice outcomes were revelated, value information flowed in a subcortical-to-cortical direction: from the amygdala and VS to OFC through MD. Our results revealed the distributed representation of gains and losses of secondary reinforcers and the flow of value information in the ventral frontostriatal system during RL.

Disclosures: H. Tang: None. **R. Bartolo:** None. **B.B. Averbeck:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.21

Topic: H.08. Learning and Memory

Title: Differential value coding of learning from gains and losses in multiple prefrontal areas

Authors: *C. TASWELL[1], R. BARTOLO[2], M. JANSSEN[3], B. B. AVERBECK[4];
[1]NIH/NIMH, Washington, DC; [2]SLDM, NIMH, Bethesda, MD; [3]NIMH, North Potomac, MD;
[4]NIMH/NIH, Bethesda, MD

Abstract: Adaptive behavior requires learning to gain rewards and avoid losses. Most work on learning has focused on learning to gain rewards. Therefore, we have developed a paradigm that allows us to examine whether the same or different neural systems underlie learning from gains vs. losses. We have previously shown that Ventral Striatum (VS) plays a specific role in learning to select between rewarded outcomes (Taswell, Costa et al. 2018). In a follow-up study (under review) we found that the learning deficits VS monkeys displayed in (Taswell, Costa et al. 2018) was due to motivation, and not learning ability. Additionally, in this same study, we showed that monkeys with lesions to the amygdala performed as well as controls. These results suggest that there is different neural circuitry that underlies learning values associated with gains and losses. In the present study, we recorded single and multiunit activity using 3 microelectrode arrays (96 channels per array) implanted in the orbitofrontal cortex (OFC), dorsolateral prefrontal cortex (DLPFC), and ventrolateral prefrontal cortex (VLPFC). In addition to these arrays, we also simultaneously recorded single and multiunit activity using 2 microelectrode arrays (64 channels per array) implanted in the anterior cingulate cortex (ACC). Activity was simultaneously recorded in these areas while monkeys completed the stochastic token experiment presented in (Taswell, Costa et al. 2018). In this experiment, we conditioned tokens as reinforcers in a task where animals could both gain and lose tokens. We introduced four novel images in each block which had associated outcomes of +2, +1,-1,-2 tokens, 75% of the time, the outcome was the value of the cue, and 25% of the time, the outcome was 0. Monkeys had to learn over trials, in each block, the outcomes associated with each cue, and choose the best cue in each trial. Preliminary results show that information about chosen stimuli and their associated value is encoded in the OFC, VLPFC, and ACC at different time periods during trials. Additional analyses will further allow us to characterize these effects.

Disclosures: C. Taswell: None. **R. Bartolo:** None. **M. Janssen:** None. **B.B. Averbeck:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.22

Topic: H.08. Learning and Memory

Support: ZIA MH002928

Title: Behavioral dissection of goals and actions

Authors: *S. WANG, C. ROBINSON, B. B. AVERBECK;
NIMH/NIH, Bethesda, MD

Abstract: Animals adaptively plan actions to achieve goals. The successful conversion of a goal into a sequence of actions to achieve the goal is crucial in adaptive behavior. One critical challenge in studying the neural mechanism underpinning goal-action conversion is the behavioral dissection of goals and actions. In previous experiments, goals and actions are often confounded. For example, in saccade-based learning experiments in macaques, the saccade direction (action) is confounded by the location of the chosen image (goal). To address this limitation, we designed a spatial sequential learning task in which we explicitly separated the choice of image (goal) and the choice of action by allowing multiple routes of actions to reach the same goal.

We trained macaques to choose between two images that deliver rewards at complementary probabilities. The images can appear in any two corners of a computer screen randomly. To choose an image, monkeys were trained to make horizontal and vertical saccades following one of the two L-shaped paths from the screen center to the target image. For example, to choose the upper right image, the monkey can either go up and then right, or right and then up. In each trial, only one of the two paths leading to each image is available. The path availability is indicated by the presence of a white square at the joint of the L-shaped path (L-joint). The monkeys started each trial by looking at the central fixation point. The images were flashed for 100ms at two random target locations. Then, two white squares were flashed at either the vertical (up and down) or horizontal (left and right) L-joints. The monkeys were then asked to make two saccades, the first from the center to one of the four L-joints (only 2 are valid), and the second from the chosen L-joint (if valid) to the target image. Behaviorally, we showed that our monkeys successfully learned the task and are capable of planning actions based on action availability to achieve goals on a trial-by-trial basis.

Neurally, previous research has suggested that goals and actions may be encoded in different areas of the brain. Goal values are mostly encoded in the ventral cortico-striatal circuit, whereas saccade directions are mostly encoded in the dorsal cortico-striatal circuit. However, how goal information is communicated between ventral and dorsal cortico-striatal circuits to drive action selection remains unclear. In our study, we plan to record simultaneously from anatomically connected nodes in both ventral and dorsal circuits (namely, amygdala and dorsal lateral prefrontal cortex) while monkeys perform the task to study the information flow between the two areas that underlies goal-action conversion.

Disclosures: S. Wang: None. **C. Robinson:** None. **B.B. Averbeck:** None.

Poster

570. Integrative Behavioral and Circuit-Based Approaches

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 570.23

Topic: H.08. Learning and Memory

Support: NIAAA U01AA025932
NIAAA R01AA021505
R01AA027768

Title: The critical roles of CIN burst-pause firing in extinction of learned action-outcome contingencies and acquisition of new contingencies

Authors: *Z. HUANG[1], J. KILLINGWORTH[1], M. CHILDS[1], J. WANG[2];
[1]Texas A&M Univ. Hlth. Sci. Ctr., Bryan, TX; [2]Texas A&M Univ. Hlth. Sci., Bryan, TX

Abstract: Although cholinergic interneurons (CINs) compose only 1-2% of striatal population, CINs exert powerful impact on striatal functions. CINs exhibit a unique burst-pause firing pattern in response to salient stimulus. This firing pattern is believed to critically modulate striatal outputs and dopamine-dependent corticostriatal plasticity. However, the behavioral consequences of altered burst-pause firing of CINs in neuropsychiatric disorders have largely been unexplored. Here, we used two different methods to manipulate the burst-pause firing of CINs and tested their behavioral consequences in alcohol use disorder. First, using a chronic alcohol-drinking animal model we observed the reduced burst-pause firing of CINs and impaired behavioral flexibility (failed to learn reversed action-outcome contingencies) in alcohol-drinking animals compared to water-drinking animals. Second, we optogenetically stimulated the burst firing of CINs and found that CIN excitation enhanced extinction of instrumental learning. According to these results we propose a model in which burst firing of CINs is important for the extinction of learned action-outcome contingencies by modulating striatal output; pause response of CINs is critical for acquiring new action-outcome contingencies by allowing dopamine-dependent plasticity. Current experiments aim to manipulate the pause response of CINs to further test our hypothesis. Our data indicate that chronic alcohol intake altered CIN firing pattern, leading to reduced flexibility. These results provide insight into reduced cognitive flexibility in alcohol use disorder.

Disclosures: Z. Huang: None. **J. Killingworth:** None. **M. Childs:** None. **J. Wang:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.01

Topic: H.08. Learning and Memory

Support: ARO MURI W911NF1810327

Title: Hippocampal place cell encoding during gap-crossing behaviors

Authors: *S. G. LASHKARI[1,2], B. M. WORONOWICZ[1,2], P. OZEL[3,4], B. KRISHNAN[3,5,6], J. J. KNIERIM[3,4,5], N. J. COWAN[1,2];
[1]Dept. of Mechanical Engin., [2]Lab. for Computat. Sensing and Robotics, [3]Zanvyl Krieger Mind/Brain Inst., [4]Solomon H. Snyder Dept. of Neurosci., [5]Kavli Neurosci. Discovery Inst. (NDI), [6]Dept. of Biomed. Engin., Johns Hopkins Univ., Baltimore, MD

Abstract: Place cells are neurons in the hippocampus that fire when an animal occupies a particular space within its environment. A subset of place cells, called splitter cells, encode the same location differently depending on the route taken to or from that location and the contingency of the reward (Wood et al. 2000). Although studies have investigated splitter cells in rats running on planar mazes, it remains unclear whether place cells exhibit splitter-like activity when trajectories are defined not by different 2D paths but by different complex behaviors available to the animal at a specific location in a common path. To investigate the firing properties of place cells during such complex locomotor behaviors, we studied Long-Evans rats (2 male, 1 female) as they ran across a linear track with an adjustable gap in the middle. The animals crossed the gap back and forth to get to the reward locations at each end of the track. When crossing, rats had to decide to either jump over the gap ("jumping") or leap into and out of the gap ("ditching"), choices that generated distinct 3D trajectories of the rat but similar 2D projections onto the experimental rig. The reward was not contingent upon the animals' current or prior decisions. Hippocampal recordings from CA1 revealed that place cells fired while the animals were airborne during jumping. For one animal, we recorded sessions with both jumping and ditching behavior and found that, in general, place cells encoded these trajectories differently. For example, in one session (n=41 CA1 cells), 18 place cells had firing field locations in the gap region, of which 14 displayed splitter-like behavior: 4 were strongly selective for jumping and 10 were strongly selective for ditching. Additionally, 6 cells fired immediately after the rat crossed the gap, of which 3 fired only after jumping, 2 fired only after ditching, and 1 fired in both cases but with different firing rates. In this study, we showed that place cells encode 3D trajectories when rats have the option to jump or ditch and that they fire while the animal is in midair. Place cells discriminated between jumping and ditching, including at the locations on the other side of the gap after the rat crossed it, thus demonstrating retrospective coding. This study provides new evidence of splitter cells differentiating 3D trajectories, even when the animal's choices do not affect the reward contingency, and further supports the notion that place cells adjust their firing properties to reflect the structure of the task at hand (Griffin et al. 2007).

Disclosures: S.G. Lashkari: None. **B.M. Woronowicz:** None. **P. Ozel:** None. **B. Krishnan:** None. **J.J. Knierim:** None. **N.J. Cowan:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.02

Topic: H.08. Learning and Memory

Support: NIH Grant R01MH118926

Title: Grid cell populations maintain their hexagonal firing patterns on an annulus

Authors: M. YIM, S. WALTON, ***K. HEDRICK**;
Southern Methodist Univ., Dallas, TX

Abstract: In an open two-dimensional environment, grid cells in the medial entorhinal cortex are known to be active in multiple locations, displaying a strikingly periodic hexagonal firing pattern covering the entire space. Both modeling and experimental data suggest that such periodic spatial representations may emerge from a continuous attractor network. According to this theory, grid cell activity in any stable 1D environment is a slice through an underlying 2D hexagonal pattern, which is supported by some experimental studies but challenged by others. Grid cells are believed to play a fundamental role in path integration, and so understanding their behavior in all environments is crucial for understanding the flow of information through the entorhinal-hippocampal system.
We analyze both simulated and experimental data from grid cells on an annulus. In our simulations, we find that the spatial frequencies associated with the grid periods are overrepresented in the 2D autocorrelation given a sufficiently large circular track, and they become more prominent when responses of a population of grid cells are combined. These predictions are consistent with our analysis of experimental grid cell data from an annulus. As expected, individual grid cells did not provide sufficient data for determining the underlying spatial activity pattern. To circumvent this, we pooled together population responses from grid cells in the same module, in which the activity pattern is characterized by a specific spacing and orientation. We observed a significant peak at the value of the track circumference in the autocorrelation of the linearized responses of the grid population, supporting an allocentric code in grid cell populations. Despite the limited and noisy nature of the data, the six-peak hexagonal pattern is recovered in the population autocorrelation, which has a gridness score significantly above controls. In addition, the orientations and period of the population response are consistent with the geometry of a hexagonal lattice. These results provide strong evidence that grid cells do maintain their regular spatial patterns on an annulus, and they also demonstrate how our novel statistical methods could be used to uncover structure in undersampled, noisy data.

Disclosures: M. Yim: None. **S. Walton:** None. **K. Hedrick:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.03

Topic: H.08. Learning and Memory

Support: NIH-NIMH Grant 1R01MH118926-01
NIH-NINDS Grant 5R01NS102537-0
ARO-MURI Grant 72929-EG-MUR-01

Title: Representation of Path-Integration Error in A Ring Attractor Model

Authors: *G. SECER[1,2], R. P. JAYAKUMAR[1,2], M. S. MADHAV[2,1], J. J. KNIERIM[2,3], N. J. COWAN[1,4];
[1]Lab. for Computat. Sensing and Robotics, [2]Zanvyl Krieger Mind/Brain Inst., [3]Solomon H. Snyder Dept. of Neurosci., [4]Dept. of Mechanical Engin., Johns Hopkins Univ., Baltimore, MD

Abstract: Animals can keep track of their position by integrating their velocities over time through path integration, a computation underlying the neural representation of position in an internally generated "cognitive map". Because path integration is intrinsically noisy, in the absence of external landmarks it accumulates error, causing the representation of position to drift. External landmarks correct drift by anchoring the cognitive map to the environment. Recent experiments in a circular VR apparatus have shown that path integration has a plastic gain that maps the magnitude of a rat's movement in physical space to that in its internal map. These experiments identified a new role of landmarks in providing a teaching signal for precise calibration of this gain (Jayakumar et al., 2019). How do landmarks modulate the dynamics of path integration to simultaneously provide drift correction and a teaching signal for the gain calibration?
To garner theoretical insight into this question, we studied the ring attractor model of path integration. Without imposing a mechanism for the gain calibration, we first sought universal conditions for stable, precise calibration of the gain and found that the instantaneous change in the gain must occur in the same direction as the product of the animal's velocity and path-integration error. Assuming Hebbian plasticity as the mechanism, we then showed that this stability condition translates to a fundamental requirement that some neurons in the network must encode the error via a rate code. This encoded error can be the neural correlate of a teaching signal provided by landmarks for gain calibration. As a preliminary test of this prediction, we analyzed place cells from 5 male, Long-Evans rats in an existing dataset in which gain calibration was shown (Jayakumar et al., 2019). In ~60% of these cells (248/400), firing rates were correlated with path-integration error, as measured by a slight drift of field locations relative to landmarks. Across sessions, the slope of this relationship was negatively correlated with the total error accumulated in a session. Finally, to close the loop between data and the model, we added afferent connections from visual landmark cells onto the angular rotation neurons in the model and reproduced the hypothesized rate code with the same negative correlation between slope and total error. This rate code is controlled by landmarks to simultaneously provide drift correction and a teaching signal for gain calibration. Although alternative explanations exist, our results suggest that an explicit rate code of path-integration error is a mechanism of landmark influence over place cells.

Disclosures: G. Secer: None. **R.P. Jayakumar:** None. **M.S. Madhav:** None. **J.J. Knierim:** None. **N.J. Cowan:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.04

Topic: H.08. Learning and Memory

Support: NIH-NINDS Grant 5R01NS102537-02
ARO-MURI Grant 72929-EG-MUR-01
NIH-NIMH Grant 1R01MH118926-01
Kavli Neuroscience Discovery Institute Fellowship (B.K.)

Title: Population Responses in Medial Entorhinal Cortex During Recalibration of Path Integration Gain

Authors: *B. KRISHNAN[1,2,3], G. SECER[2,4], F. SAVELLI[2,7], S. G. LASHKARI[5,4], R. P. JAYAKUMAR[2,4], K. L. WRIGHT[2], N. J. COWAN[5,4], J. J. KNIERIM[2,6,3];
[1]Dept. of Biomed. Engin., [2]Zanvyl Krieger Mind/Brain Inst., [3]Kavli Neurosci. Discovery Inst., [4]Lab. for Computat. Sensing and Robotics, [5]Dept. of Mechanical Engin., [6]Solomon H. Snyder Dept. of Neurosci., Johns Hopkins Univ., Baltimore, MD; [7]Dept. of Neuroscience, Developmental and Regenerative Biol., Univ. of Texas at San Antonio, San Antonio, TX

Abstract: Animals combine information from landmarks in the environment with self-motion inputs (such as vestibular cues, optic flow, proprioception, and motor copy) to navigate. The process of utilizing self-motion information to derive an estimate of position in a world-based ('allocentric') frame of reference is termed path integration. Path integration is governed by a gain factor that converts distances covered in the real world to distances traveled in the cognitive map. Recent recordings from CA1 place cells have shown that the path integration gain is a highly plastic variable, and its value can be recalibrated through a sustained conflict between landmarks and path integration (Jayakumar et al., 2019, Madhav et al., submitted). The synaptic modifications and functional changes in the firing properties of the network that bring about a recalibration of the path integration gain are unknown. The medial entorhinal cortex (MEC) is one of the primary inputs to the hippocampus and is believed to be the locus of the path integration computation (McNaughton et al., 2006), making it a good candidate to explore the neurophysiological mechanisms that underlie recalibration. We performed extracellular tetrode recordings from neurons in the MEC of Long-Evans rats (n=3) under conditions of persistent conflict between landmarks and self-motion inputs (Jayakumar et al., 2019). This conflict was produced by gradually rotating an array of landmarks in a virtual reality (VR) environment as a function of the rat's speed, producing the illusion that the rat was moving slower or faster than its actual speed. In 12/13 sessions where the firing fields of cells were locked to the position of the landmarks, the entire population of MEC cells recalibrated coherently, as demonstrated by a predicted change in the path integration gain when the landmarks were extinguished. However, in one session in which the firing locations of cells broke away from the landmarks, the MEC population split into two subgroups, with one subgroup (n = 3 cells) more closely following the landmarks than the other (n = 8 cells). These results show that MEC inputs to the hippocampus show a recalibration similar to place cells (Jayakumar et al., 2019) when the landmarks control the firing locations of cells. When the firing fields of cells dissociate from the array of landmarks, different subgroups of the MEC show varied responses, reflecting the overall functional diversity of the MEC. This heterogeneity could arise from each subgroup differentially weighting visual versus self-motion inputs.

Disclosures: B. Krishnan: None. **G. Secer:** None. **F. Savelli:** None. **S.G. Lashkari:** None. **R.P. Jayakumar:** None. **K.L. Wright:** None. **N.J. Cowan:** None. **J.J. Knierim:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.05

Topic: H.08. Learning and Memory

Support: NIA P01 AG009973
Johns Hopkins Science of Learning Institute

Title: Lateral entorhinal cortex-dependent associative memory deficit parallels spatial learning impairment in aged Long-Evans rats

Authors: Y. CHEN[1], A. BRANCH[2], C. SHUAI[1], M. GALLAGHER[2], J. J. KNIERIM[3]; [1]Neurosci., [2]Psychological and Brain Sci., [3]Zanvyl Krieger Mind/Brain Inst., Johns Hopkins Univ., Baltimore, MD

Abstract: The entorhinal cortex is a key structure of the medial temporal lobe system that mediates communication between the hippocampus and the neocortex to support spatial navigation and episodic memory. It has been postulated that the lateral entorhinal cortex (LEC) provides "content" of an experience by representing local sensory information and egocentric bearing to items, while the medial entorhinal cortex comprises a "context" pathway and encodes allocentric space as the framework of experience. In Alzheimer's disease, the LEC is affected early and profoundly, with significant accumulation of tau tangles. However, it remains poorly understood whether functional deficits in LEC arise in parallel with hippocampus-dependent memory decline with age and disease. The Long-Evans rat model of aging replicates the variable age-related cognitive outcome in humans on the Morris water maze, an assay of hippocampus-dependent spatial learning, and serves as a useful model system to assess the functional status of other brain regions in the context of hippocampal impairment. Notably, alterations in the molecular and synaptic markers of neuronal plasticity of LEC in aged, memory-impaired rats parallel the early deterioration of LEC in humans with age-related memory impairment. The present study aims to relate the age-related, hippocampus-dependent impairment in spatial memory with the functional integrity of LEC in supporting associative, episodic-like recognition memory. LEC-dependent memory was assessed in 43 male Long-Evans rats (9 young, 21 aged spatial-unimpaired [AU], 13 aged spatial-impaired [AI]) by testing their ability to recognize a novel object-place-context (OPC) configuration in a spontaneous exploration paradigm. Although both young and aged rats showed memory for integrated object, place, and context information, the amount of experience needed to form such associations differed. In an initial test after 6 exposures to two distinct OPC configurations, young and AU rats successfully recognized and preferentially explored objects of novel configurations, whereas AI rats preferred objects of familiar configurations, suggestive of an exploration strategy that fails to integrate context information. In a replication of this task with new OPC configurations, all age groups demonstrated intact associative recognition memory. The results demonstrated similarities in the behavioral expression of associative memory between young and AU rats and revealed a slower

temporal progression of information integration in AI rats, laying the groundwork for future studies to elucidate LEC alterations in information processing with cognitive aging.

Disclosures: Y. Chen: None. **A. Branch:** None. **C. Shuai:** None. **M. Gallagher:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); AgeneBio. **J.J. Knierim:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.06

Topic: H.08. Learning and Memory

Support: U01 NS111695
R01 NS039456

Title: Landmark vector cells in CA1 share a common directional signal with simultaneously recorded place cells

Authors: *Y.-Q. ZHOU[1], V. PULIYADI[2], X. CHEN[1,3], J. J. KNIERIM[1,2,4,5];
[1]Zanvyl Krieger Mind/Brain Institute, Johns Hopkins Univ., Baltimore, MD; [2]Dept. of Psychological and Brain Sciences, Johns Hopkins Univ., Baltimore, MD; [3]Brain Res. Ctr. and Dept. of Biology, Southern Univ. of Sci. and Technol., Shenzhen, China; [4]The Solomon H. Snyder Dept. of Neuroscience, Johns Hopkins Univ. Sch. of Med., Baltimore, MD; [5]Kavli Neurosci. Discovery Institute, Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: Place cells in the hippocampus increase their firing rates when an animal visits a specific location in an environment. The orientation of the hippocampal "cognitive map" appears to be controlled by the head direction system (Knierim, Kudrimoti et al. 1995). Deshmukh and Knierim reported a new form of hippocampal CA1 place cell called a landmark vector cell (LVC) that fires when the animal is located at a certain distance and angle relative to a local landmark (Deshmukh and Knierim 2013). However, how landmark vector cells respond when the salient, distal cues are rotated in the environment is unknown. To investigate how head direction system information is integrated into the LVCs, we employed miniscope technology to perform single-cell calcium imaging in the CA1 region of 8 freely moving, Long-Evans rats (5 males and 3 females) while they randomly foraged for food in a square platform containing 1-4 objects. A polarizing cue card was located on the curtains surrounding the recording chamber and it was rotated 90° between sessions. Consistent with previous studies, 7.3% (101/1366) of active CA1 pyramidal cells were identified as LVCs in calcium imaging recordings. The firing fields of place cells rotated relative to the center of the platform to follow the cue card rotation, whereas the firing fields of LVCs rotated by the same amount as the place cells but relative to the local nearby objects. We also observed ~12% of landmark vector cells (12/101) that encode both place cell information and the vector relationship between the rat's position and the local object,

in that they split their firing fields in two in the cue-rotation session; one subfield rotated around the platform center and the other subfield rotated around the object. Tracking the same neurons across multiple days also reveals that landmark vector cells can be generated from place cells. The firing fields of place cells and LVCs are both thought to be strongly coupled with the head direction system, but place cells were anchored in the world-centered frames while the LVCs were anchored in the landmark-centered frames. Taken together, these data demonstrated the capacity of CA1 neurons to encode both world-centered spatial information as well as animals' location relative to the local landmarks, with a common directional anchor presumably provided by the head direction system.

Disclosures: **Y. Zhou:** None. **V. Puliyadi:** None. **X. Chen:** None. **J.J. Knierim:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.07

Topic: H.08. Learning and Memory

Support: NIH R01 NS102537
NIH R01 MH118926
NIH R21 NS095075
Johns Hopkins University Discovery Award
Masason Foundation
Ezoe Memorial Recruit Foundation

Title: Phase code of place cells is maintained under hippocampal gain manipulation

Authors: *Y. SUEOKA[1,2], R. P. JAYAKUMAR[2,3,4], M. S. MADHAV[2,4,5], F. SAVELLI[2], N. J. COWAN[3,4], J. J. KNIERIM[1,2];
[1]Solomon H. Snyder Dept. of Neurosci., [2]Zanvyl Krieger Mind/Brain Inst., [3]Dept. of Mechanical Engin., [4]Lab. for Computat. Sensing and Robotics, [5]Kavli Neurosci. Discovery Inst., Johns Hopkins Univ., Baltimore, MD

Abstract: Spatial navigation is thought to be mediated in part by place cells in the hippocampus, which increase their firing rates when animals occupy specific locations of an environment. As the animal traverses through these place fields, the spiking activity of place cells advances to increasingly earlier phases of the local field potential (LFP) theta oscillation, a phenomenon known as theta phase precession. Thus, spike timing relative to LFP theta encodes the location of the animal within a place field. Theta precession results in the formation of theta sequences, where the spatial order of place cells with overlapping fields is re-instantiated temporally within a theta cycle. However, it is unknown whether and how phase coding adapts to a constantly changing environment. To investigate the dynamics of phase coding, we recorded the activity of place cells (n = 261) from hippocampal CA1 as 5 male, Long-Evans rats ran around a circular

track inside a planetarium-style VR environment (the "Dome"). An array of visual landmarks was projected onto the wall of the dome and was rotated as a function of the rat's movement. The motion of the visual landmarks was controlled by the experimental gain G, which specified the ratio of the animal's motion in the landmark frame to its motion in the lab frame. In $G > 1$ sessions, landmarks moved in the direction opposite to the rat's movement, causing an illusion that the rat was moving faster than it actually was; in $G < 1$ sessions, landmarks moved in the same direction as the rat, causing the rat to perceive its motion as slower than it was. In 40/51 sessions, place fields were stable in the rotating landmark frame, enlarging or shrinking relative to the rat's physical position on the track (Jayakumar et al., 2019). In these sessions, place cells maintained their theta-phase coding in the landmark frame. The overall relationship between the theta phase and the position in the landmark frame was constant across gain values, even though the size of place fields in the lab frame of reference was altered dramatically. The stable phase code was achieved by place cells flexibly adjusting their theta-modulated burst frequencies (precession rate ~ 0.16G); place cells decreased their burst frequencies and slowed down phase precession when the fields were enlarged and increased their burst frequencies when the fields were shrunk. As a result, theta sequences were preserved across different gain values. This study elucidates the ability of place cells to flexibly modulate their spiking activity to maintain phase coding under altered gain conditions and points to a possibly dominant role of intrinsic place cell sequence dynamics in stabilizing the coding scheme.

Disclosures: **Y. Sueoka:** None. **R.P. Jayakumar:** None. **M.S. Madhav:** None. **F. Savelli:** None. **N.J. Cowan:** None. **J.J. Knierim:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.08

Topic: H.08. Learning and Memory

Support: NIH R01 NS102537
Kavli Neuroscience Discovery Institute Distinguished Postdoctoral Fellowship
Johns Hopkins University Discovery Award

Title: Closed-loop control and recalibration of place cells by optic flow

Authors: *M. S. MADHAV[1,2,3,5,6], R. P. JAYAKUMAR[1,2,4], B. Y. LI[1], K. L. WRIGHT[1], F. SAVELLI[1], J. J. KNIERIM[1,3], N. J. COWAN[2,4];
[1]Mind/Brain Inst., [2]Lab. for Computat. Sensing and Robotics, [3]Kavli Neurosci. Discovery Inst., [4]Dept. of Mechanical Engin., Johns Hopkins Univ., Baltimore, MD; [5]Sch. of Biomed. Engin., [6]Djawad Mowafaghian Ctr. for Brain Hlth., Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Hippocampal place cells are an ideal system to investigate the interaction between sensory input, endogenous neural dynamics, and behavioral output, as they are influenced by

both self-motion (idiothetic) signals and by external sensory landmarks. To continuously update its position on an internal "cognitive map", the hippocampus performs path integration of self-motion signals over time. In the absence of stable, external landmarks, path integration can accumulate error and cause the internal representation of position to drift relative to the external environment. We previously demonstrated that, in addition to their known roles in preventing and/or correcting path-integration error, external landmarks can be used as a putative teaching signal to recalibrate the gain of the path integration system (Jayakumar et al., 2019). However, it remains unclear whether idiothetic cues, such as optic flow, exert sufficient influence on the cognitive map to enable recalibration of path integration, or if instead an unambiguous allocentric frame of reference, anchored by polarizing landmark information, is essential for path integration recalibration. Here, we use principles of control theory to demonstrate systematic control of place fields by pure optic flow in freely moving animals. We recorded place cell activity from the CA1 region of 2 male and 2 female Long Evans rats as they ran on a circular track inside an immersive virtual reality system. We developed a closed-loop integral controller that adjusts the gain of the optic flow signal based on real-time decoding of the gain of the hippocampal spatial map (H), in order to drive it to a desired value. In 18/25 sessions from 4 rats, the closed-loop controller was able to clamp the value of H at or near the desired value. Control was much stronger when the desired value was greater than the baseline value of H compared to when it was below baseline, demonstrating an asymmetry in the ability to control H to values greater than vs. less than its normal value. For the 3 rats with strong closed-loop control, we tested for recalibration of the path integrator gain by measuring H after the stripes were extinguished. In all 3 rats, there was a linear relationship between H measured after the stripes were extinguished and the desired value, showing that optic flow cues alone could recalibrate the path integrator gain. This finding demonstrates that the brain continuously rebalances the influence of conflicting idiothetic cues to fine-tune the neural dynamics of path integration, and that this recalibration process does not require a top-down, unambiguous teaching signal from landmarks.

Disclosures: **M.S. Madhav:** None. **R.P. Jayakumar:** None. **B.Y. Li:** None. **K.L. Wright:** None. **F. Savelli:** None. **J.J. Knierim:** None. **N.J. Cowan:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.09

Topic: H.08. Learning and Memory

Support: R01 NS102537
Johns Hopkins University Kavli Neuroscience Discovery Institute Postdoctoral Fellowship
Johns Hopkins University Discovery Award

Title: Coherent path integration recalibration observed in simultaneously recorded place cell and head direction cell populations

Authors: *R. P. JAYAKUMAR[1,2], Y. SUEOKA[1,3], M. FERREYROS[3], B. Y. LI[1], M. S. MADHAV[1,2,4], N. J. COWAN[2,5], J. J. KNIERIM[1,3,4];
[1]Zanvyl Krieger Mind/Brain Institute, Johns Hopkins Univ., Baltimore, MD; [2]Lab. for Computat. Sensing and Robotics, Johns Hopkins Univ., Baltimore, MD; [3]Solomon H. Snyder Dept. of Neuroscience, Johns Hopkins Univ., Baltimore, MD; [4]Kavli Neurosci. Discovery Institute, Johns Hopkins Univ., Baltimore, MD; [5]Dept. of Mechanical Engin., Dept. of Mechanical Engineering, Johns Hopkins Univ., Baltimore, MD

Abstract: The spatial map in the rodent hippocampal formation combines information from both landmarks and self-motion cues to create a representation of the animal's environment. We previously used a planetarium-style, virtual reality dome (Madhav et al., 2022) to examine the relative influence of these two classes of information on the firing fields of hippocampal place cells (Jayakumar*, Madhav* et al., 2019). A persistent conflict between landmarks and path integration inputs was introduced by moving the array of landmarks as a function of the rat's speed, producing the illusion that the rat was moving slower or faster than its actual speed. As a result of this persistent cue conflict, the brain recalibrated the relationship between self-motion cues perceived by the rat during movement and the updating of its position on its hippocampal "cognitive map". This relationship, called the path integration gain, was thus shown to be a plastic variable learned via feedback from salient landmark cues over relatively short time scales. Head direction cells are thought to be involved in creating the directional component of the velocity vector that is integrated in path integration. Thus, to begin to address the network mechanisms that underlie the recalibration of path integration gain, we performed extracellular tetrode recordings simultaneously from neurons in the CA1 region of the hippocampus and head direction cells in the retrosplenial cortex and/or the anterior dorsal thalamic nucleus of Long-Evans rats (n=3, 2 male, 1 female) under conditions of persistent conflict between landmarks and self-motion inputs. The head direction and place cell populations stayed locked to the moving landmark frame of reference in 17/23 sessions. When the landmarks were extinguished in these 17 sessions, the head direction cells showed recalibration of the path integration gain, similar to the simultaneously recorded CA1 place cells. These results demonstrate a coherent response between place cells and head direction cells and open the possibility that the recalibration demonstrated by place cells may be, at least in part, a reflection of plasticity mechanisms in head direction cells or other networks that are upstream of the hippocampus.

Disclosures: R.P. Jayakumar: None. **Y. Sueoka:** None. **M. Ferreyros:** None. **B.Y. Li:** None. **M.S. Madhav:** None. **N.J. Cowan:** None. **J.J. Knierim:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.10

Topic: H.08. Learning and Memory

Support: R01NS039456
R35NS116843
R35NS097370
R21NS093772
2014-MSCRFF- 0711
Research award from the Johns Hopkins University Brain Science Institute

Title: Properties of adult-born granule cells and their impact on local circuitry in freely behaving mice

Authors: *S. KIM[1,6,7], K.-J. YOON[6,7], D. GOODSMITH[1,8,10], S. TEMME[1,6,7], N.-S. KIM[6,7], D. BERG[6,7], W. HUANG[6,7], F. ZHANG[1], F. MORIYA[1], Z. ZHANG[1], Y. SU[1], Y. ZHOU[1], N. KUMAR[1], J. J. KNIERIM[8,9,10], K. M. CHRISTIAN[1], G.-L. MING[1,2,3,4], H. SONG[1,2,3,5]; [1]Dept. of Neurosci. and Mahoney Inst. for Neurosciences, [2]Dept. of Cell and Developmental Biol., [3]Inst. for Regenerative Med., [4]Dept. of Psychiatry, [5]The Epigenetics Inst., Perelman Sch. for Medicine, Univ. of Pennsylvania, Philadelphia, PA; [6]Inst. for Cell Engin., [7]Dept. of Neurol., [8]The Solomon H. Snyder Dept. of Neurosci., [9]Kavli Neurosci. Discovery Inst., Johns Hopkins Univ. Sch. of Med., Baltimore, MD; [10]Zanvyl Krieger Mind/Brain Inst., Johns Hopkins Univ., Baltimore, MD

Abstract: In the mammalian brain, constitutive neurogenesis takes place in the dentate gyrus (DG) of the hippocampus throughout life. Adult-born neurons make a significant contribution to information processing and hippocampal memory, including encoding, retrieval, and memory consolidation. Acute hippocampal slice recordings suggest that adult-born neurons exhibit increased excitability and plasticity from roughly 4-6 weeks after their birth. However, little is known about the properties of adult-born neurons in vivo and how they influence local circuitry in the DG, due to the technical challenges of identifying adult-born neurons during extracellular single unit recordings. By combining optogenetics with in vivo recording, we identified immature newborn neurons of known cellular age in freely behaving mice and found that newborn neurons are more active than mature granule cells both during behavior and sleep. Interestingly, newborn neurons up to 11 weeks old still exhibited elevated firing rates and the proportion of units involved in spatial representation is higher for newborn neurons than mature granule cells. When the mice were exposed to different environments, mature granule cells and immature newborn neurons remapped to a similar extent. Further, we recorded from DG cells while optogenetically inactivating adult-born neurons at different maturational timepoints. Inactivation of adult-born neurons between 4 and 11 weeks old affected the neural activity of mossy cells. Adult-born neurons older than 22 weeks had a minimal impact on mossy cell activity, suggesting an age-dependent role of this population on local circuit activity. Taken together, our electrophysiological recordings of single dentate gyrus cells in behaving mice support predictions of higher activity levels in adult-born neurons and reveal a modulatory role of this population on the activity of mossy cells.

Disclosures: S. Kim: None. **K. Yoon:** None. **D. GoodSmith:** None. **S. Temme:** None. **N. Kim:** None. **D. Berg:** None. **W. Huang:** None. **F. Zhang:** None. **F. Moriya:** None. **Z. Zhang:** None. **Y. Su:** None. **Y. Zhou:** None. **N. Kumar:** None. **J.J. Knierim:** None. **K.M. Christian:** None. **G. Ming:** None. **H. Song:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.11

Topic: H.08. Learning and Memory

Support: NIH T32 NS007292
NIH Grant 5R01MH120228

Title: Hippocampal-prefrontal circuit mechanisms that support inferential reasoning

Authors: *B. S. PORTER, C. SHI, S. JADHAV;
Dept. of Psychology, Brandeis Univ., Waltham, MA

Abstract: Inference is a remarkable cognitive ability that allows animals to deduce novel relationships between bits of information without having to explicitly learn their relationships. Previous studies using humans and rodents have demonstrated that the hippocampus and prefrontal cortex are critical to successful inferential reasoning. However, the neural systems level processes that underlie inference are not well understood. A possible mechanism to create new relationships between stimuli are hippocampal sharp wave ripples (SWRs). During SWR events, hippocampal neuronal sequences, coordinated with prefrontal neural ensembles, are reactivated during periods of rest. We hypothesize that SWRs could reactivate distinct representations in order to form novel associations between them and thus support inference. In order to test our hypothesis, we developed a rodent spatial transitive inference task in order to speed up training times compared to odor-based tasks and facilitate the analysis of SWR replay events. Single unit and local field potential (LFP) activity was recorded with custom high-density ShuttleDrives consisting of 64-tetrodes. Male and female rats (Sprague Dawley, 3-12 months old, N=7) readily learned the transitive inference task and demonstrated clear inference-like behaviors when tested. Specifically, rats were trained to discriminate overlapping pairs of stimuli based on a task value set (A>B>C>D>E>F). Rats were then tested on novel combinations of stimuli and successfully inferred the most valuable stimulus. Hippocampal place cells had diverse selectivity profiles with some cells firing on single arms while others had fields on multiple arms. Sharp wave ripples events were prominent on the track during inference testing and showed clear reactivations of neural ensembles. Overall, these results demonstrate a novel space-based transitive inference task for use in rodents that leverages their innate spatial navigation abilities. The task paves the way for future studies on the hippocampal-prefrontal mechanisms that support inferential reasoning.

Disclosures: B.S. Porter: None. **C. Shi:** None. **S. Jadhav:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.12

Topic: H.08. Learning and Memory

Support: NIH Grant R01 MH112661
NIH Grant R01 MH120228

Title: Distinct geometries of hippocampal and prefrontal representations for memory generalization

Authors: *W. TANG, J. D. SHIN, S. P. JADHAV;
Psychology, Brandeis Univ., Waltham, MA

Abstract: The ability to generalize learned knowledge for guiding behavior in novel contexts is thought to rely upon the interactions between the hippocampus and prefrontal cortex (PFC; *Schlichting and Preston, 2015; Preston and Eichenbaum, 2013*). Emerging evidence suggests that both regions maintain representations of task space (*Zielinski et al., 2019; Hasz and Redish, 2020; Sauer et al., 2022; Tang and Jadhav, 2022*). However, the intrinsic structures of these representations and their dynamics across contexts have not been well characterized, leaving it unclear whether and how they differ in their functional contributions to memory generalization. Here, we simultaneously recorded hippocampal and prefrontal neural ensembles (n = 214 and 215 cells, respectively) as rats (n = 5) generalized behaviors of a hippocampal-prefrontal dependent memory task across different environments. We show that although the hippocampus and PFC both encode task information with a manifold-like representation, these areas have major differences in their representational geometry. Specifically, the geometry of the PFC representation enabled generalization across locations and contexts where similar behaviors are task-appropriate, whereas the hippocampal representation provided characteristics of distinct spatial contexts. Furthermore, the context-general patterns in PFC were prominently reactivated with context-specific hippocampal ensembles during sharp-wave ripples (SWRs). Notably, following training in two different environments, the SWR events replaying recent and remote experiences were both enhanced and overlapped substantially in the hippocampal-prefrontal network. Together, these findings elucidate how task knowledge is structured and transformed into representations with distinct geometries in the hippocampal-prefrontal network to concurrently support memory specificity and generalization across experiences.

Disclosures: **W. Tang:** None. **J.D. Shin:** None. **S.P. Jadhav:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.13

Topic: H.08. Learning and Memory

Support: NIH Grant R01 MH112661
NIH Grant R01 MH120228

Title: Nonlocal coding in hippocampal-prefrontal circuits during cue-guided and memory-guided navigation

Authors: *R. YOUNG[1], S. P. JADHAV[2];
[1]Biol., [2]Psychology, Brandeis Univ., Waltham, MA

Abstract: Navigation is an active process that requires combining knowledge of "here and now" with upcoming and former states. Previously, it was noted that animals encode a span of spatiotemporal locations in the hippocampus (HPC) that bias toward the future (Dotson & Yartsev, 2021). More broadly, the hippocampus functionally coordinates with the prefrontal cortex (PFC), which influences firing around nonlocal hippocampal (HPC) representations (Yu & Frank, 2020; Berners-lee, Wu & Foster, 2021). Both HPC and PFC are required for memory-guided navigation, but it is unclear whether different task demands-- such as cue- and memory-guided states --reorganize nonlocal coding and hippocampal-prefrontal coordination thereof . To investigate this question, we implanted 64 tetrode drives in rats performing a task exhibiting both cue- and memory-guided phases in a 2-d maze. Animals were shown a rewarded location for two consecutive trials and then tested with two uninstructed trials to ascertain their memory of the location, with the reward location switching randomly in subsequent trial blocks. Surprisingly animals exhibited different distributions of nonlocal coding in hippocampal and prefrontal ensembles under these cue- and memory-guided regimes. This may imply flexibility in predictive and postdictive coding under various task demands.

Disclosures: R. Young: None. **S.P. Jadhav:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.14

Topic: H.08. Learning and Memory

Support: NIH NINDS T32 (NS 7292-33)
Swartz Foundation
Brandeis Innovation SPROUT grant
Smith Foundation Odyssey Award
NIH/NIMH R01 MH112661
NIH/NIMH R01 MH120228

Title: Subiculum and CA1 activity in rats during learning of a novel complex navigation task

Authors: *J. M. OLSON, C. W. REES, S. P. JADHAV;
Brandeis Univ., Waltham, MA

Abstract: Memory formation and recall underpin cognitive abilities such as decision-making and spatial navigation. Intact hippocampal function is necessary for memory encoding and retrieval, and coordination with extrahippocampal regions through rhythmic network patterns such as sharp-wave ripples and theta oscillations are essential for spatial learning and decision-making. During sharp-wave ripples, hippocampus "replays" memory sequences, condensing neural activity patterns from behavioral timescales into a timescale amenable to Hebbian plasticity. Hippocampal research has focused on hippocampal subregion CA1 as the main hippocampal output, but anatomical connectivity suggests subiculum (SUB) is equally vital. SUB receives strong inputs from CA1 and entorhinal cortex, and its outputs largely mirror and complement those of CA1. SUB also has extensive outputs to downstream regions such as prefrontal cortex and nucleus accumbens. Despite extensive anatomical connectivity, little is known regarding CA1/SUB coordination during memory-guided navigation. We hypothesize that during rhythmic network activity, neurons in SUB and CA1 with overlapping spatial firing fields will be active, jointly "replaying" previous experience and linking categorical SUB representations with CA1 ensembles encoding specific experiences. Here, we recorded dorsal CA1 and SUB single cell activity using *in vivo* electrophysiology while adult male Long-Evans rats navigated a novel complex environment. Through the use of dynamic barrier locations, we adapted the available paths to rewards over learning. SUB and CA1 ensembles do indeed show coordination during sharp-wave ripples during learning. Differences in field patterns between SUB and CA1 were also observed. Overall, this work adds to a growing body of evidence that SUB plays an important part in hippocampal output during navigation.

Disclosures: J.M. Olson: None. **C.W. Rees:** None. **S.P. Jadhav:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.15

Topic: H.08. Learning and Memory

Support: NIH Grant R01 NS104818
NIH Grant T90 DA032435
NIH Grant R01 MH112661
NIH Grant R01 MH120228

Title: Preexisting randomly clustered recurrent circuit structure can lead to place field activity and correlated preplay

Authors: *J. BREFFLE[1], H. GERMAINE[1], S. P. JADHAV[2], P. MILLER[3];
[1]Brandeis Univ. Grad. Neurosci. Program, Waltham, MA; [2]Psychology, [3]Biol., Brandeis Univ., Waltham, MA

Abstract: The recurrent connectivity of the CA3 region of the hippocampus is thought to be important for storing patterns of activity corresponding to spatial and episodic memories, but it is not well understood how the preexisting structure of the network shapes and constrains the expression of future neural activity. During movement through an environment, the firing of place cells in the hippocampus correspond to an animal's current location. During pauses in movement and rest, the hippocampus replays sequences of place cells that correspond to spatial trajectories through the environment. The hippocampal place code changes with each different environment. Although plasticity modifies this representation with experience, a new place code is immediately expressed in a novel environment and hippocampal replay has been observed after single laps on novel tracks. Additionally, correlations to future place codes can be found in spontaneous activity during rest, a process known as preplay. This suggests that the preexisting structure of the network shapes the future place code. Here, we demonstrate how an untrained randomly connected, recurrent clustered network can produce preplay events correlated with a place field code generated by a combination of minimally placed landmarks, an environmental context signal, and history-dependent activity. We simulated the model to represent the CA3 region of the hippocampus, with recurrent connections that were randomly clustered. All clusters in the network were comprised of randomly assigned excitatory cells, such that cells could inhabit multiple clusters, while connections between excitatory cells depended, probabilistically, on shared cluster membership. Inhibitory cells provided feedback inhibition through random but non-clustered connections. The only spatially tuned input to the network was via two location cues that monotonically varied with distance from the two ends of a linear track. All excitatory neurons received input from both location cues with randomly selected strengths. We find that the recurrent connections shape activity such that place field peaks tile the track, despite the simplicity of the spatial input. The relative strength of the feed forward and recurrent inputs must be balanced to preserve both place field-like spatial receptive fields and spatially correlated preplay events. This simple model demonstrates how preexisting structure can constrain the expression of spatially modulated neural activity in a manner that generates spatially correlated spontaneous burst events.

Disclosures: **J. Breffle:** None. **H. Germaine:** None. **S.P. Jadhav:** None. **P. Miller:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.16

Topic: H.08. Learning and Memory

Support: NIH Grant R01 MH112661
NIH Grant R01 MH120228

Title: Coordination of the hippocampus, prefrontal cortex and ventral tegmental area during learning and reward-guided behavior

Authors: *M. DING, S. JADHAV;
Brandeis Univ., Waltham, MA

Abstract: Goal-directed behavior arises through an interplay of contextual representation and action outcomes, e.g., reward. The hippocampus (HPC) and medial prefrontal cortex (mPFC) are known for their complementary functions in spatial working memory and navigation. The dopaminergic (DA) neurons in the ventral tegmental area (VTA) represent reward prediction error (RPE) and contribute to reinforcement learning. Yet it remains unclear how reward information is associated with specific context representation for model-based learning, and further utilized to support decision-making processes in dynamic environments. We hypothesize that VTA, HPC and mPFC coordinate during hippocampal sharp-wave ripple (SWR) associated replay to fulfill the function of value assignment to specific trajectories, and assist in navigational planning. In support of this idea, VTA neurons are known to be activated during hippocampal replay (Gomperts et al., 2015), and reward changes are known to influence reverse replay occurrence (Ambrose et al., 2016; Singer & Frank, 2009). In addition, our prior work has shown that interregional HPC-PFC replays are biased toward actual past and future trajectories, (Shin et al., 2019), providing a possible mechanism to associate trajectory reactivation with reward. To test our hypothesis, animals were trained to learn and perform a rule-switching W maze task, during which the change of rules was only indicated by reward outcomes without external cues. We recorded from the three brain regions simultaneously in behaving animals and optogenetically tagged DA (TH+) neurons in the VTA. TH+ neurons responded robustly to different reward outcomes. As RPE decreased along with performance improvement, the firing rates of the majority of TH+ neurons decreased. A subpopulation of VTA neurons showed firing rate modulation during SWRs, suggesting heightened communication between the HPC and VTA during replay. Further experiments and analysis will be performed to investigate how interregional communication is facilitated at a fine time scale to support learning and goal-directed behavior.

Disclosures: M. Ding: None. **S. Jadhav:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.17

Topic: H.08. Learning and Memory

Support: HIMH/NIH Grant R01 MH119102

Title: A raphe-hippocampus glutamatergic pathway controls memory specificity during consolidation

Authors: *W. HUANG, D. V. WANG;
Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: Maintenance of memory specificity is critical for our daily life while loss of memory specificity or overgeneralization can lead to abnormal or pathological memories, such as post-traumatic stress disorder (PTSD). Memories encoded during waking need to go through memory consolidation, mostly during sleep, before they become stabilized and resistant to interference. This process is slow that takes days, weeks, or even longer. Therefore, multiple memories are consolidated inevitably overlapping in time or in parallel. Yet, it remains unclear how memory specificity is maintained during consolidation of multiple memory traces. We have previously shown that the median raphe (MnR) plays a key role in regulating hippocampal ripple activity, a fast oscillation (~200 Hz) that occurs predominantly during sleep and is critical for memory consolidation. In particular, one subtype of MnR neurons fires synchronously at the termination of ripple events and suppresses subsequent ripple occurrence. In the current study, we aim to determine the identity of this MnR neural type and its specific role during memory consolidation. We hypothesized its activity is essential for controlling memory specificity during consolidation by separating ripples and associated reactivation contents. Combining electrophysiology recording and optogenetics, we confirmed that the MnR Vglut3 neurons play a causal role in up- or down-regulation of ripple activity. Using the ripple trigged closed-loop approach during slow-wave sleep, we precisely increased or decreased hippocampal ripple density via Opto-stimulation of Vglut3 neurons. In our ongoing experiments, we employ a contextual fear discrimination task and aim to determine if the closed-loop Opto-inhibition of MnR Vglut3 neurons during the post-training sleep leads to a failure in subsequent discrimination of the fear and neutral contexts. These results suggest that the MnR Vglut3 neurons play an important role in regulating hippocampal ripple activity and controlling memory specificity during consolidation.

Disclosures: W. Huang: None. **D.V. Wang:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.18

Topic: H.08. Learning and Memory

Support: NIMH/NIH R01 MH119102

Title: An assembly of retrosplenial cortical neurons is positioned to orchestrate memory consolidation

Authors: *A. N. OPALKA, K. J. DOUGHERTY, D. V. WANG;
Neurobio. and Anat., Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: Memory consolidation is a process where newly learned experiences are incorporated into pre-existing brain networks to become permanent. Understanding how memories are

consolidated is important for designing improved interventions for amnestic, psychiatric, or pathologic states. It is well-established that slow-wave sleep is essential for this consolidation process, where delta oscillations (1-4 Hz) coordinate communications among various cortical, hippocampal, and thalamic regions. These delta oscillations have periods of Up and Down states, with the latter previously thought to represent complete cortical silence; however, new evidence suggests that these silent Down states may serve important functions for information exchange during consolidation. Here, we study a key memory processing region, the retrosplenial cortex (RSC), in the delta oscillation and memory consolidation. We employed multi-channel *in vivo* electrophysiology and advanced data analyses, including cell assembly analysis, to study RSC neuronal activity in freely behaving mice during consolidation. We discovered that the RSC contains a discrete assembly of neurons (~20%) that started to fire at the Down state and reached maximal firing at the Down-to-Up transition. Therefore, we termed these RSC neurons the Down state assembly (DSA), and the remaining RSC neurons as non-DSA. This DSA activity preceded the activity of RSC non-DSA, hippocampal, and other cortical neurons, leading to our hypothesis that the DSA initiates and maintains subsequent non-DSA network activity crucial for memory consolidation. In support of this, DSA activity increases during consolidation of a contextual fear experience. Our ongoing experiments aim to explore and manipulate direct inputs to the RSC that can selectively target the DSA. Specifically, we explore two major RSC inputs, one from the anteroventral thalamic Shox2 neurons, and the other from the claustrum, both of which have been implicated in regulating neural oscillations and memory functions. Altogether, these findings will provide insight on the mechanisms of memory consolidation, which could shed light on how detailed information is integrated and organized into brain networks, along with better interventions for memory disorders.

Disclosures: **A.N. Opalka:** None. **K.J. Dougherty:** None. **D.V. Wang:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.19

Title: WITHDRAWN

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.20

Topic: H.08. Learning and Memory

Support: NIMH/NIH (R01 MH119102)

Title: Opposite dorsal versus ventral Lateral Septum firing to fear and other contexts

Authors: *C. A. RIZZI-WISE, D. V. WANG;
Neurobio. and Anat., Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: The ability to regulate internal emotional states is necessary to express appropriate behaviors in most situations. This is especially true in fearful or dangerous contexts where an internal state of fear informs what behavior would be most conducive for a higher chance of survival. Despite this central idea, the exact circuitry involved in this is not fully understood; however, an emerging brain region of importance is the Lateral Septum (LS). A specific function of the LS remains elusive as it has been implicated in a variety of behaviors due to its extensive connections to multiple brain regions. Here we investigated hippocampal-to-LS projections, one of the most prominent LS connections. We found that manipulation of the dorsal (d) versus ventral (v) hippocampal CA1 projections to LS subregions led to divergent behaviors during a contextual fear paradigm: the dCA1-to-dLS pathway promoted fear while the vCA1-to-vLS pathway suppressed it. Using in vivo electrophysiology, we found neuronal populations, within both the dLS and vLS subregions, that fired specifically to a fear associated context compared to other contexts that evoke either a neutral or positive valence. Between the dLS and vLS, these populations had distinct, identifiable firing properties where dLS neurons tended to increase and vLS neurons tended to decrease their firing rate in response to contextual/affective changes. Our findings demonstrate that the LS plays a critical role in differentially regulating fear due to anatomically different hippocampal inputs in addition to opposing firing rates in response to fearful and other affective contexts.

Disclosures: C.A. Rizzi-Wise: None. **D.V. Wang:** None.

Poster

571. Learning and Memory: Hippocampal: Cortical Interactions II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 571.21

Topic: H.08. Learning and Memory

Support: R01 MH119102

Title: Bidirectional communication between the Anterior Cingulate Cortex and Dorsal Hippocampus CA1 deep cells during sharp-wave ripples facilitates memory consolidation

Authors: *A. HALL[1], D. V. WANG[2];
[1]Drexel Univ. Col. of Med. Neurosci. Program, Philadelphia, PA; [2]Neurobio. and Anat., Drexel Univ., Philadelphia, PA

Abstract: Systems consolidation involves the transformation of impermanent, hippocampus-dependent memories into permanent, long-term memories stored throughout cortical regions. The underlying mechanism which facilitates systems memory consolidation remains limited. Sharp-wave ripples (SPWs; ripple events), short (50- 100ms), 100-250Hz neural oscillations originating from the dorsal CA1 (dCA1) pyramidal cell layer of the hippocampus during slow wave sleep (SWS) have emerged as a possible mediating factor of system consolidation. These oscillations have been shown to reactivate hippocampal cells and cortical engrams (neural units encoding memory) during SWS in the same manner seen during wakefulness; this reactivation is necessary for memory consolidation. Previous evidence has implicated the anterior cingulate cortex (ACC) as potential cortical target with correlated communication between the ACC and hippocampal dCA1 cells during SWS SPWs. This has been believed to be a feature of a dCA1 → ACC pathway, however, we have recently found, through dual-site electrophysiology, evidence to suggest that this communication is bidirectional. Using a general linear model, we have discovered that ACC activity preceding SPWs predicts the activity of dCA1 cells during subsequent SPWs. Utilizing a contextual fear learning paradigm, we have found that the predictive strength of ACC neuronal activity on dCA1 activity, as well as previously reported correlated activity, is increased during post-learning SPWs as compared to pre-learning SPWs suggesting a potential role of ACC ↔ dCA1 activity in memory consolidation. Interestingly, we discovered that ACC neural activity during SPWs specifically correlates to and predicts the activity of deep cells, a sublayer of dCA1 pyramidal cells. Deep cells are thought to be involved in ripple dependent learning, exhibiting dynamic differences in activity before and after learning during SPWs. Together, these findings establish a bidirectional pathway of communication between the ACC and dCA1 deep cells during SPWs which adapts to fear learning. These results provide foundational evidence which demonstrates a novel memory-related pathway that may be critical for systems consolidation.

Disclosures: A. Hall: None. **D.V. Wang:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.01

Topic: H.08. Learning and Memory

Support: NSF CAREER IOS-2145814

Title: Medial Entorhinal Cortex is necessary to learn novel temporal Delayed Non-Match to Sample task

Authors: *E. R. BIGUS[1], A. A. CHAGOVETZ[2], J. G. HEYS[2];
[1]Interdepartmental Neurosci. Program, [2]Dept. of Neurobio., Univ. of Utah, Salt Lake City, UT

Abstract: The medial temporal lobe (MTL) of the brain encodes individual experiences into memory so they can be flexibly used to guide future behavior. Although experiences and memories are rooted in a sense of time, it remains unknown how the MTL memory system encodes elapsed time on the order of seconds to minutes, called interval time. Numerous recent studies have uncovered a role of the medial entorhinal cortex (MEC) in interval timing tasks, suggesting this region helps the MTL track time. However, these studies also demonstrate that MEC is not always necessary for performance of interval timing tasks, raising uncertainty about the role of MEC in timing. To date, it is unknown under what circumstances MEC is necessary for interval timing. To address this gap, we propose that multiple memory systems are capable of driving interval timing behavior, depending upon timing demands. MTL structures including MEC may be necessary when animals quickly and flexibly learn and recall durations, while tasks requiring more rigid temporal responses may instead be mediated by a procedural memory system. We are examining this framework by testing the hypothesis that MEC is necessary for interval timing when animals must quickly and flexibly encode durations in a manner suited to the demands of the MTL memory system. Due to the lack of behavioral paradigms designed for studying MTL timekeeping in mice, we first established a novel temporal Delayed Non-Match to Sample (tDNMS) task in which mice quickly and relatively flexibly make decisions based upon relative stimulus durations. To test the necessity of MEC, we then trained mice expressing either a control virus or inhibitory DREADDs on the tDNMS task (n = 16 control mice; n = 15 DREADD mice; a mix of male and female C57BL/6). Administration of the DREADD agonist DCZ selectively impaired the ability of DREADD mice to learn the tDNMS task but did not affect performance post-learning. Results show a clear necessity of MEC in learning the tDNMS task, supporting the hypothesis that MEC is necessary for timing when behavioral demands are appropriate. In parallel we have used cellular-resolution Ca2+ imaging experiments to determine the neural dynamics in MEC that may underlie tDNMS performance. Our preliminary results indicate that populations of MEC neurons fire in regular, time-locked sequences across trials. Interestingly, we find that many time cells display context-dependent timing activity that is able to decode trial-type. Together, this work supports the overarching hypothesis that MEC plays a role in interval timing behavior through regular sequential neural activity across populations of MEC time cells.

Disclosures: **E.R. Bigus:** None. **A.A. Chagovetz:** None. **J.G. Heys:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.02

Topic: H.08. Learning and Memory

Support: NSF DGE-1839285
NIH R01-MH115697
NIH R01-DC017687

NSF BCS-1439267
Whitehall Foundation 2010-05-84

Title: Prefrontal cortex represents nonspatial event sequences as an ordinal schema

Authors: *K. W. COOPER[1], M. P. SARAF[1], B. SHAHBABA[2], G. A. ELIAS[1], N. J. FORTIN[1]; [1]Dept. of Neurobio. & Behavior, [2]Dept. of Statistics, Univ. of California, Irvine, Irvine, CA

Abstract: The prefrontal cortex (PFC) plays a fundamental role in the temporal organization of memory and behavior, though the specific neural mechanisms supporting this capacity remain unclear. Previous research suggests PFC activity contains information about the ordinal position within a sequence of events. We hypothesize this form of coding reflects a schematic representation of the key task structure and demands, which would be consistent with PFC's known role in abstracting and schematizing information. To test this hypothesis, we recorded neural activity from the medial PFC of rats as they performed an odor sequence memory task, in which they had to determine whether each presented odor occurred in the correct ordinal position in the sequence. Then, we quantified the information contained in the ensemble activity using Bayesian decoding techniques, focusing specifically on sessions in which animals were concurrently tested on two different sequences (one sequence consisting of odors ABCD, the other of odors WXYZ). First, we found that the dynamics of ordinal coding were nearly identical during the two sequences, suggesting a shared representation. In both sequences, ordinal information was decoded at all time points throughout the inter-trial and trial periods as well as across all item positions. In fact, individual neurons were selectively active during specific trial periods across both sequences, and modulated their activity levels during these time points according to the trial's ordinal position. Notably, the influence of ordinal information also varied during the trial period for both sequences, where it was strongest during the pre-trial and early trial period. Second, we found that item-specific information also emerged late in the trial period, suggesting PFC also extracts a representation of the differences between the two sequences. Specifically, activity late in the trial, when the animal has made its decision but is waiting to be instructed to respond, also contained information about the identity of the presented odor (e.g., B vs X). Collectively, these results suggest PFC supports the temporal organization of memory and behavior by providing a schematic representation of the expected series of task events.

Disclosures: K.W. Cooper: None. **M.P. Saraf:** None. **B. Shahbaba:** None. **G.A. Elias:** None. **N.J. Fortin:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.03

Topic: H.08. Learning and Memory

Title: Cortical Circuit Mechanisms of Multimodal Temporal Pattern Discrimination

Authors: *S. POST[1], W. MOL[2], O. ABU-WISHAH[2], S. ALI[3], N. RAHMATULLAH[4], A. GOEL[3];
[1]UC Riverside, [2]UC Riverside, Riverside, CA; [3]Univ. of California Riverside, Univ. of California Riverside, Riverside, CA; [4]Univ. of California, Riverside, Univ. of California, Riverside, Chino Hills, CA

Abstract: Timing is a critical ability across species and behaviors: prey must feint predators at precisely the right moment; drivers must judge whether or not to go through a yellow light; musicians must keep rhythm when playing in a band; and language users must produce and comprehend sequences of syllables in a temporally structured manner. However, very little is known of the circuit mechanisms that give rise to timing in these and other subsecond contexts. To elucidate the mechanistic underpinnings of timing, we recorded from V1 using 2-photon calcium imaging in water deprived, awake-behaving mice while they performed a go/no-go discrimination timing task that was composed of patterns of subsecond audio-visual stimuli. Mice reached a *d'* of 2 or greater in 10.67 ± 2.66 sessions. Learning was primarily driven by changes in response to the nonpreferred stimulus in that correct rejection rates increased. Additionally, mice's licking behavior demonstrated greatest changes across sessions in the nonpreferred stimulus, in which they learned to inhibit licking approximately 500 ms prior to a water reward. Consequently, licking profiles were highly predictive of stimuli in learned but not naive days, which also confirmed learning. Our imaging data showed that in both conditions, activity was temporally coordinated with the preferred stimulus. However, while network activity increased in the preferred condition, network activity was increasingly suppressed in the nonpreferred condition over the stimulus period. We hypothesize that the same excitatory network is recruited in both conditions, but that interneuron activity increases in the nonpreferred condition which broadly inhibits the network. Our results demonstrate that sequential timing can be accomplished by local networks and suggest inhibition as its neural mechanism. Additionally, we suspect that inhibitory dysfunction would cause timing deficiencies, particularly in disorders characterized by excitatory-inhibitory imbalance, such as autism spectrum disorder (ASD), in which temporal processing deficits are a common symptom.

Disclosures: S. Post: None. **W. Mol:** None. **O. Abu-Wishah:** None. **S. Ali:** None. **N. Rahmatullah:** None. **A. Goel:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.04

Topic: H.08. Learning and Memory

Support: NIH Grant R01-MH115697
NIH Grant R01-DC017687
NIH Grant T32-DC010775
NSF Award IOS-1150292

NSF Award BCS-1439267
NSF Award DGE-1839285
Whitehall Foundation (award 2010-05-84)

Title: Persistent and dynamic codes in prefrontal cortex reflect maintenance and updating of a neural representation of position across sequential events

Authors: *G. A. ELIAS[1], M. P. SARAF[1], K. COOPER[1], B. SHAHBABA[2], N. J. FORTIN[1]; [1]Dept. of Neurobio. and Behavior, [2]Departments of Statistics and Computer Sci., Univ. of California, Irvine, Irvine, CA

Abstract: The prefrontal cortex (PFC) is important for the temporal organization of behavior and memory, but its underlying mechanisms are unclear. To investigate this, we implanted rats (n=5) with tetrode hyperdrives targeting medial PFC and recorded neural activity as they performed a complex odor sequence memory task. This task requires rats to identify whether odors are presented at the correct ordinal position across a sequence of trials. We previously found that PFC ensembles contain sufficient information, both during and between trials, to identify the rat's current ordinal position in the sequence. Here we seek to understand how this ordinal information is coded and when it is updated using a cross-temporal decoding analysis with naïve Bayesian classifiers. We find that different forms of coding are used during inter-trial and trial periods. Specifically, ordinal information is represented using a persistent code between trials, while a dynamic code is used during trials. During the inter-trial period, ensemble states are stable over longer periods of time resulting in broad cross-temporal accuracy where activity from any timepoint is equally accurate decoding itself, or almost any other timepoint during the inter-trial. In contrast, during the trial period, ensemble states change more rapidly resulting in a more dynamic, temporally constrained code where activity is only accurate for a ~200ms. Evaluation of cross-epoch accuracy suggests these persistent and dynamic codes reflect representational maintenance and updating respectively. First, ordinal information during the inter-trial period contains more retrospective than prospective content. Specifically, post-trial activity is significantly more accurate at decoding the trial period than the pre-trial period (although cross-epoch decoding between the inter-trial and trial period was poor in general). Second, ordinal information changes around the trial period. Models trained on pre-trial activity decoded post-trial activity as the next position, while models trained on post-trial activity decoded the pre-trial period as the previous position. Collectively, these results demonstrate distinct forms of coding represent ordinal information at different times and suggest these forms support an evolving representation of ordinal position in PFC.

Disclosures: G.A. Elias: None. **M.P. Saraf:** None. **K. Cooper:** None. **B. Shahbaba:** None. **N.J. Fortin:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.05

Topic: H.08. Learning and Memory

Support: NIH Grant R01-MH115697
NIH Grant R01-DC017687
NIH Grant T32-DC010775
NSF Award IOS-1150292
NSF Award BCS-1439267
Whitehall Foundation Award 2010-05-84

Title: Awake hippocampal replay reflects stimuli information during a nonspatial sequence memory task

Authors: *M. P. SARAF[1], G. A. ELIAS[1], K. COOPER[1], B. SHAHBABA[2], N. J. FORTIN[1]; [1]Neurobio. & Behavior, [2]Departments of Statistics and Computer Sci., Univ. of California, Irvine, Irvine, CA

Abstract: The hippocampus is critical to the temporal organization of our experiences. Although this fundamental capacity is conserved across modalities and species, its underlying neuronal mechanisms remain unclear. Accumulating evidence from the place cell literature points to sharp-wave ripple (SWR) associated replay as a potential mechanism supporting this capacity. In fact, such replay of sequences of spatial locations has been linked to the formation and consolidation of spatial memories and to the planning of future trajectories, however, it remains to be determined if this form of sequence coding extends beyond the domain of spatial information. To address this important issue, we re-analyzed an existing dataset in which CA1 ensemble activity was recorded in rats performing a nonspatial sequence memory task. Briefly, the task involved repeated presentations of a sequence of five odors (odors ABCDE), and required the rats to determine whether each odor was presented in the correct order to receive a water reward. We focused our analysis on the intervals between odor presentations during which the reward was delivered and SWR events were observed. In order to identify potential odor replay events, we adapted a "sequenceness" metric previously developed to quantify nonspatial replay in a human MEG study (Liu et al., 2019, Cell). Consistent with evidence from spatial studies, we observed forward replay when testing a familiar sequence. More specifically, replay events rapidly reactivated remaining odors of the sequence (e.g., replay of BCDE during interval preceding odor B). Notably, these replay events were not limited to moments when SWRs were detected - they are distributed throughout the intervals between odor presentations. We also observed reverse replay events during these intervals (e.g., replay of CBA during interval preceding odor C), though they were less prevalent than the forward replay events. Ongoing analyses are examining whether the replay content and direction varies with learning and whether it predicts the accuracy of upcoming odor judgments. Together, these preliminary results suggest that the phenomenon of sequence replay, observed in the hippocampus during offline moments, extends to the processing of nonspatial information.

Disclosures: M.P. Saraf: None. **G.A. Elias:** None. **K. Cooper:** None. **B. Shahbaba:** None. **N.J. Fortin:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.06

Topic: H.08. Learning and Memory

Support: MRC Grant MR/T022922/1
ERC Grant AdG 695709
Wellcome Grant 201225/Z/16/Z
Wellcome Grant 224668/Z/21/Z

Title: Hippocampal dynamics for associating temporally separated events

Authors: *M. BUCHHOLZ[1], N. ROBINSON[1], D. NITU[1], S. MOLLARD[1], E. BAUMLER[1], C. BARRY[2], M. HAUSSER[1];
[1]Wolfson Inst. for Biomed. Res., [2]Res. department of Cell and Developmental Biol., Univ. Col. London, London, United Kingdom

Abstract: Associating events across time is essential for the formation of episodic memories. The hippocampus is thought to support this ability by generating neural activity sequences which tile the temporal gap between events to be linked. Here, we investigate hippocampal dynamics while mice learn and retrieve associations between temporally disparate events. We used two-photon calcium imaging to record the activity of hundreds of CA1 neurons in mice performing an olfactory delayed paired-associates task. During this task, mice had to form specific associations between pairs of odors that were separated by a 5 s mnemonic delay. Presentation of the first odor activated stimulus representations that launched trial-type specific activity sequences maintaining stimulus information across the temporal gap. Upon presentation of the second odor, a group of neurons was modulated by first odor identity, suggesting that these neurons might be involved in integrating information about both stimuli across time to inform choice. In order to study the rapid formation of novel stimulus associations independent of learning the general task rules, we next let expert mice perform the same conceptual task with new odors. Mice were able to generalize the task rules and learn the associations between new odor pairs within two days. Interestingly, mice that ran learnt the new associations faster than mice that did not run. Furthermore, runners exhibited more reliable sequences consisting of a higher number of neurons, suggesting that running organizes the activity of hippocampal sequences. Finally, we investigated whether new memories are acquired by binding event representations onto hippocampal sequence templates. We used an all-optical approach combining two-photon imaging with simultaneous holographic two-photon optogenetics to drive an arbitrary sequence across the mnemonic delay while mice learned novel associations. We hypothesize that this arbitrary sequence might acquire meaning to the brain by starting to encode the new associations and becoming incorporated into the endogenous activity. In summary, our study provides important new insights into the mechanisms by which experiences are organized into episodic memories.

Disclosures: M. Buchholz: None. **N. Robinson:** None. **D. Nitu:** None. **S. Mollard:** None. **E. Baumler:** None. **C. Barry:** None. **M. Hausser:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.07

Topic: H.08. Learning and Memory

Support: KAKENHI JP 19H01131
KAKENHI JP 21H05296
KAKENHI JP 19K12768

Title: Representation of active time perception in the hippocampal CA1 neurons

Authors: *F. SAWATANI, H. AZECHI, K. IDE, S. TAKAHASHI;
Grad. Sch. of Brain Sci., Doshisha Univ., Kyotanabe, Japan

Abstract: We can control and plan our actions in a temporally organized sequence. This ability depends on time perception. “Time cells” in the hippocampus, which are known as time-associated neurons, were found as a neuronal population that sequentially fire during forced delay. Subsequently, several lines of evidence revealed that time cells encode implicit temporal information as they retain memory across time and provide a temporal component of episodic memory integrated with spatial and contextual information. Recent reports suggested that time cells were found in the hippocampus and entorhinal cortex during tasks that require explicit perception of the passage of time, such as temporal discrimination task of sensory stimulus or of a target duration. While most neural correlates studies of time have examined neural activity to passively provided sensory stimulus or delay, the neural activity of the hippocampus during the active generation of specific time events has not been examined in detail. In this study, we established a behavioral task that requires active tracking of the passage of time. Using the task, we examined the neural representations of the hippocampus to the active and explicit time perception by means of extracellular multiple single-unit recording. First, we designed a behavioral task in which rats actively timed in an L-shaped track. Rats were able to start the timer counting by placing their nose into a nose-poke hole at one end of the L-shaped track. Second, the rats could stop the timer and the treadmill rotation by placing their nose in the nose-poke hole on the right side of the treadmill during running. When they were able to stop the timer within a 5-10s time window, they were rewarded at the opposite end of the L-shaped track. Four rats that achieved over 70 % correct response rate for 2 successive sessions (100 trials per session) were implanted with 16 tetrodes in the hippocampal CA1. The activity of putative pyramidal neurons in the CA1 recorded simultaneously from the start to the end of the treadmill running was ordered by latency of their peak firing rate. Pyramidal cells activity sequence was observed until about 2 s elapsed, but thereafter the cells showed no transient activity at a specific moment during treadmill running. Results suggest that the role of hippocampal time cells in time perception may be different from the findings of previous studies. In this presentation, we will report and discuss the differences in the neural underpinning of active and passive time perception tasks, focusing on the role of the hippocampus.

Disclosures: **F. Sawatani:** None. **H. Azechi:** None. **K. Ide:** None. **S. Takahashi:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.08

Topic: H.08. Learning and Memory

Support: NIH Grant R01MH068073

Title: Chemogenetic inactivation of striatal spiny projection neurons modulates timing behavior

Authors: *B. AKDOGAN[1], T. SOHRABI[1], E. H. SIMPSON[1], P. D. BALSAM[2];
[1]Columbia Univ., New York, NY; [2]Barnard Coll Columbia Univ., New York, NY

Abstract: There is a growing body of research demonstrating striatal involvement in establishing temporal control over behavior. However, due to limited investigation of detailed circuit anatomy in the striatum, the contribution of different striatal cell types and subregions to temporal cognition remains largely unknown. To begin addressing this question, we used a virally mediated chemogenetic approach to remotely and temporarily inhibit neural activity selectively in the indirect pathway spiny projection neurons (iSPNs) in the dorsomedial striatum (DMS). We achieved this by injecting a Cre-dependent virus expressing the Designer Receptor Exclusively Activated by Designer Drug (DREADD), hM4Di, (or a control virus) in the DMS of A2a-Cre mice. After recovery from surgery, mice were trained on a timing task that required them to wait for a certain minimum amount of time before making a lever press to earn food rewards. In two experimental phases, all mice were tested with either a short (4 s) or a long (8 s) criterion value. After they learned to withhold responding for the appropriate amount of time, mice were tested 30 minutes after intraperitoneal injection with a DREADD ligand, JHU37160 (0.2 mg/kg or 0.8 mg/kg), or saline. Our analysis of wait times revealed that chemogenetic inactivation of iSPNs in the DMS resulted in earlier and more variable response latencies in a dose-dependent manner. The administration of the higher JHU37160 dose had a more detrimental effect on the accuracy and trial-to-trial variability of wait times. The magnitude of leftward shifts in wait times was not the same absolute amount of time in the 4- and 8-s conditions. Instead, responses distributions changed in proportion to target wait times (15-25%) in both conditions. Therefore, the observed reduction in wait times cannot be the result of a simple motoric effect. Our findings provide evidence for a significant role of the indirect (striatopallidal) pathway in key information-processing mechanisms of temporal cognition, such as time-keeping and criterion-setting. Currently, we are also testing the effects of chemogenetic inactivation of direct (striatonigral) pathway spiny projection neurons in the DMS on waiting behavior. This will delineate the contribution of dopamine D1 and D2 receptor-expressing striatal neurons to temporal processing.

Disclosures: **B. Akdogan:** None. **T. Sohrabi:** None. **E.H. Simpson:** None. **P.D. Balsam:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.09

Topic: H.08. Learning and Memory

Support: NIH R01NS120987
NIH R01MH116043

Title: Optogenetic manipulation of direct and indirect pathways during interval timing in mice

Authors: *V. KHANDELWAL[1], A. S. BOVA[2], T. M. LARSON[4], M. M. CONLON[3], N. S. NARAYANAN[5];
[1]Dept. of Neurol., [2]Dept. of Psychiatry, [3]Med. Scientist Training Program, Univ. of Iowa, Iowa City, IA; [4]Duke Med. Sch., Durham, NC; [5]Univ. of Iowa Roy J and Lucille A Carver Col. of Med., Univ. of Iowa Roy J and Lucille A Carver Col. of Med., Iowa City, IA

Abstract: Precisely controlling the timing of movement is a critical human faculty that is impaired in many neuropsychiatric disorders. Timing depends on dopamine (DA) activity in the dorsomedial striatum and manipulating striatal dopamine levels causes changes in temporal estimation. However, dopamine has differential effects on direct (dMSN) and indirect (iMSN) pathway spiny projection neurons. Canonically, these distinct pathways are thought to play oppositional roles in movement, and therefore we sought to characterize their roles in timing. We tested timing behavior using a task which requires mice to respond at either a short or a long port based on their estimation of the duration of an interval. We optogenetically stimulated or inhibited dMSNs or iMSNS by delivering pulses of light to dMSN terminals in the substantia nigra pars reticulata or iMSN terminals in the external globus pallidus. We found that inhibition of both pathways led to an increase in the estimation of time. Activation of the direct pathway also led to similar overestimations, but activation of the indirect pathway led to an underestimation of temporal intervals. Our results suggest a heterogenous role for striatal DA in timing, and complexity in dMSN and iMSN response. We will test this hypothesis by recording from neuronal ensembles in downstream targets of each pathway, and through optogenetic manipulations of these structures. Our work may lead to targeted, cell-type specific, and novel pharmacological interventions for disorders of dopamine like Parkinson's Disease.

Disclosures: **V. Khandelwal:** None. **A.S. Bova:** None. **T.M. Larson:** None. **M.M. Conlon:** None. **N.S. Narayanan:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.10

Topic: H.08. Learning and Memory

Support: NIH Grant 5R01MH116043-04

Title: Dorsalstriatal dopamine dynamics regulate temporal encoding by striatal projection neurons

Authors: *A. BOVA[1], V. KHANDELWAL[2], M. A. WEBER[1], N. S. NARAYANAN[3]; [2]Dept. of Neurol., [1]Univ. of Iowa, Iowa City, IA; [3]Neurol., Univ. of Iowa Roy J and Lucille A Carver Col. of Med., Iowa City, IA

Abstract: Temporal control of movement is an essential component of everyday behaviors and relies on cognitive processes including working memory and attention to the passage of time. Parkinson's disease patients display deficits in estimating temporal intervals and optogenetically manipulating substantia nigra dopamine neurons in rodents disrupts time estimation, indicating that dopamine can directly modulate judgement of time. Dorsal striatal medium spiny projection neurons display time-related ramping activity across temporal intervals. However, how dopamine release in dorsal striatum regulates striatal projection neuron temporal encoding is unknown. We investigated this question by recording dorsal striatal dopamine dynamics using fiber photometry and the genetically encoded optical dopamine sensor, dLight, while simultaneously recording dorsal striatal neuronal ensemble activity as mice performed an interval timing task. In this task mice are required to 'switch' from one response port to another if a reward has not been delivered after a specific interval of time has passed. We found that dLight progressively increased across the temporal interval until reward delivery only in trials where the mouse correctly 'switched'. This progressive increase was greater in trials when mice 'switched' earlier in the interval. Therefore, the magnitude of the dLight signal may predict trial performance and is temporally-locked with striatal projection neuron activity. Ongoing work is exploring how optogenetic manipulations of substantia nigra dopamine neurons influence striatal neuronal activity during interval timing. These results will provide a mechanistic understanding of how nigrostriatal dopamine regulates time estimation, and more broadly cognitive processing, which will inform development of novel therapeutic interventions for cognitive deficits in Parkinson's disease and other dopamine-related disorders.

Disclosures: **A. Bova:** None. **V. Khandelwal:** None. **M.A. Weber:** None. **N.S. Narayanan:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.11

Topic: H.08. Learning and Memory

Support: NIH R01NS120987
NIH R01MH116043

Title: Differences in sex and dopamine during interval timing and amphetamine withdrawal in mice.

Authors: *H. STUTT[1], M. A. WEBER[1], N. S. NARAYANAN[2];
[1]Univ. of Iowa, Iowa City, IA; [2]Neurol., Univ. of Iowa Roy J and Lucille A Carver Col. of Med., Iowa City, IA

Abstract: There are still no effective treatments for amphetamine and other substance use disorders (SUD). Developing novel treatments for the heterogeneous population of people suffering from SUD requires understanding the mechanisms that underlie the differences between SUD populations. For example, females report more adverse withdrawal symptoms and have higher cue-induced relapse rates. Cognitive dysfunction during withdrawal contributes to the maintenance of SUD. However, the mechanisms underlying cognitive impairments during withdrawal and if these impairments differ based on sex are unknown. Dorsal striatal dopamine is implicated in both sex differences in SUD and cognitive dysfunction. Amphetamine-induced dopamine release is enhanced following administration of estradiol, a hormone that fluctuates during the reproductive cycle, in females and lesioning dorsal striatal dopamine impairs performance on cognitive tasks. To examine sex differences in SUD-related cognitive dysfunction, male and female mice were administered amphetamine (2.5 mg/kg i.p. daily) for 14 days followed by a forced withdrawal period of 14 days during which they were tested in an interval timing 'switch' task. In this task, mice determine if a light-tone cue has been presented for more than six seconds by 'switching' from one response port to another. This task relies on cognitive processes such as working memory and attention and is dependent on striatal dopamine. During withdrawal, both females in the diestrus stage of the reproductive cycle and males displayed later 'switch' times compared to saline controls. However, when females were in the estrus stage, 'switch' times did not differ from saline controls. This suggests that amphetamine produces a change in time perception during withdrawal in an estrus cycle-dependent way. To determine if this effect is dopamine-dependent, we will use fiber photometry and the fluorescent dopamine indicator, dLight, to observe dopamine dynamics in the dorsomedial striatum during interval timing. We hypothesize that during amphetamine withdrawal, task-related dopamine dynamics will be dampened in females in diestrus and males compared to amphetamine naïve mice. However, females in estrus will have similar dopamine dynamics compared to amphetamine naïve mice. These data will uncover the sex-specific mechanisms underlying cognitive impairments during amphetamine withdrawal, which will be key in finding more effective treatment options for SUD.

Disclosures: H. Stutt: None. **M.A. Weber:** None. **N.S. Narayanan:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.12

Topic: H.08. Learning and Memory

Support: NIH Grant 5 RO1 MH116043

Title: Effects of Levodopa treatment on temporal encoding activity in dopamine-depleted medium spiny neurons

Authors: *R. A. VOLKMAN, R. A. BRUCE, M. A. WEBER, Y. KIM, N. S. NARAYANAN; Neurol., Univ. of Iowa, Iowa City, IA

Abstract: Time-based decision-making requires nigrostriatal dopamine signaling. Striatal medium spiny neurons (MSNs) are strongly modulated by dopamine, and MSNs display prominent 'ramping' activity during timing tasks. Parkinson's disease (PD) involves the degeneration of dopaminergic nigrostriatal projections, and patients with PD suffer from deficits in temporal processing. However, how the dopamine-depleted striatum encodes time, and how Levodopa treatment modulates time-related neuronal activity remains poorly understood. Here, we selectively lesioned nigrostriatal dopamine projections via focal stereotactic injections of 6-hydroxy-dopamine (6-OHDA) (2mg/ml), and implanted 4x4 multielectrode arrays targeting the dorsal striatum (AP +.5mm, ML +1.4mm, DV -2.7mm) in mice trained to perform a novel interval timing task. We report 3 main findings. Firstly, 6-OHDA lesioned mice saw an increased latency to 'switch' compared to a saline control cohort, suggesting disrupted temporal processing. Second, the proportion of dorsal striatal MSNs exhibiting time-related 'ramping' activity was reduced amongst 6-OHDA lesioned mice compared to saline control mice. Finally, we report that treating both lesioned and control mice with Levodopa (10mg/kg) and Benserazide (12mg/kg) resulted in a reduction in ramping activity compared to control saline administration days, hinting that ramping may require phasic rather than tonic dopamine signaling. Our findings may provide explanatory power for PD-related deficits in timing and cognition more broadly which are notoriously resistant to first-line therapies such as Levodopa treatment.

Disclosures: R.A. Volkman: None. **R.A. Bruce:** None. **M.A. Weber:** None. **Y. Kim:** None. **N.S. Narayanan:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.13

Topic: H.08. Learning and Memory

Support: NIH Grant RO1 MH116043
NIH Grant RO1 NS120987

Title: Ventral tegmental area stimulation modulates interval timing deficits in dopamine depleted mice

Authors: *K. SIVAKUMAR, G. GAUDENCIO, M. A. WEBER, N. S. NARAYANAN; Neurol., Univ. of Iowa, Iowa City, IA

Abstract: Parkinson's disease (PD) patients can experience debilitating cognitive symptoms and deep brain stimulation only targets motor dysfunction; currently, there are no effective treatments for PD-related cognition. PD patients with mild cognitive impairment or dementia reliably display deficits in interval timing, or the ability to estimate time over seconds, and lower cognitive scores are associated with greater interval timing variability (Singh et al., 2021). Previous work has shown that depleting dopamine in the rodent ventral tegmental area (VTA) increases interval timing variability and optogenetically stimulating the frontal cortex D1-type dopamine receptors ameliorates these deficits (Kim et al., 2017). However, stimulation of an acute frontal cortex region only targets a proportion of terminals originating from the VTA. Therefore, we hypothesized that stimulating the source (VTA) can more powerfully improve interval timing in dopamine depleted mice. We first trained dopamine transporter (DAT)-Cre mice (n=8; counterbalanced across sex) to perform an interval timing switch task in which internal processes guide the animal's time to switch from one response port to another. We then bilaterally depleted dopamine in the VTA (AP: -3.3, ML: +/-1.1, DV: -4.55) using 1 or 2 µg/µL 6-hydroxydopamine (6OHDA) and injected Cre-dependent channelrhodopsin (ChR2) in the same location. After one week of recovery and two-three weeks of retraining, we bilaterally stimulated VTA-ChR2 at either 4 or 20 Hz for the first two seconds of interval timing trials. Our preliminary results show that both 4 and 20 Hz stimulation of the remaining VTA neurons decreases interval timing variability, suggesting that VTA stimulation can modulate interval timing in dopamine depleted mice. We are currently training more animals to increase experimental power. These findings are of particular interest in understanding the neural circuitry that underlies PD-related cognition. Further, this work provides insight into how brain stimulation has the potential to mitigate cognitive dysfunction associated with PD and other dopamine-related neurological and neuropsychiatric disorders.

Disclosures: K. Sivakumar: None. **G. Gaudencio:** None. **M.A. Weber:** None. **N.S. Narayanan:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.14

Topic: H.08. Learning and Memory

Support: EMBO ALTF 1161-2017
LT0000717/2018
NIH MH54671
NIH MH107396
NS 090583
NSF PIRE 1545858
U19 NS107616

Title: Intrinsic and network properties of interneuron diversity in the hippocampus

Authors: *M. VALERO[1], P. ABAD[2,3], R. P. MACHOLD[4], B. RUDY[5], G. BUZSAKI[6];
[1]NYU Neurosci. Inst., New York, NY; [2]Univ. Cardenal Herrera CEU, Elche, Spain; [3]Inst. de Neurosciencias UMH-CSIC, Alicante, Spain; [4]Neurosci., [5]Neurosci. & Physiol., NYU Sch. of Med., New York, NY; [6]Neurosci., New York University, Langone Med. Ctr., New York, NY

Abstract: Neuronal diversity in the cortex is the largest amongst GABAergic neurons. In the hippocampus, GABAergic cells have been sorted in several groups based on their morphology, firing patterns, connectivity, molecular profiling, and RNA content. It is thought that these specialized GABAergic groups aid specific circuit computations. Identification and manipulation of these distinct neuron types is a prerequisite to deciphering not only their role in circuit dynamics and behavior but also the computational mechanisms of the cortical networks they are embedded in. To catalog the interneuron diversity in the hippocampal area CA1, we first grouped the virtual totality of GABAergic neurons into four major families, based on novel and standard genetic markers. Parvalbumin (PVALB)-expressing neurons, Somatostatin (SST)-expressing neurons, Vasoactive intestinal polypeptide (VIP) and Inhibitor of DNA binding 2 (ID2)-expression GABAergic cells comprised around 97% of CA1 GABAergic diversity. We then generated double and triple transgenic mice lines expressing channelrhodopsin (ChR) under the control of our four target genes (PVALB::Ai32, SST::Ai32, VIP::Ai80, and ID2/Dlx::Ai80). When necessary, we restricted the expression to only GABAergic cells by the intersection with distaless homeobox (Dlx) 5/6 to avoid expression in non-GABAergic cells. Using chronically implanted silicon probes coupled with optic fibers, we recorded and optogenetically identified large numbers of interneurons from these main four families in freely behaving mice (n = 3, 2, 3 and 4 mice for PVALB, SST, VIP and ID2/Dlx, respectively, more than 30 opto-tagged units per group). The four interneuron families show distinct intrinsic features and exhibited specific activity dynamics during NREM and REM sleep, theta oscillations, sharp-wave ripples, reward consumption and spatial exploration. Further subclasses within these four main families were identified by triple transgenic intersections (including ID2/Nkx2.1::Ai80, VIP/CCK::Ai80 and VIP/CR::Ai80), and validated with in vitro recordings and morphological reconstructions. Finally, we built an automatic classification tool based on the observed levels of complexity (waveform and auto-correlogram features, soma location, network interactions and brain state dynamics) which enable ground-truth-based classification of interneurons from hippocampal extracellular recordings. These experiments provided a high precision physiological characterization of interneuron types, a prerequisite for understanding their collective organization for supporting circuit computation.

Disclosures: **M. Valero:** None. **P. Abad:** None. **R.P. Machold:** None. **B. Rudy:** None. **G. Buzsaki:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.15

Topic: H.08. Learning and Memory

Support: NIH Grant 5R01MH129046
Paul and Lilah Newton Brain Science Award
NSERC PDF-516867-2018
FRQNT B3X-258512-2018
McGovern Institute for Brain Research

Title: Mental navigation recruits complimentary abstract computations in the entorhinal and parietal cortex

Authors: *S. NEUPANE, I. R. FIETE, M. JAZAYERI;
Brain and Cognitive Sci., MIT, Cambridge, MA

Abstract: The nervous system richly represents spatial relationships and uses this information to solve cognitive problems such as planning a trip via a new route. However, it is not known how these representations are put to use to perform flexible computation. To address this question, we developed a task for monkeys in which the animals travel virtually (via a joystick) between two points on a 1-dimensional path containing a sequence of 6 equidistant images. On each trial, animals had to deflect the joystick in the correct direction and for the appropriate duration to move from a start image to an end image along the path. Crucially, the task had to be solved mentally; i.e., without sensory feedback about the intervening images.
This task can be solved either through online simulation of images based on their ordinal position (mental counting), or by inferring the temporal distance between the start and end image and producing that interval without simulating the intervening images (timing). These strategies have distinct behavioral signatures. Counting predicts lower behavioral variability compared to timing, especially for higher counts. Using a rigorous behavioral modeling approach, we found that animals' behavioral variability was better accounted for by counting. These strategies also have distinct neural signatures. Counting starts from a common initial point (count of 0), goes through intermediate states with a periodicity that matches the inter-image interval, and terminates at different points depending on the ordinal distance (1, 2, 3, ⋯). In contrast, timing is governed by dynamics that start from different initial conditions and advance at different rates toward a common threshold [Wang et al. 2018]. We recorded from the entorhinal cortex (EC) and posterior parietal cortex (PPC) to test these predictions. We found single neurons in both areas whose profile was either more similar to counting or timing. However, at the population level, EC and PPC signals diverged. EC neurons had hallmarks of counting. They had a weak representation of distances before joystick deflection, a striking periodic activity during navigation, and terminal points representing the total ordinal distance. PPC, in contrast, had distance-dependent initial states and ramping activity with different slopes reaching a common

terminal point. These findings suggest that the brain may solve mental navigation tasks using complementary computations, one that occurs before navigation to generate a plan based on an initial inference and another that unfolds during navigation and simulates intermediate states.

Disclosures: S. Neupane: None. **I.R. Fiete:** None. **M. Jazayeri:** None.

Poster

572. Timing and Temporal Processing: Cortex, Hippocampus, Striatum

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 572.16

Topic: C.09.Stroke

Support: NWO Grant 406.18.GO.063
KNAW WF/1627

Title: On the phylogenesis of rhythm cognition and the causal involvement of cortico-subcortical structures

Authors: *A. CRISCUOLO[1], M. J. HENRY[2], M. SCHWARTZE[1], H. MERCHANT[3], S. A. KOTZ[1,4];
[1]Maastricht Univ., Maastricht, Netherlands; [2]Max Planck Inst. for Empirical Aesthetics, Max Planck Inst. for Empirical Aesthetics, Frankfurt Am Main, Germany; [3]Inst. de Neurobiologia UNAM, Inst. de Neurobiologia UNAM, Queretaro, Mexico; [4]Neuropsychology, Max Planck Inst., Leipzig, Germany

Abstract: The ability to successfully navigate in a dynamically changing environment relies on *adaptive* mechanisms which allow to *encode* and *predict* the timing of external events. Since Darwin, comparative research has shown that most animals share basic *timing* capacities, such as the ability to process temporal *regularities* and produce *rhythmic* behaviors. What seems to be more exclusive, however, are the capacities to generate *temporal predictions* and to display *anticipatory* behavior at *salient* time points. These abilities are associated with subcortical structures like basal ganglia (BG) and cerebellum (CE), which are more developed in humans as compared to nonhuman animals. The current work adopts a comparative and translational approach, by examining the phylogenetic trajectories of human's rhythm cognition as well as the causal involvement of cortico-subcortical structures. We made use of a unique dataset including 2 macaque monkeys, 20 healthy young (HY), 11 healthy old participants (HO) and 22 stroke patients, 11 with focal lesions in the BG and 11 in the CE. We recorded EEG while participants listened to isochronous equitone sequences presented at a rate of 1.5Hz. We examined whether neural oscillatory activity in the delta-band (1-3Hz) internalized the timing of external events, by encoding temporal regularity. Next, we tested whether delta-activity showed an anticipatory phase-alignment to expected tone onsets, which would indicate the ability to predict the precise timing of events. Interestingly, macaque monkeys showed striking similarities with human participants: they showed a clear peak in the Fourier spectrum at 1.5Hz, thus confirming the

ability to encode temporal regularities. Furthermore, healthy participants' and macaque monkeys' delta-band activity displayed a coherent and anticipatory phase-alignment to expected tone onsets, as indexed by mean vector length (MVL). HO and patients showed a similar peak in the Fourier spectrum at 1.5Hz, but significantly differed in their MVL: BG patients showed a stronger phase-alignment to tone onsets as compared to the other groups. When compared to HY, however, HO and CE patients showed lower MVL, while BG patients were comparable. Our phylogenetic and translational approach demonstrates that, similarly to humans, macaque monkeys are able to encode temporal regularities and formulate temporal predictions. We further show that ageing and CE lesions, but not BG, alter temporal predictions, reducing the ability to track environmental rhythms. These observations provide crucial evidence for a differential but complementary role of CE and BG in the phylogenesis of human's rhythm cognition.

Disclosures: **A. Criscuolo:** None. **M.J. Henry:** None. **M. Schwartze:** None. **H. Merchant:** None. **S.A. Kotz:** None.

Poster

573. Spatial Cognition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 573.01

Topic: H.09. Spatial Navigation

Support: NSF IIS #1703340
NSF IIS #1703225
UBFC ANER Robotself

Title: Integrating velocity-dependent spatio-temporal place cell information to a reservoir model of prefrontal cortex

Authors: P. SCLEIDOROVICH[1], ***A. WEITZENFELD**[2], J.-M. FELLOUS[3], P. DOMINEY[4]; [1]Computer Sci. & Engin., USF, Tampa, FL; [2]Univ. of South Florida, Univ. of South Florida, Tampa, FL; [3]Psychology, Univ. of Arizona, Tucson, AZ; [4]INSERM U1093, Dijon, France

Abstract: In this work we investigate how speed patterns might be behaviorally significant in rats during sequential multigoal spatial navigation and how cortical networks might encode this information. We show how variations in speed patterns can teach the rat different goals while following the same spatial trajectory in the same overall time. We use a novel experimental paradigm, where we train rats to follow a small baited robot in a large open-field megaspace where the velocity of the robot is used to precisely control the speed of the rat. We show that rats can make spatial navigation decisions based on the navigation speed pattern they just experienced. Based on these results and previous research we show that recurrent reservoir networks can appropriately represent spatio-temporal structures. We test reservoir networks in simulated navigation contexts and demonstrate they can discriminate between traversals of the same path with identical durations but different speed patterns. We then test the networks in an

embodied robotic setup, where we use place cell representations obtained from physically navigating robots to evaluate the model. To demonstrate that this capability is inherent to recurrent networks, we compare the model against linear integrators. Results show that reservoir neurons can display a form of statistical mixed selectivity as a complex interaction between spatial location and speed that was not as abundant in the linear integrators. This mixed selectivity is characteristic of the cortex and reservoirs generating specific predictions about the neural activity that can be recorded in rat prefrontal cortex in future experiments.

Disclosures: P. Scleidorovich: None. **A. Weitzenfeld:** None. **J. Fellous:** None. **P. Dominey:** None.

Poster

573. Spatial Cognition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 573.02

Topic: H.09. Spatial Navigation

Support: Marie Sklodowska-Curie 101022757
Wellcome 223144

Title: Spatial modulation of sensory activity during virtual navigation

Authors: E. H. VAN BEEST, R. BI, K. D. HARRIS, M. CARANDINI;
Univ. Col. London, London, United Kingdom

Abstract: Spatial navigation is a complex form of goal-directed behavior that depends on cognitive processes such as memory, attention, and perception. A key center for spatial memory and spatial navigation is region CA1 of the hippocampus, where place cells enhance their firing at a certain position in a (physical or virtual) environment. Spatial signals have been found in many other brain regions, including regions that were considered purely sensory such as the visual cortex. The interaction between sensory and spatial input is unclear. Also, the extent to which spatial coding is distributed across other brain regions is unknown. Using Neuropixels probes we are investigating the distribution of spatial coding across the brain. Additionally, we aim to understand how spatial signals interact with sensory signals.
We recorded and extracted single-unit activity from different brain regions in mice (N=13) navigating a linear corridor with two sensory identical halves (Saleem et al *Nature* 2018). The visual contrast of landmarks in the corridor varied across trials, and in a subset of trials, the landmarks were also the source of an auditory stimulus.
Most single units (70-90% for most areas we recorded from) were significantly modulated by the position in the virtual corridor ($p<0.05$, Rayleigh's test). This modulation could be a consequence of spatial preference, sensory modulation (responses to the auditory and visual landmarks), or other factors such as motion. More than 60% of units in hippocampal and cortical regions (and a smaller fraction in thalamus) exhibited spatial preference, meaning that they

responded differently in the two identical halves of the corridor. Hippocampal regions showed less sensory modulation than other areas.
These results show that many units across the brain are modulated by sensory input as well as subjective position in the virtual corridor. We are currently investigating whether the spatial modulation on sensory input is additive or multiplicative.

Disclosures: E.H. van Beest: None. **R. Bi:** None. **K.D. Harris:** None. **M. Carandini:** None.

Poster

573. Spatial Cognition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 573.03

Topic: H.09. Spatial Navigation

Title: Orbitofrontal and hippocampal responses to hidden spatial goals

Authors: *S. GUPTA, A. M. WIKENHEISER;
Psychology, UCLA, Los Angeles, CA

Abstract: Both orbitofrontal cortex (OFC) and hippocampus have been implicated in representing task state. Place cells in the hippocampus encode state as defined by spatial locations, while neurons in OFC encode more abstract features of state, integrating information about responses, reward expectancies, value, and other attributes. Theoretical work has argued that OFC is particularly important in situations where no single piece of information unambiguously signals the current state. Here, we examined representations in OFC and hippocampus of male rats (n = 7) as they performed a behavioral task that required them to learn about unmarked spatial goal locations in an open field arena. Pausing in either of two hidden goal locations caused one food pellet to be delivered, but the two goals differed in where they caused food to be delivered. Pausing in the “fixed” goal dispensed a pellet to a consistent location in the arena while pausing in the “random” goal dispensed a pellet to a random location in the arena. Both goal locations were randomly assigned for each behavioral session. Rats were able to learn both goal locations, earning an average of 36 pellets per session from the random goal and 33 pellets per session from the fixed goal. Further, rats learned to distinguish the fixed and random goals, as evidenced by the development of increasingly fast and direct trajectories between the fixed goal and the fixed reward location over the course of behavioral sessions. Rats did not, however, prefer one goal type over the other. Each rat was implanted with one bundle of sixteen microwire electrodes targeting the lateral orbitofrontal cortex, and one bundle of 8 stereotrodes targeting the dorsal hippocampus. Over 101 behavioral sessions, we recorded 847 OFC units and 661 hippocampal units. A large fraction of hippocampal units showed spatially-specific firing. Place fields did not cluster around goal locations relative to non-goal locations, and approximately equal numbers of fields occurred near the fixed and random goals. Consistent with previous reports, many OFC neurons showed clear reward responses. In addition, many OFC neurons show reliable changes in firing rate as rats entered and waited at goal locations,

with a sizeable fraction of goal-modulated neurons discriminating the fixed and random goal locations.

Disclosures: S. Gupta: None. **A.M. Wikenheiser:** None.

Poster

573. Spatial Cognition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 573.04

Topic: H.09. Spatial Navigation

Support: NIH F31 EY031582
NSF/IOS 1924732
NIH R01 MH123260-01

Title: Retrosplenial representations of space and hippocampal circuitry underlying spatial reorientation

Authors: *C. GAGLIARDI, M. E. NORMANDIN, N. PUNJAALA, I. A. MUZZIO;
Univ. of Texas At San Antonio, San Antonio, TX

Abstract: Reorientation, the process by which lost navigators regain their bearings, is fundamental for navigation. Under oriented conditions, navigators use both external and internal cues to navigate. However, during disorientation, the internal sense of direction is unreliable and lost navigators must rely on external cues to reorient. Studies have shown that geometric cues are especially important for reorientation. However, over time, learned associations of goal locations/rewards with non-geometric, featural cues (odors, textures, sounds, etc.) are formed and lost navigators begin to rely on these associations over geometric cues to minimize errors in geometrically similar locations. The retrosplenial cortex (RSC) has been shown to be important for coding the shape of environments. The RSC also receives inhibition from long-range GABAergic projections originating in hippocampal area CA1 (H-RSC), providing a potential pathway for attenuating the use of geometry to bolster the use of reinforced cues. Disoriented mice were trained to find a reward hidden in one out of four cups placed near one corner of a rectangular chamber with visual cues. Using chemogenetic silencing of RSC neurons during distinct stages of reorientation, we demonstrate that the RSC is required early during reorientation, when lost subjects reorient using geometry, but not late in training, when mice have learned the directional value of featural cues and use them to reorient. Optogenetic activation of H-RSC during reorientation during early and late training periods display distinct impacts on behavor. We also conducted single-unit and calcium imaging recordings of RSC cells during reorientation, finding alignment of RSC representations to environmental geometry. We are currently characterizing bi-directional head-direction cells and egocentric boundary tuning to explore how these spatial representations change as mice gain experience with the task. We hypothesize that the spatial properties of these representations will change as animals switch

geometric strategies to featural ones. These results will be critical to understand the role of the RSC in reorientation and will help to elucidate how different aspects of the environment control single cell activity in this region.

Disclosures: C. Gagliardi: None. **M.E. Normandin:** None. **N. Punjaala:** None. **I.A. Muzzio:** None.

Poster

573. Spatial Cognition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 573.05

Topic: H.09. Spatial Navigation

Title: Projection-specific retrosplenial cortex circuits and the transformation of spatial cognition to action

Authors: *X. LIN[1], M. AMALRAJ[2], N. M. L. HA[2], B. AVILA[2], D. A. NITZ[3], X. XU[4]; [1]Univ. of California Irvine, Univ. of California Irvine, Irvine, CA; [2]Univ. of California, Irvine, Irvine, CA; [3]Univ. of California San Diego, La Jolla, CA; [4]Anat. and Neurobio., Univ. California, Irvine, Irvine, CA

Abstract: Retrosplenial cortex (RSC) plays a key role in the neural coding of location and orientation relative to environment boundaries as well as left/right tunning actions within path networks. RSG is anatomically interconnected with multiple cortical and subcortical brain regions, including sensory cortices, the hippocampal/parahippocampal formation, and the anterior thalamus. Prior work has suggested that the encoded spatial information from the hippocampal formation is integrated with RSC and then is transformed into the action-related secondary motor cortex. However, neural circuit mechanisms for the transformation of spatial information into planned action through pathway-specific circuitry remain largely undefined. To address this, we used Cre-dependent retrograde monosynaptic rabies virus and adenovirus mapped and compared circuit input connections of layer-specific RSC excitatory neurons, M2-projecting, anterior thalamus-projecting, and postsubiculum-projecting RSC neurons in mouse brain. We find significant stronger inputs from the subiculum, visual cortex, somatosensory cortex, auditory cortex, and thalamus to M2-projecting RSG neurons while AD-projecting RSC neurons receive stronger inputs from the cingulate cortex, motor cortex, and media septal nucleus. Moreover, we developed a spatial place-based cross-maze task that required mice to make left/right choices depending on where they were located in the room. We find that genetic inactivation of the projection of RSG to M2 impairs the accuracy of space-based L/R turn decision-making. These results provide an anatomical circuit basis to understand RSG neurophysiological function and its role in the transformation of spatial cognition to action planning.

Disclosures: **X. Lin:** None. **M. Amalraj:** None. **N.M.L. Ha:** None. **B. Avila:** None. **D.A. Nitz:** None. **X. Xu:** None.

Poster

573. Spatial Cognition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 573.06

Topic: H.09. Spatial Navigation

Support: NRF 2018R1A4A1025616
NRF 2019R1A2C2088799
NRF 2021R1A4A2001803
NRF 2022M3E5E8017723
BK21 program

Title: A virtual place preference task dependent on the interactions between the prefrontal cortex and hippocampus in a VR environment

Authors: *S. PARK[1], S.-W. JIN[1], H. HWANG[1], J. SHIN[1], H.-W. LEE[2], I. LEE[1];
[1]Seoul Natl. Univ., Seoul Natl. Univ., Seoul, Korea, Republic of; [2]Neurobio., Univ. of Utah, Salt Lake City, UT

Abstract: We hypothesize that the iHPC-mPFC interactions are critical for an animal to navigate a space in a goal-directed fashion. To test this, we trained rats (n=6) to navigate to an unmarked goal location in a VR environment to receive water rewards. The rat navigated a circular VR arena by running on a ball-shaped treadmill. The rat started from the center of the arena, facing either north or south, to visit the unmarked reward zone on the west (WRZ). Rats typically showed a stereotyped turning behavior (turning to the right) as a trial started and then adjusted the length of the trajectory depending on the starting heading direction. Once the rat was trained, the rat was trained to go to a new reward zone on the east (ERZ). Afterward, a 24-tetrode-carrying hyperdrive was implanted, targeting the mPFC and iHPC. After recovery, tetrodes were lowered to the target regions and rats were retrained. As the main recording commenced, the rat was required to visit the WRZ. Then, rats were required to reverse the goal zone again to the ERZ until they reached the performance criterion. When we checked how accurately the rat arrived at the reward zone by measuring the angular difference between the center of the reward zone and the first contact point of the circular boundary of the arena, there was a significant difference between the sessions in which rats performed below and above the performance criterion ($p<0.0005$, Kruskal-Wallis test). However, a similar analysis on the departure angle at the start point did not result in performance-related correlates, suggesting that rats used allocentric visual scenes encountered during the navigation to adjust their travel paths for wayfinding to the goal zone in our task. This allocentric strategy was also confirmed when we examined the navigation patterns of rats at different learning stages. Specifically, rats tended to show an egocentric strategy by entering the closest reward zone (ERZ or WRZ) associated with

each start heading direction (ERZ and WRZ when heading north and south at the start, respectively) before learning. However, as rats learned the task (reaching the goal zone > 75%), they inhibited the egocentric tendency to travel to the nearest goal zone to take a longer path to reach the goal zone on the opposite side in those trials in which their initial departing orientations carried them farther from the goal zone as they started their navigation in a stereotypic way (i.e., right turn). Such a strategic shift made the rat's hitting rate for the goal zone significantly higher after learning compared to the pre-learning stage ($p<0.0001$, Chi-square test). We plan to analyze spiking activities of single units recorded from the iHPC and mPFC in this task.

Disclosures: S. Park: None. **S. Jin:** None. **H. Hwang:** None. **J. Shin:** None. **H. Lee:** None. **I. Lee:** None.

Poster

573. Spatial Cognition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 573.07

Topic: H.09. Spatial Navigation

Support: NIH grant MH119391

Title: A circuit for spatial based decisions - Oscillatory dynamics of the medial prefrontal cortex, dorsal and ventral hippocampus

Authors: *K. S. KIDDER[1], J. T. MILES[2], S. J. Y. MIZUMORI[1];
[1]Psychology, [2]Neurosci., Univ. of Washington, Seattle, WA

Abstract: Past evidence from electrophysiological analyses and experimental manipulations have suggested that decision-making which involves spatial working memory, relies on information exchange between dorsal hippocampus (HPC) and the medial prefrontal cortex (mPFC). However, in the last decade evidence has arisen which suggests ventral HPC and its communication with the mPFC as well as dorsal-ventral HPC communication may also be important for successful spatial working memory performance. To better understand the interplay between these three areas, we implemented tri-site neural recordings of the mPFC, and both (dorsal/ventral) hippocampal regions as rats performed a spatial delayed alternation task. We then analyzed neural data by task epoch (delay, choice, return). Preliminary analyses reveal prominent theta peaks in both dorsal and ventral HPC during the choice epoch of the spatial delayed alternation task. Additionally, theta coherence between each pair of structures is highest during the choice epoch. These results suggest that the broader "dorsal HPC - ventral HPC - mPFC" circuit communicates at select timepoints during the successful execution of decision making involving spatial working memory.

Disclosures: K.S. Kidder: None. **J.T. Miles:** None. **S.J.Y. Mizumori:** None.

Poster

573. Spatial Cognition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 573.08

Topic: H.09. Spatial Navigation

Title: Computational neural mechanisms of scalable planning to arbitrary goals

Authors: *H. CHENG, J. W. BROWN;
Dept. of Psychological and Brain Sci., Indiana Univ. Bloomington, Bloomington, IN

Abstract: The neural mechanisms of goal-directed behaviors remains an open question. The hippocampus is associated with path integration and replays. RL-based algorithms address the problem of seeking goals by learning a Q value landscape, but when the rewarded state (I.e. goal) changes, the Q-values must be recomputed, which makes switching goals inefficient. The GOLSA model (Zarr & Brown, 2019; Fine et al. 2020) suggested how the orbitofrontal cortex and hippocampus work together to reach arbitrary goal states efficiently when the goals vary. Still, the GOLSA model requires a one-hot representation that fails to scale up to large problems. Here we revise the GOLSA model by replacing the one-hot representations with a deep network, which takes as input both the current state and the goal state representations. Planning then becomes an iterative directed search for how to achieve the goal state. The model orbitofrontal cortex represents the value of the planned state transitions with respect to the current goal, while the model hippocampus performs replays that represent planned path integration toward the specified goal.
We trained the model on various tasks involving short planning sequences. We found that it can plan nearly optimal paths from arbitrary starting to arbitrary goal states. Furthermore, additional training on longer planning sequences converged more quickly if the model was pre-trained on shorter sequences.
Also, we noticed the emergence of grid-like representations in the path integration network and a series of subgoal-like representations in the decision network. As such, this implies a more flexible and biologically plausible model for goal-directed behaviors, and the representation that emerged matches empirical results in animal navigation and human goal-directed behavior studies.
The model as a whole provides a scalable proposal for how the hippocampus and orbitofrontal cortex might work together to plan and evaluate actions aimed at achieving arbitrary and time-varying goals, both spatial and non-spatial.

Disclosures: H. Cheng: None. **J.W. Brown:** None.

Poster

573. Spatial Cognition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 573.09

Topic: H.12. Aging and Development

Support: McKnight Brain Research Foundation
NIH grants AG055544
NIH grants MH109548

Title: Theta Dominates Cross-frequency Coupling in Hippocampal-Medial Entorhinal Circuit During Run and Sleep in Rats

Authors: *Y. ZHOU[1,2], A. SHEREMET[3], J. P. KENNEDY[4], Y. QIN[5], N. M. DICOLA[6], S. D. LOVETT[6], S. N. BURKE[6], A. P. MAURER[7];
[1]Yale university, New Haven, CT; [3]Engin. Sch. for Sustainable Infrastructure and Envrn. (ESSIE), [2]Univ. of Florida, Gainesville, FL; [4]Neurosci., Univ. of Florida, Jacksonville, FL; [5]Civil & Coastal Eng., [6]Neurosci., [7]Evelyn F. McKnight Brain Inst., Univ. of Florida, Gainesville, FL

Abstract: Hippocampal theta and gamma rhythms are hypothesized to play a key role in the physiology of higher cognition. Prior research has reported that an offset in theta cycles between the entorhinal cortex, CA3, and CA1 regions potentially promotes independence of population activity across the hippocampus (HPC). In line with this idea, it has recently been observed that CA1 pyramidal cells can establish and maintain coordinated place cell activity intrinsically, with minimal reliance on afferent input. Counter to these observations is the contemporary hypothesis that CA1 neuron activity is driven by a slow gamma oscillation arising from CA3 and a fast gamma reflecting the input from the medial entorhinal cortex (MEC), with both rhythms providing a millisecond-level of synchrony across regions. In reinvestigating this, we recorded local field potentials simultaneously in the HPC and MEC from rats during maze-running and REM sleep. We found theta dominates cross-frequency coupling between the MEC and hippocampus, with gamma coupling falling to levels expected by a regional independence model.

Disclosures: Y. Zhou: None. **A. Sheremet:** None. **J.P. Kennedy:** None. **Y. Qin:** None. **N.M. Dicola:** None. **S.D. Lovett:** None. **S.N. Burke:** None. **A.P. Maurer:** None.

Poster

573. Spatial Cognition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 573.10

Topic: H.12. Aging and Development

Support: NIA R01 AG049722
NIH T32 AG020499-16A1

McKnight Brain Research Foundation
Ed and Ethel Moore Alzheimer's Disease Research Program

Title: Elucidating the role of hippocampal and perirhinal connectivity in a paired associates learning task

Authors: *T. COOPER, C. LOGAN, A. BROTGANDEL, B. PARRA, M. RAMIREZ, C. WATSON, S. BURKE;
Univ. of Florida Dept. of Neurosci., Gainesville, FL

Abstract: Higher cognitive function is supported by functional connectivity between several brain regions that work together to support complex behaviors. These regions are highly vulnerable to neurodegenerative diseases and aging and can limit the ability of older adults to live independently. The prefrontal cortex, hippocampus, lateral entorhinal cortex, and perirhinal cortex have been identified as areas crucial for learning and memory, and degeneration of these regions is associated with Alzheimer's Disease and other dementias. However, their individual and combined function in paired associate learning that involves integrating information about identity and location of a stimulus is not yet understood. This current study uses a version of the CANTAB paired associate learning (PAL) task adapted to a rodent model to further elucidate the role of each of these brain regions in a paired associate learning (PAL) task and how this learning is influenced by age. Pilot data suggested that hippocampus and perirhinal function are correlated to PAL performance, but prefrontal cortex and lateral entorhinal cortex inactivation did not significantly affect behavior. The current study builds upon this finding by using Designer Receptors Activated by Designer Drugs (DREADDs) to specifically target muscarinic G-protein coupled receptors in the perirhinal cortex through infusion of actuators into dorsal hippocampus to selectively inactivate perirhinal-hippocampal connections and evaluate behavioral changes due to loss of functional connectivity between these two key regions. Preliminary data suggests that the ability to learn object-location associations and subsequent performance on the PAL task relies upon communication between the perirhinal cortex and CA1.

Disclosures: T. Cooper: None. **C. Logan:** None. **A. Brotgandel:** None. **B. Parra:** None. **M. Ramirez:** None. **C. Watson:** None. **S. Burke:** None.

Poster

573. Spatial Cognition

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 573.11

Topic: H.12. Aging and Development

Support: NIH/NIMH R01MH126236
MBRF 0011249

Title: Cross-frequency interactions in rodent hippocampal LFP and cortical EEG

Authors: *S. D. LOVETT[1,2], N. M. DICOLA[1,2], M. S. LOVETT[1,2], J. P. KENNEDY[1,2], A. BROTGANDEL[1,2], K. L. ROBERTSON[1,2], S. N. BURKE[1,2,3], A. P. MAURER[1,2,3,4,5];
[1]Neurosci., Univ. of Florida, Gainesville, FL; [2]McKnight Brain Inst., Gainesville, FL; [3]Ctr. for Cognitive Aging and Memory, Gainesville, FL; [4]Dept. of Biomed. Engin., Gainesville, FL; [5]Engin. Sch. of Sustainable Infrastructure and Envrn., Gainesville, FL

Abstract: Common neurophysiological approaches in rodents tend to implement intercortical electrodes to measure the local field potentials (LFP). Human neurophysiological monitoring, on the other hand, is often measured through the use of non-invasive scalp EEG recordings. While EEG recordings can also come from rodents, there is a dearth of approaches that relate scalp EEG to the intracortical LFP. Our experimental goal is to derive a relationship between these approaches by demonstrating how EEG recorded through stainless steel screws placed on the skull surface can correlate with the hippocampal LFP obtained from intracranial silicon probes. After training five male Fisher Brown Norway hybrid rats to appetitively run on a circular track for a food reward, they were implanted with an intracranial silicon probe in the CA1 region of the dorsal hippocampus (AP: -3.2 ML: 1.5 DV: 4.0), and three screw electrodes were adhered to the skull surface, without fully penetrating the bone: One screw over the contralateral prefrontal cortex (AP: 3.3 ML: -0.7), one over the ipsilateral prefrontal cortex (AP: 2.7 ML: 0.7), and one over the ipsilateral parietal cortex (AP: -4.5 ML: 4.0). After a recovery period, the rats were reintroduced to the circular track, and data was then collected from each site. We observed that signals from the CA1 pyramidal layer, the CA1 stratum lacunosum moleculare (LM) layer, the prefrontal cortices, and the parietal region were modulated by running speed. Theta power increased as a function of velocity across all regions, and power-power correlations showed a strong coupling between theta frequencies and their harmonics. Interestingly, there was little variability between the cortical recordings, despite their spatial distance from each other, suggesting a correlation between hippocampal LFP and cortical EEG. We believe that further exploration of these findings may advance diagnostic inferences of hippocampal integrity using scalp EEG.

Disclosures: S.D. Lovett: None. **N.M. Dicola:** None. **M.S. Lovett:** None. **J.P. Kennedy:** None. **A. Brotgandel:** None. **K.L. Robertson:** None. **S.N. Burke:** None. **A.P. Maurer:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.01

Topic: H.12. Aging and Development

Support: NIH/NIMH K99/R00-MH101234
NIH/NIMH R01-MH116008
NIH/NIA R01-AG059028
NIH/NIA RF1-AG043640
NIH/NIA RF1-AG062831

Title: Changes to the morpho-electric properties of layer 3 pyramidal neurons of the rhesus monkey anterior cingulate cortex during aging

Authors: *C. A. MOJICA, D. DE ALBA, B. J. SNYDER, W. CHANG, A. TSOLIAS, T. L. MOORE, D. L. ROSENE, J. I. LUEBKE, M. MEDALLA;
Anat. & Neurobio., Boston Univ. Sch. of Med., Boston, MA

Abstract: A decline in some executive functions, such as working memory and cognitive flexibility is associated with normal aging, with the onset of deficits observed during middle age in both humans and non-human primates. These higher-order capacities are primarily mediated by the prefrontal cortex and involve layer 2/3 cortico-cortical connections in the lateral prefrontal cortex (LPFC) and in the anterior cingulate cortex (ACC). While previous work on the LPFC of rhesus monkeys has shown an age-related increase in excitability, alteration in synaptic currents, and reduction in spine density in layer 3 (L3) pyramidal neurons, the effects of aging on the structure and function of ACC pyramidal neurons are unknown. To this end, we characterized the morpho-electric properties of L3 pyramidal neurons in the ACC of rhesus monkeys across the adult lifespan (n=22, 6.0-27.5 yrs, 9 males and 13 females) using whole-cell patch clamp recordings and morphological reconstructions. Linear regression analysis show that input resistance increased (R^2=0.042, p<0.05) from young age (6 yrs) through the onset of old age (21 yrs). Resting membrane potential increased (R^2=0.07, p=0.0014), while rheobase decreased significantly with age (R^2=0.081, p<0.001). These changes in passive and active membrane properties indicate increased excitability that may be associated with changes in ion channel expression levels and in the structural compartments of the neuron. Whole-neuron reconstructions revealed no significant effect of aging on apical dendritic length, but a significant increase in the basal dendritic length of ACC pyramidal neurons across age (R^2=0.212, p=0.018). Neither apical nor basal dendritic branch number differed with age, but basilar branch density was significantly lower for aged neurons (R^2=0.346, p=0.0016), which implies decreased complexity. In addition, we observed reduced spine density (R^2=0.187, p=0.0243) on the apical but not the basal dendrites of aged compared to young neurons, with a significant decrease specifically in the number of thin and mushroom spine subtypes. Since postsynaptic spines are the sites of most excitatory synapses and are important in synaptic plasticity, an age-related reduction in spine density in the dendritic arbors of ACC neurons would be expected to significantly affect the signaling properties of these neurons during aging. Changes in LPFC pyramidal neuron properties significantly correlate with working memory deficits in aging. It is imperative to assess whether alterations in the morpho-electric properties of ACC neurons will correlate with similar or distinct aspects of age-associated impairment in executive function.

Disclosures: C.A. Mojica: None. **D. De Alba:** None. **B.J. Snyder:** None. **W. Chang:** None. **A. Tsolias:** None. **T.L. Moore:** None. **D.L. Rosene:** None. **J.I. Luebke:** None. **M. Medalla:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.02

Topic: H.12. Aging and Development

Support: NIH Grant R01AG059028

Title: Altered action potential propagation caused by age-related myelin dystrophies leads to impaired working memory in dlPFC neuron and circuit models

Authors: *N. SENGUPTA[1], S. IBAÑEZ[2], M. MEDALLA[1], K. WIMMER[2], J. I. LUEBKE[1], C. M. WEAVER[3];
[1]Anat. & Neurobio., Boston Univ. Sch. of Med., Boston, MA; [2]Ctr. de Recerca Matemàtica, Bellaterra (Barcelona), Spain; [3]Mathematics, Franklin & Marshall Col., Lancaster, PA

Abstract: Working memory, vital for complex cognitive tasks, is often impaired with aging in human and non-human primates. Behavioral studies involving spatial working memory have shown cognitive decline across the adult life span in rhesus monkeys. The action potential (AP) firing activity of pyramidal neurons in the dorsolateral prefrontal cortex (dlPFC) of these animals is crucial for spatial working memory. These neurons undergo structural and physiological changes with aging, some of which are associated with cognitive decline. Computational modeling can help to evaluate how various changes affect the function of individual neurons and networks that they form. This study quantifies how specific myelin dystrophies (namely demyelination, remyelination, and occasional excess myelination), which have been observed with aging in the rhesus dlPFC, affect AP propagation. Using the NEURON simulation framework, we integrated two published neuron models into a multicompartment model with a soma, dendritic arbor, and detailed axon with nodes and myelinated segments comprising paranodes, juxtaparanodes and internodes each with tight junctions. We identified a broad cohort of these models with plausible combinations of morphological and biophysical axon parameters, constrained by empirical data including conduction velocity (CV) of propagating APs and *in vitro* firing rates (FR). Subsequently we simulated several dystrophic myelin conditions, having perturbation magnitudes consistent with past empirical observations. When simulating demyelination alone, with 25% of internodes losing half of their myelin lamellae, CV was 5% lower. Complete demyelination of affected segments occasionally led to AP failure. During remyelination, a fraction of myelinated segments are replaced by shorter and thinner ones. When simulating this condition, remyelinating 45% of segments resulted in CV being 8% lower. Empirical observations suggest that aged axons exhibit both remyelinated internodes and occasional internodes with increased myelin lamellae. Our simulations of this scenario revealed that the additional lamellae partially compensate the CV reduction, therefore reducing conduction delay. Finally, we incorporated these conduction delays and AP failures into a spiking neuron network model of spatial working memory, and found some of these changes were sufficient to disrupt working memory precision. Future studies will combine myelin changes with other structural and physiological changes known to occur with normal aging in dlPFC pyramidal neurons.

Disclosures: **N. Sengupta:** None. **S. Ibañez:** None. **M. Medalla:** None. **K. Wimmer:** None. **J.I. Luebke:** None. **C.M. Weaver:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.03

Topic: H.12. Aging and Development

Support: R01 AG003376
McKnight Brain Research Foundation

Title: Reduced intra- and inter-regional heterogeneity in the human MTL subfields underly CA1 remapping deficits in normal aging

Authors: *L. ZHENG[1], S. DONER[1], M. R. FORLOINES[2], A. OYAO[1], A. D. EKSTROM[1], C. A. BARNES[1,3];
[1]Psychology, The Univ. of Arizona, Tucson, AZ; [2]Univ. of California, Davis, Citrus Heights, CA; [3]Evelyn F. McKnight Brain Inst., Tucson, AZ

Abstract: Healthy aging is accompanied by spatial memory declines, with animal studies suggesting that some of these age-related differences in spatial memory may relate to differences in hippocampal "remapping." Hippocampal remapping involves changing representations to be consistent with the current environment, which previous studies suggest may be deficient in older rats. This idea, however, has not been tested in aged humans. To address this issue, younger and older adults watched videos of virtual environments to learn the locations of stores. They then retrieved the details of stores[BCA(1] within a single environment during high-resolution functional magnetic resonance imaging (fMRI). Behavioral results demonstrate significant impairment of spatial memory in older adults. [MOU2] [MOU3] Using representational similarity analysis (RSA), we found that younger adults showed environment-specific neural codes (remapping) in CA1 whereas this effect was not observed in older adults, consistent with differences in hippocampus remapping in healthy older adults. In addition, older adults showed greater variability of in pattern similarity than did younger adults in CA1. This less stable neural representation in CA1 is a possible contributor to the underlying failure of remapping in older adults. Moreover, using a principal component analysis, we also found that older adults showed reduced intra-regional heterogeneity compared to younger adults, which was further negatively correlated with pattern similarity and memory performance. Together, these findings indicate that lower representational complexity resulted in worse behavioral performance in the task. Finally, using informational connectivity to assess inter-subfield interactions, our results revealed increased inter-regional coupling between MTL subregions in older compared to younger adults. These findings again suggested that the neural representations across MTL subregions were more homogeneous in older adults than in younger adults. In particular, the higher inter-regional coupling between PRC and ERC or ERC and CA1 was associated with worse memory performance. Taken together, these observations help to provide a better mechanistic understanding of spatial memory declines in normal aging, suggesting both age-variant and age-invariant differences.

Disclosures: L. Zheng: None. **S. Doner:** None. **M.R. Forloines:** None. **A. Oyao:** None. **A.D. Ekstrom:** None. **C.A. Barnes:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.04

Topic: H.12. Aging and Development

Support: R01 AG003376
McKnight Brain Research Foundation
P51RR000169
Arizona Alzheimer's Consortium, Department of Health Services

Title: Establishing an immunohistochemical protocol for visualization and quantification of noradrenergic fibers and localization of α1, α2, and β receptors in the hippocampus of macaque monkeys

Authors: *K. MCDERMOTT[1], I. SINAKEVITCH[1], S. KHATTAB[1], C. A. BARNES[1,2];
[1]Evelyn F. McKnight Brain Inst., Tucson, AZ; [2], Dept. of Psychology, Neurol. and Neurosciences, Univ. of Arizona, Tucson, AZ

Abstract: The Locus Coeruleus (LC) is a noradrenaline (NA)-producing brainstem nucleus with wide projections throughout the cortex. NA acts via 3 classes of receptors (α1, α2, β) and this signaling is critical for optimization of cognitive performance. Some histological studies have suggested age-related decreases in NA fiber and varicosity density in the cortex, and autoradiography studies have shown age-related decreases in α1 and α2 receptor densities. NA fiber density has not been investigated with density of all 3 NA receptor types, and no protocol exists for histological analysis of NA receptors in primates. In our studies we establish such a protocol for analysis of the NA system in rhesus macaques, while will be of great use for normative aging research. Our research utilizes coronal brainstem sections from a colony of 30 adult and aged rhesus macaque monkeys. We use commercially available primary antibodies: anti Dopamine Beta Hydroxylase (DBH, a marker for noradrenergic innervation), anti-α1 receptors, anti-α2a receptors, and anti- β1 receptors. Antibodies were tested at various concentrations, with and without antigen retrieval protocols, and with various types of secondary antibodies. We performed extended controls provide evidence of specific staining with each antibody. Receptor and DBH stains were then stained together in sequential sections for colocalization analysis. Reliable staining of NA fibers and varicosities were found homogenously projecting in all layers of the hippocampus using anti DBH antibody (1:1000, Millipore Sigma AB1537). α1 receptors (1:250, Millipore Sigma A270) were found on cell bodies in all layers of the hippocampus. α2a receptors (1:250, Abcam AB85770) were found predominantly on cell bodies and extended fibers, as well as colocalized with DBH+ fibers. β1 receptors (1:500, Abcam AB3442) were found predominantly on cell bodies and were the least dense of the three receptor types. Tests showed no undesirable interactions between layers of primary and secondary antibodies. Control tests indicate specific staining for each antibody. Thus far, we have developed a staining protocol for the visualization of noradrenergic fibers and

varicosities and receptors in the primate hippocampus. This protocol also includes markers for astrocytes, microglia, vessels, and DAPI for visualization of receptors on different cell types and in different cell layers of the hippocampus. A serial staining protocol such as the one developed here will allow for analysis of age-related changes in the LC-NA system of primates.

Disclosures: K. McDermott: None. **I. Sinakevitch:** None. **S. Khattab:** None. **C.A. Barnes:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.05

Topic: H.12. Aging and Development

Support: McKnight Brain Research Foundation

Title: Age-related changes in performance on the frontal cortex-dependent temporal order memory task

Authors: *O. GUSWILER[1], S. KHATTAB[1], K. BOHNE[1], C. A. BARNES[1,2];
[1]Evelyn F. McKnight Brain Inst., Tucson, AZ; [2]Dept. of Psychology, Neurol. and Neurosciences, Univ. of Arizona, Tucson, AZ

Abstract: It is known that cognitive performance declines with normative aging in several domains, but few studies have attempted to tease apart the specific regions that may be more susceptible to or robust against age-related changes over time. The temporal order memory (TOR) task is a simple and efficient test used to assess recognition memory, specifically the ability to recall when an object or event was committed to memory. In aging humans, prefrontal cortex-dependent memory exhibits some of the most dramatic and early changes relative to other brain functions (Park et al., Psychology and Aging, 2002, 17:299). Previous work has shown that lesions to the medial prefrontal cortex (mPFC) significantly disrupt performance on this task in young rats (Barker et al. J. Neurosci, 2007, 27:2948). Although the TOR task has been utilized in many studies, there has been little to no research on the effect of age on performance. We therefore investigated the sensitivity of the TOR task to detect age differences in a rodent model of normative aging. We tested male Fischer 344 (F344) rats of two separate ages, young (9mo) and old (23-27mo). Animals were exposed to the test box for 10 minutes for two consecutive days. On days 3 and 4, they were exposed first to two identical objects (i.e., A and A) for 4 minutes. One hour after this, the rats were exposed to two different identical objects (i.e., B, B) for 4 minutes. Two hours following the second sample phase, the test phase began during which the rat was presented with one copy of both objects (i.e., A and B). In adult rats, the object from the first sample phase (A) is explored more, indicating that the rat recognizes that this object was explored during a temporal window that was more remote than the second object explored. Our results indicate a trend for the aged rats to be impaired compared to the younger animals in their

ability to recognize which object was most recently experienced. This simple temporal order memory task will be extremely useful for further exploration of the differences between young and aged individuals, particularly in combination with high-density cell recordings in the mPFC and related structures.

Disclosures: O. Guswiler: None. **S. Khattab:** None. **K. Bohne:** None. **C.A. Barnes:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.06

Topic: H.12. Aging and Development

Support: R01 AG049465
McKnight Brain Research Foundation

Title: Volumetric MRI Analysis of Rodent Brains as a Function of Age and Cognition

Authors: *L. DO[1], M. A. ZEMPARE[4], A. S. BERNSTEIN[1], P. K. BHARADWAJ[2], N. CAREY[4], C. NGUYEN[4], G. E. ALEXANDER[4,2], C. A. BARNES[4,3], T. P. TROUARD[4,1]; [1]Dept. of Biomed. Engin., [2]Psychology, [3]Dept. of Psychology, Neurol. and Neurosciences, Univ. of Arizona, Tucson, AZ; [4]Evelyn F. McKnight Brain Inst., Tucson, AZ

Abstract: Animal models play an important role in preclinical and translational studies of the human brain. Magnetic Resonance Imaging (MRI) is a non-invasive technique that has the potential to provide reliable anatomical measures of the brain. Atlas-based tools for neuro-informatics can be utilized in studying age-dependent changes in brain anatomy associated with cognitive function in both animal models and in humans. Here we present results from a large cross-sectional study employing a rodent model of normative aging to investigate the correlates of healthy cognitive aging. Initial analysis of MRI data utilized a rat brain template and associated atlas (Goerzen et al. 2020, Sci Rep 10:6952.) for comparison of rodent brain volumes both globally and regionally at 3 ages across the lifespan and at 3 levels of cognitive status within each age category. Male Fisher 344 rats (n=114) were acquired at young adult (6 months, n=48), middle aged (15 months, n= 38) and old adult (23 months, n= 28) ages. These rats underwent a battery of behavioral tasks over the course of 6 weeks resulting in each age group being sub-divided into 3 categorized subgroups of high, average, and low cognition using a corrected integrated pathlength score from the spatial version of the hippocampus-dependent Morris watermaze task. At the end of the 6 weeks, body weights were measured, and neurological MRI was carried out. Whole-brain T2-weighted images were collected at 150μm isotropic resolution. Images underwent brain extraction using a semi-automated process as well as bias correction due to non-uniform surface coil sensitivity. A Fischer 344 T2-weighted template image (60 μm isotropic voxels) and its labeled atlas (115 regions) were registered to each individual animal in the study using linear and non-linear registration. Two factor ANOVA

models were used to assess differences in regional brain volumes, and total intracranial volumes across age and cognition groups. Significant main effect for age group was observed for total intracranial volume ($p<0.0001$) and hippocampus volume ($p<0.0001$). No significant interaction effects, or main effects of cognitive status were observed. While there are many other ROIs to consider further, these initial findings indicate that intracranial volume and hippocampus volume are good predictors of the age of the animal, the intracranial volume is still increasing through middle age while the hippocampus continues to significantly increase into old age, however, the volume of the hippocampus is not related to cognitive status of the animals in these analyses.

Disclosures: L. Do: None. **M.A. Zempare:** None. **A.S. Bernstein:** None. **P.K. Bharadwaj:** None. **N. Carey:** None. **C. Nguyen:** None. **G.E. Alexander:** None. **C.A. Barnes:** None. **T.P. Trouard:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.07

Topic: H.12. Aging and Development

Support: McKnight Brain Research Foundation
NIH grant AG049465

Title: Identification of transcriptional patterns in hippocampus CA1 subregion associated with differential cognitive aptitude across the entire lifespan of rats

Authors: *M. CHAWLA[1], Y. J. CHEN[1], M. ZEMPARE[1], C. A. BARNES[1,2], A. DALMENDRAY[1], K. YOUNG[1];
[1]Evelyn F. McKnight Brain Inst., Tucson, AZ; [2]Dept. of Psychology, Neurol. and Neurosciences, Univ. of Arizona, Tucson, AZ

Abstract: Each hippocampus primary cell type has a unique transcriptomic composition. Therefore, it is possible that CA1 and CA3 pyramidal cells or DG granule cells may have distinct age-sensitive trajectories. Additionally, these trajectories of cognitive decline may depend on the cognitive status of individual rats. Here we utilize the immediate early gene *Arc* to assess the transcription pattern in cognitively categorized rats. Male Fisher-344 rats (6 mo, 15 mo, and 23 mo old), were given a battery of cognitive tests and were categorized into three groups - low, average, or high performers depending on their performance on the spatial version of the Morris watermaze. Rats were given two-5 min exploratory sessions separated by a 20 min rest in the home cage and brains from behavior as well as two additional controls (caged, a negative control, and maximal electroconvulsive shock-treated, a positive control) were quickly extracted, hemisected and rapidly frozen until sectioning and processing for *in situ* hybridization. *In situ* hybridization was performed as described previously (Guzowski et al., 1999), and slides were imaged using an SP5 Leica or a Zeiss LSM 880 confocal microscope. Three different areas of

CA1 were imaged: distal CA1, which receives input primarily from the lateral entorhinal cortex; proximal CA1, which receives inputs primarily from the medial entorhinal cortex; and middle CA1, which receives a mixture of entorhinal inputs. Cells with *Arc* mRNA expression in the nucleus, cytoplasm or both compartments were counted using Image J software. Since cell-counting is still ongoing, we can report that *Arc* mRNA expression is following a pattern similar to that of previous studies; that is, very low *Arc* expression in caged controls, robust expression in maximal electroconvulsive shock-treated rats and intermediate in behavior-treated rats. Using compartment analysis of temporal activity using FISH (catFISH) we can determine the reliability of cell firing in brain networks by two experiences in the same environment separated by 20 minutes. In this manner, we can assess the circuit stability of these specific brain regions, across age and across different cognitive competences. The overall goal is to identify the circuit characteristics associated with successful cognitive outcomes during normal aging.

Disclosures: M. Chawla: None. **Y.J. Chen:** None. **M. Zempare:** None. **C.A. Barnes:** None. **A. Dalmendray:** None. **K. Young:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.08

Topic: H.12. Aging and Development

Support: R01 AG003376
McKnight Brain Research Foundation
P51RR000169
Arizona Alzheimer's Consortium, Department of Health Services

Title: A high-resolution 3D reconstruction of the locus coeruleus in aged macaques: a combined MRI, Nissl and anti-Tyrosine Hydroxylase (TH) immunofluorescence study

Authors: *I. SINAKEVITCH[1], K. MCDERMOTT[1], S. KHATTAB[1], D. T. GRAY[1,2], C. A. BARNES[1,3];
[1]Evelyn F. McKnight Brain Inst., Tucson, AZ; [2]Dept. of Physiol., UCLA, Los Angeles, CA; [3]Dept. of Psychology, Neurol. and Neurosciences, Univ. of Arizona, Tucson, AZ

Abstract: The Locus Coeruleus (LC) is a brainstem nucleus with the largest group of noradrenaline producing neurons. Dysregulation of LC systems contributes to cognitive dysfunctions in both healthy aged brains and brains that succumb to Alzheimer's disease. Notably, the LC is heterogeneous along the rostral-caudal and dorsal-ventral axes with respect to neuron morphology, projection targets, and vulnerability to the impact of normative brain aging and neurodegenerative disease. In previous studies in our laboratory, we identified three distinct subnuclei in the macaque LC: a medial nucleus that was confined to the central gray area, a lateral nucleus that lies outside of the gray area laterally and blends within the mesencephalic

tract (me5), and a compact area within the medial nucleus. In this study, we describe in detail the 3D anatomy of the LC nucleus using the Nissl, Tyrosine hydroxylase (TH)-immunoreactivity, and MRI data of one macaque. Next, we describe the neuroarchitecture of the long-range processes of TH-positive LC neurons in the midbrain. Finally, we establish a protocol using AMIRA software for counting cells along the rostral-caudal axis and within the described compartments of LC. AMIRA software was employed to reconstruct the LC from the TH-immunofluorescence and Nissl sections that were aligned with previously collected *in vivo* structural MRI data. The macaque LC proper nucleus extends approximately 2.4mm along the rostro-caudal axis with an overall volume up to $3mm^3$. We also found that the extranuclear LC processes extend laterally to an adjacent area in the midbrain and surrounds the lateral and dorsal superior cerebellar peduncle. AMIRA software was also used to analyze the cell counts in each compartment. To accomplish this, LC TH-positive cells were first segmented manually to determine the range of possible cell dimensions to use for later segmentation. Next, all TH-positive neurons and processes were segmented using automatic AMIRA procedures, and the previously established cell dimensions were used to select putative cells within LC compartments. These automatic cell counts were compared with manual cell counts from within the LC nucleus to verify their accuracy. In conclusion, this analysis pipeline will allow us to standardize our data on adult and aged macaques to find out the specific sites of vulnerability along the rostral-caudal axis of the LC compartments. Further neuronal analyses will be aimed at understanding the mechanisms responsible for LC vulnerability and its impacts on cognition in normative aging and disease.

Disclosures: I. Sinakevitch: None. **K. McDermott:** None. **S. Khattab:** None. **D.T. Gray:** None. **C.A. Barnes:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.09

Topic: H.12. Aging and Development

Support: McKnight Brain Research Foundation
R01 AG049464

Title: Diffusion-weighted mri and cognitive evaluation of the effects of induced hypertension in middle aged cyp1a1-ren2 transgenic rats

Authors: *M. ZEMPARE[1], N. J. CAREY[1], A. L. DALMENDRAY[1], K. YOUNG[1], K. M. BOHNE[1], H. WISKOSKI[2], L. DO[2], T. P. TROUARD[2,1], M. K. CHAWLA[1], K. D. MITCHELL[4], M. J. HUENTELMAN[5,1], C. A. BARNES[1,3];
[1]Evelyn F. McKnight Brain Inst., Tucson, AZ; [2]Biomed. Engin., [3]Dept. of Psychology, Neurol. and Neurosciences, Univ. of Arizona, Tucson, AZ; [4]Tulane Univ. Hlth. Sci. Ctr., New Orleans, LA; [5]The Translational Genomics Res. Inst., Phoenix, AZ

Abstract: Hypertension (HTN) is associated with an increased risk of cardiovascular disease (CVD) and cognitive decline in aging humans, with the onset occurring around middle-age. While prior research has suggested an association between CVD and cognitive decline in the elderly, it is also critical to investigate how this dynamic may evolve from middle to older age and the related changes in brain region-specific anatomy and function prior to and after onset of HTN. We have carried out diffusion weighted MRI (dMRI), a powerful non-invasive translational tool that can be utilized to analyze microstructural changes to the brain, and cognitive evaluation before and after induced HTN in transgenic rats. In this study, Cyp1a1-Ren2 xenobiotic-inducible transgenic rats (n=50) were used to model the gradual rate and age-of-onset of HTN observed in humans. 15-month-old male rats were assigned to either control (n=21) or treatment (n=29) groups and given a 6-week battery of behavior tests. The battery included the hippocampus-dependent spatial version of the Morris watermaze. Following the pre-treatment tests, the treatment group received a diet with 0.15% Indole-3-Carbinol (I3C) while control rats received the same chow without I3C. A post-treatment behavioral battery was given to assess the effect of HTN on cognition. Gradual onset of HTN was confirmed through systolic and diastolic blood pressure changes. Shortly after the pre- and post-treatment tests, body weights were measured, and neurological MRI was carried out. Multishell (b=1000, 2000 and 3000 s/mm2), multidirection (64) dMRI, was carried out with a resolution of 300x300x1000 μm. Fiber orientation distribution functions (FODs) were calculated via constrained spherical deconvolution (CSD) and used to register images to a study-specific template space. A T2-weighted template and labeled atlas (115 regions of the brain) were registered to the study-specific template space and values of Fractional Anisotropy (FA) and mean diffusivity were compared between groups and HTN status. The I3C-treated group showed a significant increase in cardiac and renal end organ damage as observed previously (Willeman et al. 2019, Physiol. Report, 7:2051). Analysis of the hippocampus region-specific Morris watermaze data indicate no significant changes in cognitive performance with renal-induced HTN. Consistent with the behavioral results, noninvasive imaging techniques also found no statistically significant differences in left (p=0.488) and right (p=0.7078) hippocampus FA of control versus I3C treated groups. The protection of brain and cognitive function in spite of HTN, may suggest a novel brain resilience mechanism.

Disclosures: **M. Zempare:** None. **N.J. Carey:** None. **A.L. Dalmendray:** None. **K. Young:** None. **K.M. Bohne:** None. **H. Wiskoski:** None. **L. Do:** None. **T.P. Trouard:** None. **M.K. Chawla:** None. **K.D. Mitchell:** None. **M.J. Huentelman:** None. **C.A. Barnes:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.10

Topic: H.12. Aging and Development

Support: R01 AG003376
McKnight Brain Research Foundation
Alzheimer's Association AARF-22-926755

Title: Age-related spatial navigation impairments are moderated by testing modality

Authors: *P. HILL[1], A. S. MCAVAN[1], C. A. BARNES[1,3,2], A. D. EKSTROM[1,3];
[1]Dept. of Psychology, [2]Dept. of Neurol. and Neurosciences, Univ. of Arizona, Tucson, AZ;
[3]Evelyn F. McKnight Brain Inst., Tucson, AZ

Abstract: Spatial navigation declines during healthy aging and is exacerbated by age-related neurodegenerative disorders, such as Alzheimer's disease. In humans, age-related navigation deficits have been attributed to morphological changes in the hippocampus and neighboring medial temporal lobe structures. Spatial computations in these regions are generally assumed to support formation of externally referenced (i.e., 'allocentric') map-like representations of the environment via gradual integration of idiothetic cues and visual landmarks. The few studies in humans to examine spatial navigation in older age have often relied on experimental paradigms that require using a keyboard/joystick to navigate a virtual environment. These types of paradigms restrict natural movements and idiothetic feedback and could be confounded by age-related differences in computer gaming experience and fine motor control. In the present study, cognitively healthy young (N=20) and older (N=20) adults navigated a virtual variant of the Morris watermaze task which is presumed to rely on hippocampal mediated allocentric navigation strategies. Participants learned the location of hidden targets in each of two conditions: a desktop-based condition requiring a keyboard/mouse to navigate and a fully immersive virtual reality (VR) condition that permitted unrestricted locomotion. Following a learning phase, participants were tested on their immediate and delayed recall of the hidden target locations. A significant age x condition interaction indicated that the effect of age on spatial memory was moderated by VR modality. Spatial recall was significantly reduced in older adults relative to younger individuals when navigating the desktop VR environment. By contrast, the two groups did not significantly differ when navigating the fully immersive VR environment. These results are consistent with the hypothesis that the effect of age on spatial navigation is modified by availability and integration of idiothetic cues and visual landmarks. Our results suggest that age differences in spatial navigation may be inflated when testing in desktop-based VR environments, which may be less familiar and natural for an older cohort. Reconciling how older navigators integrate multisensory cues is critical for understanding the biological significance of findings from navigation paradigms that restrict self-motion and body-based feedback, such as those used in an MRI environment.

Disclosures: P. Hill: None. **A.S. McAvan:** None. **C.A. Barnes:** None. **A.D. Ekstrom:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.11

Topic: H.12. Aging and Development

Support: McKnight Brain Research Foundation
Arizona Alzheimer's Consortium, Department of Health Services

Title: Investigating age-related changes of mPFC neural responses to ventral hippocampus stimulation

Authors: *S. SRIVATHSA[1], A. VISHWANATH[2], S. L. COWEN[1,2], C. A. BARNES[1,3];
[1]Evelyn F. McKnight Brain Inst., Tucson, AZ; [2]Psychology, [3]Dept. of Psychology, Neurol. and Neurosciences, Univ. of Arizona, Tucson, AZ

Abstract: Neural ensembles in the hippocampus and medial prefrontal cortex (mPFC) play a crucial role in spatial working memory, a process susceptible to decline during aging. These regions are connected via a monosynaptic, unidirectional projection from the ventral hippocampus (vHC) to the mPFC (Jay and Witter, 1991, J. Com. Neurol. 313:574), and damage or inhibition of this connection leads to impairments in spatial working memory tasks. Performance on spatial working memory tasks is known to correlate with increased synchrony of vHC theta (8-12 Hz) rhythms to mPFC neuron activity and is also coupled with mPFC local theta and gamma (30-100 Hz) oscillations. Additionally, the temporal offset of mPFC neurons phase-locking to vHC theta corresponds to the conduction delay between vHC and mPFC neurons, suggesting that the vHC-mPFC synchronization is a direct result of this projection. In the mPFC of aged rats, the frequency of gamma oscillations has been shown to be reduced. These gamma oscillations are known to result from an interaction between fast spiking inhibitory interneurons and their local excitatory neuron targets. The interactions between inhibitory and excitatory neurons are reduced by 1-2 msec in old rats. Little is understood about how monosynaptic vHC inputs engage mPFC, how this changes with age, or how vHC activation differentially affects neural activity along the dorsal-ventral axis of the mPFC. To investigate these questions, we delivered electrical pulses to the CA1 layer of vHC in anesthetized male F344 young (9 months) and old (27 months) rats while simultaneously recording neural activity along the dorsoventral length of the mPFC using Neuropixels probes. Recordings were obtained from neurons spanning 5.5 mm along the mPFC, including the anterior cingulate cortex, prelimbic, and infralimbic regions (areas 24b, 32, and 25). Since vHC projections as well as general excitability characteristics are not uniform across the mPFC (Liu and Carter, 2018, J. Neurosci. 38:7351), we also compare neural activity across different layers of mPFC in response to vHC stimulation by recording from layer V and II/III in mPFC. This allows a comparison of the effects of direct vHC axonal input onto different layers and cell types within the three mPFC subregions recorded, as well as whether these connections are altered by aging.

Disclosures: S. Srivathsa: None. **A. Vishwanath:** None. **S.L. Cowen:** None. **C.A. Barnes:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.12

Topic: H.12. Aging and Development

Support: McKnight Brain Research Foundation
Arizona Alzheimer's Consortium, Department of Health Services
UA translational bioimaging resource (TBIR)
NIH small instrumentation grant (S10 OD025016)

Title: Comparative Analysis of Microstructural Features of Bonnet Macaque Hippocampal Subfields Using MRI Microscopy

Authors: *L. DIECKHAUS[1], K. MCDERMOTT[4], D. T. GRAY[4,5], C. A. BARNES[4,2], A. ECKSTROM[4,3], A. MURLIKRISHNAN[1], E. B. HUTICHINSON[1];
[1]Dept. of Biomed. Engin., [2]Dept. of Psychology, Neurol. and Neurosciences, [3]Dept. of Psychology, Univ. of Arizona, Tucson, AZ; [4]Evelyn F. McKnight Brain Inst., Tucson, AZ; [5]Dept. of Physiol., David Geffen Sch. of Med. at UCLA, Los Angeles, CA

Abstract: The subfields of the hippocampus are highly organized with prominent microscale features that may be detectable by diffusion and relaxometry-based MRI techniques. Understanding microstructural variations between hippocampal subfields is important to assessing subtle structural changes that may have behavioral correlates or lead to the discovery of biomarkers in brain disorders. Nonhuman primates are an excellent animal model to investigate subregion- specific differences in MRI metrics because of their similar microstructural features to the human brain and can be used for radiologic-pathologic validation studies. We performed high resolution *ex vivo* imaging (600μ isotropic) on perfused whole brains of 8 behaviorally characterized female bonnet macaques ranging from 10 to 25 years old (30 to 75 human equivalent years). Multiple MRI metrics from diffusion and relaxometry-based techniques offered insight into brain microstructures to quantify myelin, macromolecules and iron content: Fractional Anisotropy (FA), Trace (TR), Myelin Water Fraction (MWF), Bound Pool Fraction (BPF), and R2* maps. High resolution maps were calculated for each brain and manual Region of Interest (ROI) segmentation of the hippocampus subfields: CA1, CA3, subiculum, and Dentate Gyrus (DG) were drawn using T2 weighted anatomical (200μ isotropic). The ROIs were drawn utilizing a technique utilized in human hippocampal subfield segmentation and used to extract values for FA, TR, R2*, MWF and BPF in each subfield. Spearman correlations revealed no correlation with age but did find significant correlations between some MR metrics for some subfields. FA and TR were significantly correlated in CA1 ($R=-0.83, p=0.01$) and DG ($R=-0.905, p=0.002$). Only 2 of the subfields had significant correlations between MWF and BPF: CA3 ($R=0.976, p=3.3\times10^{-5}$) and the subiculum ($R=0.833, p=0.01$). We have previously reported that at the whole hippocampal level, BPF and MWF were significantly correlated and this appears to be partially explained by these 2 specific regions that make up only 20% of voxels in the hippocampus. FA and TR were correlated in only 2 subfields (CA1, DG) while MWF and BPF were correlated with the other 2 subfields (CA3, subiculum). By analyzing the hippocampus subfields, we were able to identify two regions (CA3,subiculum) that potentially gave rise to the previous correlation between MWF and BPF of the whole hippocampus. These correlations have the potential offer further insight on the environmental landscape of the hippocampus that would otherwise be limited when only investigating volumetric changes or exclusively FA changes.

Disclosures: **L. Dieckhaus:** None. **K. McDermott:** None. **D.T. Gray:** None. **C.A. Barnes:** None. **A. Eckstrom:** None. **A. Murlikrishnan:** None. **E.B. Hutichinson:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.13

Topic: H.12. Aging and Development

Support: NIH/NIA R01AG038465-06

Title: Topological Data Analysis Predicts Longitudinal Behavioral Change of Fluid Reasoning in the RANN Cohort

Authors: *G. ARGIRIS[1], Y. STERN[2], C. G. HABECK[3];
[1]Cognitive Neurosci. Div., Columbia Univ., New York, NY; [2]Cognitive Neurosci. Div., Columbia Univ., New York City, NY; [3]Taub Inst., Columbia Univ., New York, NY

Abstract: Graph theoretic metrics have shown that increasing age has been associated with decreases in segregation of functional brain systems (e.g., Chan et al., 2014). Recent attention though has been given to topological data analysis (TDA) in order to identify the underlying shape of brain network connectivity beyond simple pairings of edges (see Sizemore et al., 2019). Persistent homology (PH) is one tool of TDA that allows for the computation of topological features across different connectivity thresholds, with one focus being on the number of connected components of 0-dimension, or Betti-0 numbers, used to create a curve that, when integrated over iterations (i.e., area under the curve; AUC), indicates how quickly a single connected component is formed; in principle, lower AUC should reflect overall fewer connected components and is thus analogous to lower brain segregation (e.g., Gracia-Tabuenca et al., 2020). In the present study, we applied PH to task-based functional connectivity data from the Reference Ability Neural Network (RANN) longitudinal lifespan cohort in order to 1.) test for individual factors that might predict change in the AUC of Betti-0 curves over time, 2.) predict change in behavioral performance based on change in Betti-0 curves, and 3.) compare systems segregation to the PH approach. One hundred and sixty-three participants were tested in-scanner at both baseline and five-year follow-up on a battery of tests comprising four domains of cognition (see Stern et al., 2014). Task-based functional connectivity values were calculated between a pre-defined set of 264 ROIs (nodes) according to Power et al.'s (2011) parcellation scheme. Results showed that, specifically in the Fluid Reasoning domain, the AUC of the Betti-0 curve significantly decreased with age. Systems segregation also significantly decreased with age across all four cognitive domains. However, when investigating the link between connectivity and behavior, only the AUC of the Betti-0 curve significantly predicted longitudinal cognitive change in the Fluid Reasoning domain. That is, an increase in how quickly a network formed a single component between time points was linked to behavioral decline. Furthermore, this finding was significant even after controlling for specific demographic factors in addition to

brain integrity and white matter hyperintensity burden. These results argue for the importance of TDA as an investigative tool for functional alterations across time and its link to cognitive outcomes.

Disclosures: G. Argiris: None. **Y. Stern:** None. **C.G. Habeck:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.14

Topic: H.12. Aging and Development

Support: NIA K01AG051777
AARG-17-529121
NIA R01AG038465
NIA R01AG026158

Title: Lower vulnerability for AD pathology for crystallized than for fluid abilities in older adults

Authors: *Y. GAZES, J. SAKHARDANDE, R. BABUKUTTY, C. G. HABECK; Columbia Univ. Irving Med. Ctr., New York, NY

Abstract: A well-known phenomenon in cognitive aging is the seemingly contradictory observation that cognitive abilities relying on crystallized intelligence are well maintained until as late as past 80 years old while other cognitive abilities that rely on fluid intelligence start to decline as early as 20 to 30 years old. Not only are crystallized abilities better preserved in aging, they are also more resilient to ADRD pathology than fluid abilities, as demonstrated by the use of vocabulary as an estimate of premorbid IQ. It is still unclear what mechanisms underly the disjuncture between crystallized and fluid abilities in aging and ADRD. To test the hypothesis that crystallized abilities are less vulnerable to AD pathology than fluid abilities, we examined the topographic overlap between the fMRI activation patterns with each participant's amyloid-ß/tau PET images. For 63 older adults, three vocabulary fMRI tasks were estimated with the fMRI patterns for crystallized ability, and patterns for three different fluid abilities (processing speed, reasoning, and episodic memory) were each estimated with three fMRI tasks. Any overlapping voxels between any of the ability patterns were excluded from the final fMRI patterns. Thus, each ability's fMRI pattern contained only voxels uniquely associated with each ability. Amyloid-ß was imaged with ^{18}F-Florbetaben PET scans. Topographic overlap between amyloid-β PET standardized uptake value ratio (SUVR) images and the fMRI activation pattern for each of the four abilities was calculated using dot product across non-zero voxels after grey matter masking. The resulting overlap score was normalized by the number of non-zero voxels for each ability. Vocabulary overlap scores were lower than the scores for the three fluid abilities with $p < .001$ for all pairings, and the effect sizes (Cohen's D) were: 7.36 for processing speed,

3.36 for reasoning, and .584 for episodic memory. Less topographic overlap between amyloid-ß and fMRI patterns for vocabulary than the three fluid abilities provided support for vocabulary's lower vulnerability to AD pathology compared to the three fluid abilities, suggesting that one possible mechanism for crystallized abilities' greater age-resilience may be in terms of vulnerability to AD pathology.

Disclosures: Y. Gazes: None. **J. Sakhardande:** None. **R. Babukutty:** None. **C.G. Habeck:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.15

Topic: H.12. Aging and Development

Title: Thinking in the Searchlight: Identifying Local Cognitive Representations in the Brain

Authors: *J. KABIR[1], D. LEEDS[1], C. G. HABECK[2], Y. STERN[3];
[1]Fordham Univ., New York, NY; [2]Taub Inst., Columbia Univ., New York, NY; [3]Columbia Univ., NY, NY

Abstract: Previous studies have indicated that four latent variables, or reference abilities (RAs), can serve as broader elements of cognition that can help frame age-related changes in brain activities: these being vocabulary, perceptual speed, fluid reasoning, and episodic memory. Our aim is to assess the RA groupings by locating regions in the brain supporting RA or other cognitive groupings. We further study whether these cognitive groupings are affected by age. Two hundred seventy-two clinically healthy participants performed 12 cognitive tests, three tasks for each reference ability while undergoing fMRI scans. The participants were divided into three groups: young (ages 20-40), middle (ages 41-60), and old (61-80). We analyzed the fMRI data by conducting voxel searchlight and representational similarity analysis on the localized voxel responses to each of twelve tasks, generating a 12x12 matrix comparing local task responses. Response patterns were analyzed from 5x5x5 voxel searchlight cubes. To identify brain regions that group tasks similarly, a k-means clustering algorithm was used on the 12x12 task matrices. Across all age groups, we found four frequent task groupings: (a) most commonly, similar voxel responses for all tasks; (b) similar voxel response patterns for vocabulary and perceptual speed RA, and a separate set of similar patterns for fluid reasoning and episodic memory RAs; (c) similar responses for all tasks and for all RAs except picture naming in the vocabulary RA; and, (d) similar voxel response patterns for tasks in fluid reasoning RA, and a separate weaker similarity among the three other RAs. These patterns were consistent across all age groups. These results support not only the existence of the initial four RAs but also suggest the existence of cognitive groups spanning multiple RAs. It also suggests localization of different cognitive processes in various regions of the brain, complementing past studies identifying cognitive ability networks across the brain.

Disclosures: **J. Kabir:** None. **D. Leeds:** None. **C.G. Habeck:** None. **Y. Stern:** None.

Poster

574. Neural Mechanisms of Aging II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 574.16

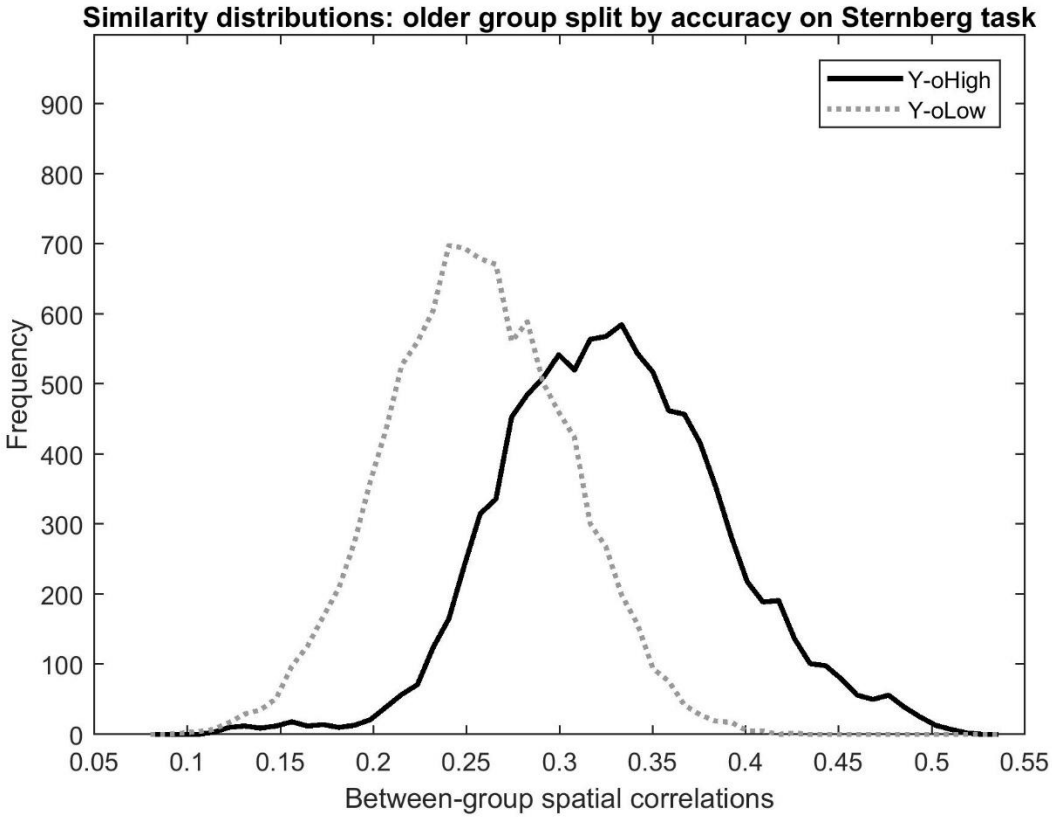
Topic: H.12. Aging and Development

Support: NIH grant R01AG026158

Title: Higher similarity to young activation profiles for higher-performing elders in verbal working memory

Authors: *C. G. HABECK, Y. GAZES, Y. STERN;
Columbia Univ., Columbia Univ., New York, NY

Abstract: We were interested to what extent older high-performing people show youthful-looking activation patterns and focused on the maintenance phase of a working-memory fMRI task for which participants studied an array of 1, 3, or 6 letters for 3 seconds, then maintained the letters while facing a blank screen for 7 seconds, with a subsequent probe to which the participants answered with a button press whether the probe letter was contained in the study array or not. 44 younger participants aged 20-30 performed the task, as well as 135 older participants aged 55-70. Ordinal Trend Canonical Variates analysis (OrT CVA; Habeck et al., 2005) derived activation patterns whose pattern scores increased with memory load within participant for as many participants as possible. There were 3 performance measures: (1) load-averaged response accuracy in Sternberg task, (2) load-averaged reaction time in Sternberg task, and (3) vocabulary ability. In the older group these measures were residualized with regard to age, sex, education, mean cortical thickness, and white-matter hyperintensities, with a subsequent median split of the residuals, leading to 3 groups: (1) young group (Y), (2) high-performing old group (oHigh), (3) low-performing old group (oLow). OrT CVA was applied to each group with bootstrap robustness computation of 100 iterations. From these patterns in each group, we generated two similarity distributions by computing 10,000 pairwise spatial correlations for the two pairings Y-oHigh and Y-oLow. For all 3 performance measures, the Y-oHigh similarity distribution was shifted to the right of the Y-oLow distribution. (See figure for the example of mean accuracy). These findings show that higher-performing older people manifest younger-looking activation patterns in a verbal working memory fMRI task. Similar findings were obtained when performing the median split in the older participants based on brain structure, but not on education which yielded no difference. These findings point to preservation of young-like activation patterns as a possible mechanism of Cognitive Reserve.



Disclosures: **C.G. Habeck:** None. **Y. Gazes:** None. **Y. Stern:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.01

Topic: H.13. Schizophrenia

Support: NIMH K00MH121382
NIMH R01MH124047

Title: Comprehensive and simultaneous 3-D imaging of interneuron subtypes in CA1 depicts deficits in interneuron activity resulting in microcircuit imbalance in a mouse model for the 22q11.2 deletion syndrome

Authors: *S. HERRLINGER[1], B. RAO[2], A. TUTTMAN[1], S. CHEN[1], M. SZOBOSZLAY[1], J. B. PRIESTLEY[3], E. VAROL[1], B. VANCURA[4], T. GEILLER[1], J. A. GOGOS[5], A.

LOSONCZY[1];
[1]Zuckerman Inst., [3]Dept. of Neurosci., [4]Neurosci., [2]Columbia Univ., New York, NY;
[5]Zuckerman Inst., Columbia Univ., New York City, NY

Abstract: Individuals with the 22q11.2 deletion syndrome (22q11.2DS), one of the strongest genetic risk factors for schizophrenia, demonstrate cognitive impairments, including episodic memory (EM) dysfunction. Our group previously showed that EM is impaired in a mouse model for the 22q11.2DS (*Df(16)A*$^{+/-}$). Place cells, cellular representations of EM, are under strong inhibitory control by heterogeneous subtypes of GABAergic interneurons, which have been implicated in the pathophysiology of schizophrenia. In this study, we examined the contribution of pyramidal cells and hippocampal interneuron subtypes to local microcircuit dysfunction in CA1. 2-photon imaging of CA1 pyramidal neuron population dynamics were performed *in vivo* to characterize plasticity during novel context exposure in a virtual environment in *Df(16)A*$^{+/-}$ and WT mice. Wild-type and *Df(16)A*$^{+/-}$ mice performed goal-oriented learning and random foraging tasks on a cued belt while undergoing large-scale, unbiased 3D GCaMP-Ca^{2+} imaging of *in vivo* CA1 interneuron dynamics. Molecular identification of major interneuron subtypes was performed post-hoc utilizing immunohistochemistry. Interneuron subtype activity was assessed through Pearson cross-correlations with velocity and through peristimulus time histograms around behavioral indicators. In *Df(16)A*$^{+/-}$ mice we observe a significant decrease in somatic bursting rate during context switch compared with WTs, suggesting that plasticity is suppressed *in vivo*. Interneurons exhibit subtype-specific alterations in activity during behavior. Results examining CA1 principal neuron dynamics and plasticity collected *in vivo* and *in vitro* suggest that inhibitory circuits are either over-compensating *in vivo* or are intrinsically deficient themselves. We identify subtype-specific alterations in interneuron dynamics, contributing to microcircuit imbalances.

Disclosures: S. Herrlinger: None. **B. Rao:** None. **A. Tuttman:** None. **S. Chen:** None. **M. Szoboszlay:** None. **J.B. Priestley:** None. **E. Varol:** None. **B. Vancura:** None. **T. Geiller:** None. **J.A. Gogos:** None. **A. Losonczy:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.02

Topic: H.13. Schizophrenia

Title: Src-heterozygous mice show alterations to CA3- CA1 oscillatory activity in a novel object recognition task

Authors: *K. PAREKH[1], L. M. CROWN[1], R. E. FEATHERSTONE[1], M. W. SALTER[2], C.-G. HAHN[3], S. J. SIEGEL[1];
[1]Dept. of Psychiatry and Behavioral Sci., USC, Los Angeles, CA; [2]Dept. of Physiol., Univ. of

Toronto, Toronto, ON, Canada; [3]Dept. of Psychiatry & Neurosci., Thomas Jefferson Univ., Philadelphia, PA

Abstract: Memory impairments are common to a variety of neuropsychiatric disorders including schizophrenia. A leading hypothesis is that such impairments arise from attenuated signaling activity of N-Methyl-D-aspartate (NMDA) receptors. Indeed, studies have found that various molecular alterations associated with schizophrenia (e.g. Dysbindin, DISC-1, erbB4) act to modulate NMDA receptor function. Intriguingly, these risk factors likely alter NMDA receptors through a common mechanism, the regulation of Sarcoma tyrosine kinase (Src). Src is highly expressed within the hippocampus, specifically in the CA3 and dentate gyrus region, further supporting a role for Src in schizophrenia-related memory deficits. Correspondingly, previous studies by our group have found that Src-deficient mice show deficit recall, but not learning in trace fear conditioning. We have also observed alterations to evoked gamma oscillations in these animals. A growing body of literature supports a crucial role for CA3 gamma oscillatory activity in facilitating the recall of episodic memory. Therefore, we hypothesized that Src-deficient mice (Src heterozygous) would show alterations to gamma oscillatory activity, particularly CA3-associated low gamma. In this study we recorded activity from the dorsal CA1, ventral CA1, CA3 and the medial prefrontal cortex (mPFC) of Src heterozygous mice (n = 8; 4 male, 4 female) and wild-type littermates (n= 9, 4 male, 5 female) as they performed a novel object recognition (NOR) task. While we observed no differences in the power of CA3 or CA1 theta, low gamma, or high gamma, we did find that the frequency of CA1 low gamma was reduced in Src animals compared to wild-type controls, while no differences were observed to in the peak frequency of theta or high gamma. Examining CA3 to CA1 coherence, we found increased low gamma coherence for Src animals. These results suggest that Src deficiency may alter the function of CA3 and its communication with CA1, perhaps underlying associated deficits in memory recall.

Disclosures: K. Parekh: None. **L.M. Crown:** None. **R.E. Featherstone:** None. **M.W. Salter:** None. **C. Hahn:** None. **S.J. Siegel:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.03

Topic: H.13. Schizophrenia

Support: Iniciativa Científica Milenio (ICM-P09-015F)
Beca Fundación Guillermo Puelma

Title: Gamma Power alterations in Schizophrenia: A developing biomarker and promising window to schizophrenia pathophysiology

Authors: *E. TABILO[1], R. MAYOL-TRONCOSO[5], R. VERDUGO[1], P. A. GASPAR[6,5], P. E. MALDONADO[1,7], J. PARRINI[8], G. ORELLANA[2], A. RUIZ[3], J. EGAÑA[4,7], C. DEVIA[1,7]; [1]Dept. de Neurociencia, Facultad de Medicina, [2]Dept. de Psiquiatría, Facultad de Medicina, [3]Hosp. Clínico Univ. de Chile, Dept. de Psiquiatría y Salud Mental, [4]Dept. de Anestesiología y Medicina Perioperativa, Univ. de Chile, Santiago, Chile; [5]Clínica Psiquiátrica Universitaria, Dept. de Psiquiatría, Facultad de Medicina, Univ. de Chile, Santiago, Chile; [6]Dept. de Psiquiatría, Facultad de Medicina, Univ. De Chile, Santiago, Chile; [7]Biomed. Neurosci. Inst., Santiago, Chile; [8]Dept. of Liaison Psychiatry, Hosp. Doctor Luis Tisné Brousse, Santiago, Chile

Abstract: A current interest in translational neuroscience is the development of psychiatric biomarkers. In the case of schizophrenia, several electroencephalography-based (EEG) biomarkers have been proposed, such as evoked potentials, entropy, or oscillatory power. Gamma band oscillations are of special relevance since the glutamatergic and gabaergic pathophysiological models of schizophrenia predict disruptions in gamma activity, as evidenced by MEG studies from visual perception of Mooney faces. However, it remains unknown if similar alterations appear in more ecological conditions, such as free exploration of natural images. This study investigates gamma-band power on EEG recordings while participants freely explored natural scenes. We analyzed 11 medicated patients with schizophrenia diagnosis (SZ), and 9 age-paired healthy controls (HC). They explored Natural Images (NI) and control images (Pink noise (PN) and Gray images (GI), among others). We extracted EEG power using a time-frequency representation. Then we compared power from the two groups with a cluster-test. This last step was performed for each image category and for each frequency band. At each category the clusters with the biggest differences were used to train a decision tree (J48). The algorithms trained with gamma-clusters obtained an Accuracy of 95% for control images and 94.9% for NI images, validated by 10-fold Cross-Validation on this dataset, and beta-clusters obtained similar accuracies. Interestingly, the direction of the gamma-band alterations was opposite between distinct conditions: NI elicited gamma power deficits in patients compared to controls, while PN and GI conditions elicited greater gamma power in patients than in controls. Although both gabaergic and glutamatergic models of schizophrenia correctly predicted the gamma power deficit of patients on the NI condition, the power augment relative to controls in the GI condition only has a precedent in the glutamatergic model research, which reported gamma power alterations in both directions in conditions of NMDAR antagonists pharmacological administration, in both non-human and human animals. In conclusion, the processing of natural images elicits cortical activity with diminished gamma band in patients but augmented for images without semantic content (PN and GI), which resembles an effect found in the glutamatergic model of schizophrenia; and the activity elicited by each image category allows, by itself, the detection of patients from controls based on their EEG signal.

Disclosures: E. Tabilo: None. **R. Mayol-Troncoso:** None. **R. Verdugo:** None. **P.A. Gaspar:** None. **P.E. Maldonado:** None. **J. Parrini:** None. **G. Orellana:** None. **A. Ruiz:** None. **J. Egaña:** None. **C. Devia:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.04

Topic: H.13. Schizophrenia

Support: NIH Grant 5K00MH125434-05
NIH Grant R01NS110776-04

Title: Astroglial calcium signaling deficits perturb synaptic function in a human glial chimeric mouse model of schizophrenia

Authors: *J. CANO[1], S. A. GOLDMAN[2,3];
[1]Univ. of Rochester Med. Ctr., Rochester, NY; [2]Dept of Neurol., Univ. Rochester Med. Ctr., Rochester, NY; [3]Fac. of Hlth. and Med. Sci., Univ. of Copenhagen, Copenhagen, Denmark

Abstract: Schizophrenia (SCZ) is one of the most disabling and poorly understood neuropsychiatric disorders, in large part due to the polygenic and heterogeneous nature of its pathogenesis. Converging studies highlight the influence of astrocyte pathology on the synaptic pathology of schizophrenia. Previously, we identified impaired astroglial differentiation with attendant phenotypic abnormalities in mice engrafted with childhood-onset SCZ human iPSC-derived glial progenitor cells (hGPCs). Subsequent *in vitro* and *in vivo* analyses revealed the impaired astroglial differentiation and disrupted potassium buffering of SCZ astroglia, which could be rescued in part by targeting the dysregulated expression of the REST transcriptional repressor. Nonetheless, the specific influence of SCZ-astroglia on neuronal and synaptic function, and that of dysregulated REST expression on astroglial calcium signaling, remain enigmatic. To address these issues, we first used CRISPR-Cas9 to generate stable knock-in control- and SCZ-derived hiPSCs expressing GCaMP7, a fluorescent calcium sensor. GCaMP7-expressing iPSCs were then differentiated into GPCs, which were CD140 FACS-sorted, validated as expressing the canonical hGPC markers PDFGRα and OLIG2, and transplanted into neonatal hypomyelinated and immunodeficient *shiverer* x *rag2*$^{-/-}$ mice. Electrophysiological assessment 19 weeks later revealed that those cortical and striatal neurons apposed to transplanted GCaMP7-expressing SCZ glia were hyperexcitable and manifested deficient intercellular astroglial calcium signaling relative to controls. With that background, we then evaluated the impact of regulating glial REST expression on astrocytic calcium signaling *in vitro*. GCaMP7-expressing GPCs were differentiated into astrocytes, then FACS-sorted on CD44 and their astrocytic phenotype validated by the expression of AQP4 and GFAP. In SCZ astrocytes, lentiviral REST knockdown (REST-KD) rescued otherwise deficient ATP-elicited calcium events *in vitro* (SCZ REST-KD vs SCZ: $p<0.01$; SCZ REST-KD vs CTRL: p=NS). Along the same line, control astrocytes overexpressing REST mimicked the deficient calcium signaling of SCZ astrocytes (CTRL-REST vs CTRL: $p<0.05$; CTRL-REST vs SCZ: p=NS). Taken together, these findings highlight the impact of astroglial impairment on the synaptic pathology of schizophrenia and provide tangible targets for its potential glial-based treatment.

Disclosures: J. Cano: None. **S.A. Goldman:** A. Employment/Salary (full or part-time):; Sana Biotechnology. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Sana Biotechnology.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.05

Topic: H.13. Schizophrenia

Support: NIMH Grant R61/R33MH112793

Title: Frontal P300 as a marker of effective neurofeedback training targeting working memory in patients with schizophrenia

Authors: *I.-W. SHU, Y. LIN, S.-H. HSU, B. CASTILLO, K. KASRAEIAN, P. LINK, J. PINEDA, E. GRANHOLM, F. SINGH;
Univ. of California San Diego, La Jolla, CA

Abstract: Complex tasks requiring cortical processing of dynamic stimuli are associated with specific electroencephalographic (EEG) signatures, including the P300 event-related potential (ERP). P300 generally peaks at midline electrodes several hundred milliseconds after novel or salient stimuli, and is modulated by intensity of stimuli (e.g., greater/lesser novelty or salience), as well as the participant's level of effort or ability. While patients with schizophrenia (SCZ) generally exhibit decreased P300 amplitudes, under specific conditions, patients with SCZ may exhibit greater P300 amplitudes, e.g., when exerting greater effort, or exhibiting reduced habituation, than controls. As such, P300 may serve as a marker of cognitive ability or effort for treatment studies targeting cognitive impairment in patients with SCZ. To explore this hypothesis, we assessed frontal P300 amplitudes during a working memory (WM) task (N-back), in participants with SCZ (n = 31), pre- and post-EEG neurofeedback (NFB) training designed to improve WM by targeting dorsal-lateral prefrontal gamma activity. In support of this approach, in addition to above features, P300 arises from multiple cortical, including frontal, sources. Initial clinical and neurophysiologic results were previously reported. In this follow-up analysis, we present new results modeling the effects of NFB training efficacy on changes in frontal P300 amplitude and WM performance during training, and training-related changes in frontal theta, alpha and gamma power. In a minimal structural equation model, greater NFB efficacy significantly predicted training-related P300 amplitude increases ($beta$ = 0.285, p = 0.05); furthermore, greater training-related P300 amplitude significantly predicted greater training-related WM performance ($beta$ = 0.317, p = 0.035). NFB training was also associated with increased frontal theta, alpha and gamma power during N-back (slope = 0.006, 0.01, 0.04; p < 0.001, 0.008, 0.004; respectively). Including training-related frontal theta, alpha and gamma changes in the above model improved the strength/significance of NFB efficacy's effect on frontal neural activity ($beta$ = 0.340, p = 0.035), and strength of training-related frontal neural activity on training-related WM performance ($beta$ = 0.327, p = 0.045). These promising early results suggest that, in patients with SCZ, frontal P300 may serve as a marker of effective NFB training targeting frontal gamma activity and WM. We aim to formally test the specificity of such effects in larger, randomized, controlled clinical trials.

Disclosures: **I. Shu:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); BioSignal Solutions LLC. **Y. Lin:** None. **S. Hsu:** None. **B. Castillo:** None. **K. Kasraeian:** None. **P. Link:** None. **J. Pineda:** None. **E. Granholm:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); BioSignal Solutions LLC. **F. Singh:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); BioSignal Solutions LLC.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.06

Topic: H.13. Schizophrenia

Support: KAKENHI Grant JP20H05064

Title: A neural network model of aberrant visual salience for approaching the heterogeneous pathophysiology of schizophrenia

Authors: *Y. FUJITA, T. MURAI, J. MIYATA;
Dept. of Psychiatry, Kyoto Univ., Kyoto, Japan

Abstract: Although various neurobiological abnormalities have been reported as possible causes of schizophrenia, the mechanism how they induce symptoms such as delusion and hallucination is still unknown. Attempts to fill the large gap between biology and symptoms include computational modeling based on the aberrant salience hypothesis, that salience attributed to contextually irrelevant stimuli gives rise to psychotic experiences. This hypothesis deals with motivational salience derived from dopamine-dependent learning. On the other hand, experimental findings regarding schizophrenia also indicate alterations in other kinds of salience, namely visual and auditory salience, and in other kinds of neurobiological features including excitatory-inhibitory (E-I) balance, structural connectivity, and cortical signal-to-noise ratio. Multiple mechanisms should underlie schizophrenia that can be considered as a heterogeneous syndrome. In this study, we use computational modeling to investigate how visual salience is altered by neurobiological disturbances including E-I imbalance. We implemented a saliency map model with neural networks so that the biological features and disturbances can be reflected in the model. It consists of feature maps receiving input features extracted from visual stimulus, conspicuity maps integrating feature representations for each stimulus dimension, and a saliency map as an output. Each map is implemented as two-dimensional arrays of neural populations that have local excitatory connections and distant inhibitory connections. These lateral connections cause competition between features within a map and thereby make the representations of salient visual stimuli stronger than those of non-salient stimuli. We introduced E-I imbalance into the model by changing the parameters of the lateral connections. Elevation of E-I balance made the salience of non-salient stimuli closer to that of salient stimuli, while reduction of E-I balance

resulted in salience for non-salient stimuli far smaller than that for salient stimuli. Such alteration of visual salience can be regarded as changing sensitivity, which is actually implied by experiences reported by schizophrenia patients. It is in contrast to the way contextually irrelevant assignment of salience generates abnormal motivational salience as described in the aberrant salience hypothesis. Our results suggest that different biological abnormalities can cause different patterns of aberrant salience. Since visual salience influences eye movements, our model may lead to novel eye-tracking data analysis to infer specific biological abnormality underlying each patient.

Disclosures: **Y. Fujita:** None. **T. Murai:** None. **J. Miyata:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.07

Topic: H.13. Schizophrenia

Support: DANA
K23MH108711
R01MH123639
MH121790

Title: Population receptive field mapping of parietal retinotopic areas in schizophrenia

Authors: ***N. C. FOLEY**[1,2], J. K. LEE[2], N. M. MACILVANE[2], A. MURATI[2], J. KRAVITZ[3], M. RAHMATI[4], C. E. CURTIS[5], G. H. PATEL[6];
[1]Psychiatry, Columbia Univ., New York, NY; [2]Psychiatry, New York State Psychiatric Inst., New York, NY; [3]Psychology, [4]New York Univ., New York, NY; [5]Psych & CNS, NYU, New York, NY; [6]Psychiatry, Columbia University/NYSPI, New York, NY

Abstract: Deficits in social cognition are a major source of disability in neuropsychiatric disorders, especially schizophrenia (SzP). One major domain in social cognition is accurate perception of social cues, such as facial expressions of emotion. Detecting visual social cues requires an individual to make saccadic eye-movements to scan a scene for salient or behaviorally relevant information. Crucially, it has been shown that SzP make about 20% fewer saccades to low speed moving facial expressions compared to healthy controls (HC). This failure to orient towards changes in expression may either be due to direct perceptual deficits, or a deficit in mentalization indicating that these changes in expression are behaviorally relevant. Planned eye movements are controlled by the dorsal attention network, which includes parietal cortex.
Here, we use fMRI to scan patients and healthy controls during a rapid serial visual presentation (RSVP) task designed to robustly activate visual and attentionally related cortices over a large area of the visual field (14.3x14.3 deg). This consisted of ‘bars’ of 6 RSVP streams of pictures of

objects that refreshed every ~300-600ms and traversed the visual field from left to right (LR), top to bottom (TB) and (RL, BT) over 8 locations in each sweep. The subject was instructed to fixate while trying to detect an instructed object in each run. Stimulus duration of the objects was varied to maintain detection performance between 50-80%. We used population receptive field mapping (pRF) to define the potential human homologue of macaque lateral interparietal cortex (LIP) in both left and right hemispheres, which is implicated in the planning of eye-movements. No group difference was found in either stimulus duration ($p = 0.81$) or detection accuracy ($p = 0.32$). Preliminary analysis indicates these ROI maps were highly heterogenous in location, with 57% sharing overlapping locations in at least 2 subjects the right hemisphere and 44% in the left hemisphere. The density of myelin decreases as a function of eccentricity in both hemispheres of HCs and right SzP (preliminary $n = 4$ and 7, data collection ongoing; all $p < 10^{-4)}$, while trending in left SzP ($p = 0.054$).
Overall, the current results demonstrate the utility of individually defined areas over group ROIs. Furthermore, the preliminary results suggest that the structure-function relationships in human homologue of LIP in schizophrenia are relatively intact.

Disclosures: **N.C. Foley:** None. **J.K. Lee:** None. **N.M. Macilvane:** None. **A. Murati:** None. **J. Kravitz:** None. **M. Rahmati:** None. **C.E. Curtis:** None. **G.H. Patel:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.08

Topic: H.13. Schizophrenia

Support: 1R01 MH111177
Lycaki-Young Funds (State of Michigan)
Mark Cohen Neuroscience Endowment
Ethel and James Flinn Foundation
UROP WSU

Title: The effective connectivity of network dynamics during memory encoding and retrieval: Extensions to Schizophrenia

Authors: *K. G. NANAAWARE[1], A. Z. CHOWDURY[1], S. BAAJOUR[1], P. THOMAS[1], U. RAJAN[1], D. KHATIB[1], L. HADDAD[1], A. AMIRSADRI[1], J. A. STANLEY[2], V. A. DIWADKAR[1];
[1]Psychiatry and Behavioral Neurosciences, Wayne State Univ. Sch. of Med., Detroit, MI; [2]Psych & Behav Neurosci, Wayne State Univ. Translational Neurosci. Program, Detroit, MI

Abstract: Schizophrenia is characterized by network dysfunction, which underpins behavioral deficits in memory and associative learning. Memory and associative learning are processes dependent upon frontal-hippocampal function and underpinned by dynamic changes in brain

network activity/connectivity. These changes may be particularly acute in paradigms that induce non-linear learning: early linear increases in learning, followed by asymptotic performance. How are these functional network processes maintained or disrupted in schizophrenia? We evaluated two questions in our modeling: 1) are descending pathways from the dorsal anterior cingulate (dACC) and dorsolateral prefrontal (dlPFC) cortices contextually modulated by task conditions and time, and 2) do estimates of contextual modulation differ between schizophrenia patients (SCZ) and healthy controls (HC). Participants (n=90; age: 18-50) provided informed consent and were compensated for participation. SCZ (n=52) were stabilized on a regimen of atypical antipsychotics. HC (n=38) were free of psychiatric/neurological conditions. Data (3.0T Siemens Verio) were processed in SPM12. Network dynamics (negatively accelerated learning) induced using an object-location task and modeled using Dynamic Causal Modeling (DCM). A model space was constructed on basis of co-activation of task-relevant regions: primary visual cortex (V1), superior parietal (SPG) and inferior temporal (ITG) gyri, hippocampus (HPC), dACC, and dlPFC. Bayesian Model Selection (BMS) used to determine likely model (and presence of contextual modulation of descending pathways by task and time). BMS identified the model sans descending connections from the frontal lobe to unimodal regions as more likely (exceedance probabilities >0.8). Subsequent analyses focused on main effects (group or time) and interactions on parameter estimates of contextual modulation. During Encoding, reduced contextual modulation of HPC->dACC (p=0.04) and dlPFC->dACC (p=0.02) pathways in SCZ and main effects of time (ps <0.05) on all pathways (except V1->ITG) were observed. During Retrieval, reduced contextual modulation (p=0.009) of dlPFC->dACC pathway in SCZ and main effects of time (ps <0.05) on HPC->dlPFC and HPC->dACC pathways were observed. A single time*group interaction (p=0.009) was observed on the ITG->HPC pathway during Retrieval, where contextual modulation *decreased* in HC but *increased* in SCZ.

Disclosures: K.G. Nanaaware: None. **A.Z. Chowdury:** None. **S. Baajour:** None. **P. Thomas:** None. **U. Rajan:** None. **D. Khatib:** None. **L. Haddad:** None. **A. Amirsadri:** None. **J.A. Stanley:** None. **V.A. Diwadkar:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.09

Topic: H.13. Schizophrenia

Support: SNSF grant SYNAPSY-51NF40-185897
ERC grant SyG-856439-CLAUSTROFUNCT

Title: Relationship between altered medial prefrontal cortex circuit dynamics and impaired cognitive flexibility in an animal model of schizophrenia

Authors: C. HUBER, M. MOULINIER, I. RODRIGUEZ, ***A. CARLETON**;
Univ. of Geneva, Geneva, Switzerland

Abstract: Schizophrenia is a chronic mental disease with a prevalence of ~1% worldwide. This psychiatric disorder is characterized by positive and negative symptoms as well as cognitive deficits. Though current medications usually alleviate positive symptoms, they are much less efficient in treating negative and cognitive ones. Although the exact causes of schizophrenia are unknown, impaired cortical communication and processing seem to be critical for the expression of the disease. Establishing a causal link between circuit dysfunction and particular behavioral traits relevant to schizophrenia may shed new light on the mechanisms underlying the pathology. Here we used a hemizygote knockout mouse of *Nr4a2* (also called *Nurr1*), a gene whose mutation represents a genetic risk factor for developing schizophrenia in humans. Though NR4A2 is a nuclear receptor contributing to the specification and maintenance of the dopaminergic phenotype in the ventral tegmental area and the substantia nigra, it is also expressed in glutamatergic neurons of various brain areas such as the hippocampus, cortex and claustrum. We monitored the physiological responses of a genetically identified neuronal population of the medial prefrontal cortex and compared the behavioral performances of the mutant to their wild-type littermates. We observed that basic associative learning and rule generalization were not affected in the mutant mice. In contrast, knockout mice exhibited a strong impairment in cognitive flexibility, a hallmark of cognitive alteration previously observed in patients. Using calcium imaging, we observed that neurons were less active and were more desynchronized in the mutant than in the WT network. We demonstrate that alteration of *Nr4a2* expression specifically in a population of glutamatergic neurons and selectively during adulthood in WT mice was sufficient to phenocopy the neurophysiological and behavioral alterations observed in the KO mouse. Importantly, the opposite manipulations during adulthood improved neurophysiological and behavioral impairment observed in the mutant line. Taken together, our data support a causal relationship between glutamatergic neuron dysfunction and cognitive deficits observed in some psychiatric disorders.

Disclosures: C. huber: None. **M. moulinier:** None. **I. Rodriguez:** None. **A. Carleton:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.10

Topic: H.13. Schizophrenia

Title: The selective GPR139 agonist TAK-041 reverses anhedonia and social interaction deficits in rodent models related to negative symptoms in schizophrenia

Authors: *H. H. SCHIFFER, J. ATIENZA, H. REICHARD, V. MULLIGAN, J. CILIA, H. MONENSCHEIN, D. COLLIA, W. J. RAY, G. KILPATRICK, N. BRICE, M. CARLTON, S. HITCHCOCK, G. CORBETT, R. HODGSON;
Takeda Develop. Ctr. Americas, Inc., San Diego, CA

Abstract: Negative symptoms in schizophrenia (SCZ) are poorly treated and interfere with the function of patients. Studies focusing on structural and functional imaging and non-invasive electrophysiology implicate perturbations of the frontocortico-temporal circuits and disruption of cortico-striatal loops to negative symptoms in SCZ. GPR139 is an orphan GPCR that is specifically expressed in the CNS and enriched in the habenula, a brain structure involved in reward and motivation. Structural and functional alterations of the habenula have been found in SCZ patients, who show deficits in feedback processing and lack habenula activation in response to negative outcomes. Rodent experiments demonstrate the involvement of direct projection from the cortex to the habenula controlling social behavior. Alterations in habenula activity have been correlated with depression, and normalization of aberrant habenula activity has been proposed as therapeutic strategy to reverse anhedonia. Building on data previously reported, we further explored modulation of GPR139 receptors and habenula circuitry *in vivo* as a novel mechanism to treat negative symptoms in SCZ. The selective GPR139 agonist, TAK-041, increased cFOS expression in the habenula in wild type mice, but not in GPR139 knock out mice. No desensitization of cFOS in the habenula was observed after chronic dosing of TAK-041. TAK-041 reduced amphetamine- and nicotine-induced dopamine release in the NAc in rats. In the rat unpredictable chronic mild stress (uCMS) model, acute and chronic treatment with TAK-041 reversed the anhedonic and anxiolytic effects, as measured by the forced swim test and the novelty-suppressed feeding test, respectively. Additionally, uCMS-exposed rats developed a disrupted circadian regulation of corticosterone secretion, which was also normalized by TAK-041 treatment. Furthermore, TAK-041 reversed the uCMS-induced atrophy in the basal dendrites of pyramidal neurons in the hippocampus and the uCMS-induced hypertrophy of medium-spiny neurons in the NAc. These results suggest a potential benefit of TAK-041 in the treatment of anhedonia and possibly depression. TAK-041 also reversed social interaction (SI) deficits in the maternal immune activation poly-I:C model of SCZ, the subchronic PCP-SI model and in Balb/C and BTBR mice. The GPR139 agonist TAK-041 is proposed as a modulator of habenula circuitry to treat negative symptoms in SCZ based on efficacy in reversing anhedonia and social interaction deficits in multiple rodent models related to negative symptoms in SCZ.

Disclosures: **H.H. Schiffer:** A. Employment/Salary (full or part-time):; Takeda Pharmaceuticals. **J. Atienza:** A. Employment/Salary (full or part-time):; Takeda Pharmaceuticals. **H. Reichard:** A. Employment/Salary (full or part-time):; Takeda Pharmaceuticals. **V. Mulligan:** None. **J. Cilia:** None. **H. Monenschein:** A. Employment/Salary (full or part-time):; Takeda Pharmaceuticals. **D. Collia:** None. **W.J. Ray:** None. **G. Kilpatrick:** None. **N. Brice:** None. **M. Carlton:** None. **S. Hitchcock:** A. Employment/Salary (full or part-time):; Takeda Pharmaceuticals. **G. Corbett:** None. **R. Hodgson:** A. Employment/Salary (full or part-time):; Takeda Pharmaceuticals.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.11

Topic: H.13. Schizophrenia

Support: 5P50MH103222

Title: Enduring adverse effects of adolescent cannabis abuse involve kynurenic acid: relevance for the etiology and treatment of cognitive dysfunctions.

Authors: *S. BEGGIATO[1], L. FERRARO[2], R. SCHWARCZ[3];
[1]Univ. of Ferrara, Univ. of Chieti, Ferrara, Italy; [2]Univ. of Ferrara, Univ. of Ferrara, Ferrara, Italy; [3]Maryland Psychiatric Res. Ctr., Baltimore, MD

Abstract: Cannabis abuse during adolescence is a risk factor for cognitive impairments in psychiatric disorders later in life (Renard et al., 2016). Specifically, early exposure to Δ9-tetrahydrocannabinol (THC; i.e. the main psychotropic component of cannabis) causes enduring cognitive deficits, which critically involve impaired glutamatergic function in the prefrontal cortex (PFC; Zamberletti et al., 2014). Remarkably, these adverse effects are qualitatively very similar to those caused in developing rodents by experimental increases in brain kynurenic acid (KYNA), a neuroactive metabolite of tryptophan degradation (Pocivavsek et al., 2014).To date, the possible causal relationship between cannabinoids, KYNA and cognition has not been investigated in adolescence. We therefore examined the effect of chronic adolescent THC exposure on KYNA levels in the rat brain. Male Wistar rats were chronically treated with vehicle or ascending intraperitoneal (i.p.) doses of THC starting on postnatal day (PND) 35 until PND 45 (Zamberletti et al., 2014). In adulthood (PND 75), cognitive assessment (Y-maze) and extracellular KYNA/glutamate levels were measured in the PFC by *in vivo* microdialysis, before and after a challenge with L-KYN (5 mg/kg i.p., the biological precursor of KYNA). By using the selective, brain-penetrable KAT II inhibitor PF-04859989 (Kozak et al. 2014), we then examined whether blockade of KYNA neosynthesis prevents the cognitive impairment. Compared to vehicle-treated controls, extracellular basal KYNA levels were higher in the PFC of adult rats chronically exposed to THC in adolescence ($p<0.01$). No changes were observed in extracellular glutamate levels. Following a challenge with L-KYN, extracellular KYNA levels similarly increased in both groups (i.e. vehicle- and THC-treated; $p<0.001$ and $p<0.01$ respectively). Chronic adolescent THC exposure negatively affected short-term memory (reduced spontaneous alternation), in adult animals ($p<0.001$), while PF-04859989 partially restored the impairment ($p<0.05$). We propose that these alterations in PFC KYNA signalling might be involved in the cognitive dysfunction induced by the exposure to THC during the adolescence. In the translational realm, these experiments raise the prospect of prevention of KYNA neosynthesis as a promising novel approach to combat some of the detrimental long-term effects of adolescence cannabis use.

Disclosures: S. Beggiato: None. **L. Ferraro:** None. **R. Schwarcz:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.12

Topic: H.13. Schizophrenia

Title: Blockade of GABA Transporter 1 (GAT1) improves the spatial learning tasks in rats neonatally treated with MK-801

Authors: *C. LOPEZ-RUBALCAVA[1], L. A. MÁRQUEZ[2], E. J. GALVAN[3];
[1]CINVESTAV-IPN, Mexico DF, Mexico; [2]CINVESTAV, Mexico City, Mexico; [3]CINVESTAV SUR, Mexico City, Mexico

Abstract: NMDA receptor hypofunction is a fundamental and convergent mechanism for the pathophysiology of schizophrenia, and transient blockade of the NMDA receptor during early development induces schizophrenia-like behaviors in rats. Dysfunction of GABAergic transmission has been reported in the hippocampus and prefrontal cortex both in patients with schizophrenia and in animal models, both structures are fundamentally involved in multiple memory types. In this study, we investigated if the increase of GABA levels by inhibiting GABA Transporter 1 (GAT1) could alleviate the deficits of hippocampus-dependent tasks in juvenile male rats neonatally treated with MK-801. Male pup rats from postnatal days (P) 7 to 11 were daily treated with NMDA receptor antagonist MK-801 (0.2 mg/kg) and evaluated in spatial pattern separation (P28-37) and Barnes maze task (P30-P35). We found that MK-801-treated animals showed reduced performance in the Barnes maze, indicating an impairment in spatial memory. In addition, in comparison with the control group, MK-801-treated animals were unable to discriminate minimal changes in the spatial position of identical objects, demonstrating an impairment in spatial pattern separation. Next, we analyzed if NNC-711, a selective inhibitor of GAT1, could alleviate the spatial memory deficits. NNC-711 treatment at 0.5 mg/kg (but not at 1.5 mg/kg) improved the performance of MK-801-treated animals in the Barnes maze. Our results suggest that GAT1 inhibition during the early or prodromal phase of schizophrenia could be an effective treatment for alleviating hippocampus-dependent memory deficits.

Disclosures: C. Lopez-Rubalcava: None. **L.A. Márquez:** None. **E.J. Galvan:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.13

Topic: H.13. Schizophrenia

Support: NSERC Discovery Grant RGPIN-2017-05401

Title: The preventive effects of ultra-micronized palmitoylethanolamide (PEA) in a preclinical model of schizophrenia

Authors: *C. CLARKE, S. DONOVAN, S. HASAN, F. R. BAMBICO;
Mem. Univ. of Newfoundland, St. John's, NL, Canada

Abstract: Schizophrenia (SCZ) is a complex neurodevelopmental disorder with early adult onset and typically follows multiple pathogenic factors (multi-hit), including neuroinflammatory events. Current antipsychotics prove to be insufficient at treating the full range of symptoms, posing the need for novel therapies. To date, the maternal immune activation (MIA) model has both high face and predictive validity, making it the most translatable model for the development of novel therapies. The endocannabinoid system has long been theorized as a target for novel interventions, largely due to its role in neurodevelopment and regulating inflammatory processes, however, targeting the main CB1R has proven to be ineffective. Brain-wide CB2Rs are a novel target for SCZ treatment and prevention as they are known to regulate inflammatory processes that contribute to its pathology. Palmitoylethanolamide (PEA) is an endogenous fatty acid amide that binds to PPAR-alpha receptors to indirectly activate CB2R, triggering an anti-inflammatory cascade, thus regulating neuroinflammation. Hallmarks of SCZ include a decrease in CB2R and PPAR-alpha function, leading to rampant neuroinflammation; therefore, we hypothesize that administering PEA during preadolescence will prevent neuroinflammation induced during the pubertal period and subsequently prevent SCZ onset. Prior to MIA, we used an acute MK-801 mouse model to test whether orally administered PEA pretreatment in adult C57BL/6 mice would prevent MK-801-induced deficits in the translatable SCZ biomarker Mismatch Negativity (MMN), an auditory EEG phenotype evoked by an oddball stimulus seen in both humans and rodents exhibiting SCZ-related symptoms. PEA pretreatment prevented MK-801-induced MMN deficits in both male and female adult mice ($p = 0.02$). Next, we used the MIA mouse model to explore whether PEA administration during preadolescence would prevent the SCZ phenotype from appearing in adult offspring from dams exposed to the immunostimulant LPS on gestation day 14.5. LPS mice presented with social deficits ($p = 0.02$) and novel object recognition impairments ($p = 0.03$), meanwhile, PEA prevented these deficits, restoring the behaviors to control levels ($p = 0.01$). Female LPS-PEA mice displayed control novel object recognition behavior ($p = 0.02$); however, PEA did not prevent social deficits in LPS females ($p > 0.05$). EEG MMN data is currently being analyzed to explore whether PEA also successfully prevented characteristic SCZ MMN deficits in adult MIA offspring. This study has thus far provided promising data indicating SCZ typical behaviors in MK-801 and MIA mouse models can be prevented with oral PEA pretreatment.

Disclosures: **C. Clarke:** None. **S. Donovan:** None. **S. Hasan:** None. **F.R. Bambico:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.14

Topic: H.13. Schizophrenia

Title: Increasing the Excitatory Drive Rescues Excitatory/Inhibitory Imbalance and Mismatch Negativity Deficit Caused by Parvalbumin Specific GluA1 Deletion

Authors: H.-J. CHEN-ENGERER[1], S. JAEGER[1], B. RIMMA[1], R. SPRENGEL[2], B. HENGERER[1], H. ROSENBROCK[1], V. MACK[3], N. SCHUELERT[1];
[1]Central Nervous Syst. Dis. Research, Boehringer Ingelheim Pharma GmbH & Co. KG, Biberach/Riss, Germany; [2]Max Planck Inst. for Med. Res. at the Inst. for Anat. and Cell Biol. at Heidelberg Univ., Heidelberg, Germany; [3]CardioMetabolic Dis. Research, Boehringer Ingelheim Pharma GmbH & Co. KG, Biberach/Riss, Germany

Abstract: Excitation-inhibition (E/I) imbalance in the prefrontal cortex has been considered as a critical pathophysiological mechanism for cognitive dysfunction, a core symptom in schizophrenia. However, the cortical network pathophysiology induced by E/I imbalance is not well characterized, and an effective therapeutic strategy is still lacking. In this study, we simulated imbalanced cortical network by using mice with parvalbumin neuron (PV) specific knockout of GluA1 (AMPA receptor subunit 1) (Gria1-PV KO) as an experimental model. Applying high-content confocal imaging and electrophysiological recordings in the medial prefrontal cortical (mPFC), we found structural and functional alterations in the local network of Gria1-PV KO mice. In addition, Gria1-PV KO animals exhibited abnormal theta oscillation and deficits in mismatch negativity (MMN), which is consistent with clinical findings in cognitively impaired patients. Remarkably, we demonstrated that application of the glycine transporter 1 (GlyT1) inhibitor, Bitopertin, ameliorates E/I imbalance, hyperexcitability, and sensory processing malfunction in Gria1-PV KO mice. Our results suggest that PV-specific deletion of GluA1 might be an experimental approach for back translating the E/I imbalance observed in schizophrenic patients. Our work offers a systematic workflow to understand the effect of GlyT1 inhibition in restoring cortical network activity from single cells to local brain circuitry. Furthermore, this study highlights that selectively boosting NMDA receptor-mediated excitatory drive to enhance the network inhibitory transmission from interneurons to pyramidal neurons is a potential therapeutic strategy for restoring E/I imbalance-associated cognitive-related abnormality.

Disclosures: H. Chen-Engerer: A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co. KG. **S. Jaeger:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co. KG. **B. Rimma:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co. KG. **R. Sprengel:** None. **B. Hengerer:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co. KG. **H. Rosenbrock:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co. KG. **V. Mack:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co. KG. **N. Schuelert:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co. KG.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.15

Topic: H.13. Schizophrenia

Support: NIH P50 MH103222

Title: Longitudinal assessment of developmental changes in the structure and function of white matter tracts in adolescent minipigs

Authors: P. KOCHUNOV, A. T. SUMMERFELT, P. L. BROWN, M. C. TERZI, K. YACHERA, K. V. SATHYASAIKUMAR, X. DU, L. E. HONG, R. SCHWARCZ, ***P. D. SHEPARD**;
Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: Most translational animal models used to study structural and functional alterations in the organization of cerebral white matter in schizophrenia have serious limitations. Rodents have a lissencephalic cortex and a grey:white matter ratio that differs from humans; ferrets undergo gyrification after birth; and use of nonhuman primates is constrained by ethical and economic considerations. By contrast, Sinclair miniature pigs are born with a fully gyrencephalic brain and have a grey:white matter volume ratio similar to humans. Pigs mature over a 4-6 month interval which is more extensive than rodents but compressed relative to primates, making them well suited for longitudinal studies of adolescent brain development. In the present series of experiments, we used quantitative diffusion weighted imaging (DWI) and evoked potential (EP) recording to assess the maturation of cerebral white matter in adolescent Sinclair miniature pigs. Animals underwent four EEG/MRI sessions on PND 98, 138, 166 and 194. Imaging was conducted in a Siemens 3 Tesla Prisma scanner and a 15-channel knee coil. A higher resolution anatomical (0.5mm isotropic) DWI protocol (1.25mm isotropic), proportionally scaled to the resolution used in humans, consisted of 15 b-shells (b = 0-3500 s/mm2) with 32-directions/shell. Data were analyzed using diffusion kurtosis and bi-exponential modeling that provide fractional anisotropy radial kurtosis, kurtosis anisotropy, axial kurtosis, tortuosity, and permeability-diffusivity index of cerebral white matter. EEG recordings were acquired with a Neuroscan Synamp2 (5 kHz sampling rate; bandpass of 0.1-200 Hz) immediately before imaging from subdermal platinum-iridium needles. Following a 5-minute resting EEG recording, monocular visual EPs were elicited by light flashes (300 stimuli @ 1 Hz, 10 - 500 ms duration) generated by a Lifelines photic stimulator. Auditory EPs were recorded in duration mismatch (80% standard, 60-ms 1000 Hz tones and 20% deviant 100 ms, tones presented in pseudorandom order) and auditory steady-state (75 click trains presented at 2.5 Hz and 40 Hz) procedures. All stimuli were applied at 72 dB through inserted earphones (Etymotic ER-1). We expect to observe significant changes in DWI parameters consistent with the structural maturation of cerebral white matter during adolescence. We further anticipate that these changes will be reflected by coincident changes in the amplitude and conduction speed of visual and auditory EPs. These studies provide foundational experiments for assessing the role of elevated kynurenic acid in white matter abnormalities and related cognitive impairments in people with schizophrenia.

Disclosures: P. Kochunov: None. **A.T. Summerfelt:** None. **P.L. Brown:** None. **M.C. Terzi:** None. **K. Yachera:** None. **K.V. Sathyasaikumar:** None. **X. Du:** None. **L.E. Hong:** None. **R. Schwarcz:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property

rights/patent holder, excluding diversified mutual funds); Kynexis, Vistagen. **P.D. Shepard:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.16

Topic: H.13. Schizophrenia

Support: NARSAD Grant
ARC Grant DP190101948

Title: Maldevelopment of primate pulvinar-prefrontal cortex connectivity reproduces the functional, behavioral and cellular changes observed in schizophrenia

Authors: *J. T. SCOTT[1], B. L. MENDÍVEZ VÁSQUEZ[1], N. S. HOSSEINI FIN[1], J. HOMMAN-LUDIYE[1], J. A. BOURNE[1,2];
[1]Australian Regenerative Med. Inst., Monash Univ., Melbourne, Australia; [2]Section on Cell. and Cognitive Neurodevelopment, Natl. Inst. of Mental Health, Natl. Inst. of Hlth., Bethesda, MD

Abstract: Schizophrenia (Sz) is characterised by the cellular and cognitive impairment of the prefrontal cortex (PFC) and dysfunction of the thalamus. One thalamic nucleus most heavily implicated is the medial pulvinar (PM), which has robust bidirectional connectivity with the PFC. Previously, we have revealed how another pulvinar nucleus, the inferior pulvinar (PIm), is crucial for the normal development of visual cortical areas and visuomotor behavior. Therefore, we propose that the PM is critical for the normal development of the PFC. Any perturbation to the PM and its connectivity with the PFC in early life will lead to the well-characterised cellular and cognitive phenotype associated with Sz. To test our hypothesis, we bilaterally lesioned (NMDA injection) the PM of neonatal (postnatal day 14) marmoset monkeys (*Callithrix jacchus*) using an MRI-guided stereotaxic surgery (*n*=5 lesion, *n=3* sham control, both sexes) and then longitudinally examined PFC anatomy, connectivity and function into adulthood (>18 months age (m.a.)). We selected primarily non-invasive methods akin to those used to assess human patients with Sz to maximise clinical translatability. First, marmosets underwent diffusion MRI to confirm the loss of frontal thalamocortical connectivity and characterise local changes in PFC cytoarchitecture. Next, resting-state epidural EEG was recorded from the PFC using fully internalised radiotelemetry implants to examine the function of local PFC circuits. Finally, animals underwent regimented behavioral training using a novel, home-cage integrated touchscreen platform and were assessed in cognitive tasks challenging PFC function. Diffusion MRI tractography revealed a reduction in PM-PFC connectivity in lesioned animals beginning from 6 m.a., coinciding with broadly reduced fractional anisotropy (potentially indicating loss of microstructural complexity) in PFC areas at 18 m.a. Further, these changes in the PFC coincided with a selective reduction of relative power in low gamma oscillations (30-80 Hz; -3.8% (+/- 2.8%)) obtained through resting-state EEG. Behaviorally, lesioned animals exhibited specific

impairment in maintaining working memory over extended delay periods (>2 s), a cognitive operation known to be sustained by gamma oscillations in the PFC. Post-mortem immunohistochemical staining of the PFC revealed a ~54% reduction in parvalbumin-expressing neurons, known generators of cortical gamma oscillations, in the thalamorecipient layer 3. Together, these findings highlight the importance of the PM in the normal development of the PFC and how its perturbation is central to anatomical and behavioral sequelae observed in Sz.

Disclosures: J.T. Scott: None. **B.L. Mendívez Vásquez:** None. **N.S. Hosseini Fin:** None. **J. Homman-Ludiye:** None. **J.A. Bourne:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.17

Topic: H.13. Schizophrenia

Support: BBSRC DTP Studentship funded

Title: Maternal immune activation induces fetal epigenetic dysfunction, developmental perturbations in glutamatergic signalling and adult cognitive deficit in a rodent model for schizophrenia

Authors: *R. M. WOODS[1], J. A. FLETCHER[1], I. G. HARRIS[1], J. M. LORUSSO[1], H. G. POTTER[2], H. M. KOWASH[1], J. D. GLAZIER[1], J. C. NEILL[1], M. HARTE[1], R. HAGER[1]; [1]Fac. Biol. Med. and Hlth., Univ. of Manchester, Manchester, United Kingdom; [2]Univ. of Central Lancaster, Lancaster, United Kingdom

Abstract: Introduction: Epidemiological studies demonstrate viral infection, resulting in maternal immune activation (mIA), during pregnancy is a risk factor for schizophrenia (SZ). We hypothesize that the maternal inflammatory response promotes epigenetic alterations in the fetal brain leading to changes in expression of key neurodevelopmental genes, which predisposes offspring to SZ. To test this hypothesis, we used our validated rat model for SZ, exposing pregnant dams to the viral mimetic polyinosinic:polycytidylic acid (poly(I:C)), which increases pro-inflammatory plasma cytokines, IL-6 and TNFα. **Objectives:** Establish DNA methylation patterns and associated gene/protein expression changes underpinning cognitive deficits in mIA-offspring. **Methods:** Pregnant Wistar rat dams were blindly assigned to receive 10mg/kg bodyweight poly(I:C) (mIA) or 0.9% saline (vehicle control) on gestational day (GD) 15 (N=8-10/group). Adult offspring (Postnatal day (PD) 100) were assessed using the attentional set-shifting task (ASST) for cognitive function underscored by prefrontal cortex (PFC) activity. Offspring PFC were dissected at multiple developmental timepoints (GD21, PD1, PD21, PD35 and PD100). DNA, RNA and protein were extracted for DNA methylation, mRNA (qPCR) and protein expression (WES) analysis, respectively (N=5-10/group). Data were analysed using general linear mixed models (GLMM). **Results:** Adult offspring exposed to mIA showed an

increased number of trials and errors in the extra-dimensional shift phase of the ASST, indicative of a cognitive deficit. Global DNA methylation, *Dnmt3a/b* mRNA expression and DNMT activity were increased in GD21 mIA-fetuses, indicative of epigenetic alterations in response to mIA. mRNA analysis across developmental timepoints showed decreases in Reelin signalling genes (*Reln, Dab1*), and both decreases (*Camk2b, Dlg4*) and increases (*Sgk1, Gpc4*) in glutamatergic modulators in mIA-offspring. Protein analysis across postnatal timepoints showed a reduction in AMPA receptor (GRIA1) expression in PD21 and PD35 mIA-offspring but no changes in expression of NMDA receptors (NR2A, NR2B). However, we found a decrease in the NR2A:2B and NMDA:AMPA receptor ratios in adult mIA-offspring. **Conclusions:** mIA induced early alterations in DNA methylation together with persistent changes in the expression of glutamatergic signalling genes and an imbalance of glutamatergic receptor ratios. We suggest that developmentally imbalanced glutamatergic signalling in mIA-offspring may contribute to adult cognitive deficits, comparable to cognitive symptoms in SZ.

Disclosures: R.M. Woods: None. **J.A. Fletcher:** None. **I.G. Harris:** None. **J.M. Lorusso:** None. **H.G. Potter:** None. **H.M. Kowash:** None. **J.D. Glazier:** None. **J.C. Neill:** None. **M. Harte:** None. **R. Hager:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.18

Topic: H.13. Schizophrenia

Support: NIH Grant 1R01MH123479-01A1

Title: Dentate Gyrus Lesion Prior to Adulthood Causes Hippocampal Hyperactivity Characterized by Synchronous Activity

Authors: *D. SCOTT[1], J. YAMAMOTO[2], C. A. TAMMINGA[3];
[1]Univ. of Texas, Southwestern Med. Ctr., Dallas, TX; [2]Psychiatry, [3]UT Southwestern Med. Ctr., Dallas, TX

Abstract: Post-mortem analyses of brain tissue from human schizophrenia shows evidence of impaired dentate gyrus (DG) function, coupled with downstream hippocampal hyperactivity. Little is known about the mechanisms of this putative psychosis biomarker. We have developed a reverse-translation mouse model to study critical features.
We expressed an inhibitory DREADD in the granule cells of the mouse dentate gyrus, and inhibited the region for 21 days in adolescent (6 week) and adult (10 week) C57BL/6J mice. Following this period, we assessed hippocampally-mediated behaviors, and quantified hippocampal subfield activity by assessing cFos expression in the mice. We further characterized the hippocampal local field potentials prior to, during, and following DG inhibition.
DG inhibition delivered during mouse adolescence, but not adulthood, results in impaired social

cognition and augmented fear conditioning, and this phenotype is associated with increased cFos expression in CA3 and CA1. Moreover, during the period of DG inhibition, we observed the emergence of bursts of EEG activity throughout the hippocampus, which we refer to as 'hyper synchronous events' (HSEs) when we chronically recorded dorsal CA1-DG axis of hippocampal neural activities. HSEs emerged in the second week of DG inhibition, and persist for months following the cessation of active DG inhibition. DG inhibition delivered during adulthood results in none of these effects. The increased CA3 activity, as well as the observed behavioral phenotype seen following adolescent DG inhibition is short-lived, normalizing within four weeks, while CA1 activity remains elevated.
Adolescence in mice appears to be a critical period in which decreased DG activity can induce hippocampal hyperactivity in CA3/CA1, characterized by HSEs, an outcome not observed in adult mice. This suggests a sensitive period of development in which the brain in sensitive to alterations in hippocampal activity and can result in psychosis-like behavioral outcomes as well as molecular changes.

Disclosures: D. Scott: None. **J. Yamamoto:** None. **C.A. Tamminga:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.19

Topic: H.13. Schizophrenia

Support: NSERC CGSD (2022-2025)
CIHR Grant #PJT-159586
CIHR Grant #CRU1126
MITACS Canada
CanaQuest Inc.

Title: The role of the basolateral amygdala in prenatal THC induced mesocorticolimbic pathophenotypes

Authors: *M. H. SARIKAHYA[1], K. WONG[1], S. COUSINEAU[2], M. DE FELICE[1], H. SZKUDLAREK[1], M. V. DEVUONO[1], K. K.-C. YEUNG[2], D. B. HARDY[3], W. RUSHLOW[1], S. R. LAVIOLETTE[1];
[1]Univ. of Western Ontario, [2]Biochem., [3]Physiol. and Pharmacol., Univ. of Western Ontario, London, ON, Canada

Abstract: Clinical and preclinical studies indicate prenatal cannabis exposure (PCE) pathologically effects fetal brain development and may increase vulnerability to neuropsychiatric disorders, including schizophrenia and mood/anxiety disorders. *In review* research from our lab suggests that fetal exposure to Δ9-THC sex-selectively impairs mesocorticolimbic (MCL) circuit function. However, there is a distinct lack of focus of PCE models on the BLA. The BLA plays a

central role within the MCL where it directly interacts with the VTA, PFC and HIPP. Importantly, our model exhibits significant VTA hyperdopaminergia, and sex-specific alterations to PFC/HIPP glutamate firing, alongside region- and sex-specific changes in dopamine (DA), glutamate/GABA molecular markers. These result with outward pathological behavioural manifestations with males exhibiting enhanced anxiety and both sexes exhibiting cognitive deficits. It is thus necessary to mechanistically explore the effect of PCE on the BLA. The present study characterized the interconnected pathophenotype of the BLA-MCL circuit using behavioural, electrophysiological, molecular, MALDI IMS, and mechanistic assays. Pregnant Wistar rats were assigned to VEH or 3mg/kg THC (daily, *i.p.*; n=10 dams/treatment; n=4 progeny/sex/dam) from gestational day (GD) 7 to GD22. A subset of progeny (n=8/treatment/sex) were sacrificed on PD21 for molecular assays of the NAc and BLA. Between PD70-85, a subset of progeny (n=20/treatment/sex) were assessed for anxiety, depression, prepulse inhibition, and contextual fear. Between PD90-120, *in vivo* electrophysiology was used to assess VTA DA-ergic neurons, glutamate, and GABA neurons in the posterior/anterior BLA, and NAc GABA neurons. Remaining offspring were sacrificed and NAc and BLA punch-outs were obtained for molecular and MALDI IMS assays. A behaviour naïve subset (n=10/sex/treatment) received intra-BLA cannulations for mechanistic assays between PD90-120. In line with previous results, males exhibit a significant anxiogenic phenotype; however, males also exhibited significantly less freezing, suggesting a deficit in contextual fear learning, consistent with significant increases in GAD67 expression; males also exhibit increases in D1R and GABAARα1. Female progeny did not exhibit any outward pathology but did exhibit significantly greater expression of vGLUT2, GABAARα1, and GABAARγ2. These suggest that the anxiogenic deficits observed in males is likely contingent on BLA disruptions, while the female progeny is protected from BLA-dependant etiology. Electrophysiological, MALDI IMS, and mechanistic assays are currently ongoing.

Disclosures: **M.H. Sarikahya:** None. **K. Wong:** None. **S. Cousineau:** None. **M. De Felice:** None. **H. Szkudlarek:** None. **M.V. Devuono:** None. **K.K. Yeung:** None. **D.B. Hardy:** None. **W. Rushlow:** None. **S.R. Laviolette:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.20

Topic: H.13. Schizophrenia

Title: Clustering Reveals Inflammatory and Behavioral Susceptibility to Maternal Immune Activation and Early Life Stress

Authors: *J. M. LORUSSO[1], R. M. WOODS[1], F. MCEWAN[1], I. HARRIS[2], H. TSUI[2], A. CHIRON[2], I. JIMENEZ PULIDO[2], I. SHAND[2], M. MAXWELL[2], J. D. GLAZIER[3], R. HAGER[1], M. HARTE[3];

[1]Div. of Evolution, Infection, and Genomics, [3]Div. of Pharm. & Optometry, [2]Univ. of Manchester, Manchester, United Kingdom

Abstract: Prenatal infection and early life stress are known risk factors for schizophrenia and other neurodevelopmental disorders. Following exposure, many individuals never develop these disorders. This resiliency is not well-understood and often neglected in animal models. The use of unbiased clustering based on behaviour can identify susceptibility at the individual level and offers the opportunity to investigate the mechanisms that may underpin it. Our study aimed to identify whether neuroinflammation predicted susceptibility in a two-hit rat model for schizophrenia. Cognition and social behaviour was investigated in offspring and used to determine individual resilience to the manipulation. Fetal brains were also investigated to identify prenatal resilience to Maternal Immune Activation (MIA)-induced neuroinflammation. We used Wistar rats in a two-hit neurodevelopmental model of schizophrenia. MIA was induced following i.p. injection of 10 mg/kg bodyweight poly(I:C), a viral mimetic, on gestational day (GD)15 with limited bedding and nesting (LBN) as a second stressor on postnatal days 1-10. Maternal cytokine responses were measured as a validation of MIA. Fetal brains were analysed at GD15 and GD16 for IL-6, IL-1β, IL-10, and TNF-α. Male and female offspring were tested on the novel object recognition (NOR) and social interaction (SI) tasks. To identify inflammatory mediators, we analzyed frontal cortices for IL-6 concentration by ELISA. MIA and LBN reduced cognition, and LBN affected sociability. Clustering analysis identified two clusters defined by either high or low cognition and social preference. MIA affected distribution to these clusters, with 50% of MIA offspring in the higher-performing cluster. No changes in adolescent IL-6 were present in the frontal cortex. Maternal TNF-α concentration predicted an increase in fetal brain IL-10, though only as a trend while MIA also increased fetal IL-6 concentrations. Clustering identified two groups based on inflammatory profiles. Cluster distribution was affected by MIA with approximately equal number of offspring in high and low inflammation clusters. Postnatally, MIA and LBN affected offspring behaviour. Half of MIA offspring appeared behaviourally susceptible to the insults, and frontal cortex inflammation did not predict resilience or susceptibility. This susceptibility may be determined prenatally, as non-response can be seen in a population of fetal MIA brains. Further investigation is needed to identify the mechanisms which maintain this susceptibility or initiate it prenatally, such as glutamatergic or glucocorticoid dysfunction.

Disclosures: **J.M. Lorusso:** None. **R.M. Woods:** None. **F. McEwan:** None. **I. Harris:** None. **H. Tsui:** None. **A. Chiron:** None. **I. Jimenez Pulido:** None. **I. Shand:** None. **M. Maxwell:** None. **J.D. Glazier:** None. **R. Hager:** None. **M. Harte:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.21

Topic: H.13. Schizophrenia

Support: Universidad Autónoma de Madrid - Comunidad Autónoma de Madrid (SI3-PJI-2021-00417)

Title: Foxp2 expression is altered in the thalamus of a rat model of schizophrenia

Authors: B. SANCHEZ MORENO[1], J. S. NACHER[2,3,4], ***J. GILABERT JUAN**[1];
[1]Univ. Autonoma de Madrid, Univ. Autónoma de Madrid, Madrid, Spain; [2]Neurobio. Unit, Dept. of Cell Biology, Inst. of Biotech. and Biomedicine, Univ. of Valencia,, Burjassot, Spain; [3]Spanish Natl. Network for Res. in Mental Health, CIBERSAM, Valencia, Spain; [4]Fundación Investigación Hosp. Clínico de Valencia, INCLIVA, Valencia, Spain

Abstract: FoxP2 is a transcriptional factor associated to schizophrenia disease in numerous studies, including our previous one where we showed the involvement of FoxP2 gene variants with reductions in grey matter density and gene expression in various brain regions of patients with schizophrenia. Furthermore, it is a gene related to language development. Indeed, deficits in FoxP2 function have been observed in individuals with alterations in language processing; also, FoxP2 has been associated with psychotic symptoms, such as auditory hallucinations. One of the main regions of the brain altered in schizophrenia is the thalamus, a region composed of multiple nuclei where sensory, motor, emotional, and cognitive neural pathways relay and connect with the cerebral cortex.
In the present study, we analyzed the expression of FoxP2 and related markers in the thalamus of a rat "dual hit" animal model of schizophrenia previously developed by our laboratory. We studied the distributions of cells containing FoxP2 protein in the different thalamic nuclei and compared the expression, at the cellular nuclei, of this protein with that of proteins related to chromatin status and gene expression.
FoxP2 expression in the rat thalamus is observed mainly in the midline and intralaminar nuclei. Specifically in the paraventricular nucleus. There is an increase of FoxP2 expression in the medial dorsal and paraventricular thalamic nuclei of the "dual hit" animal model of schizophrenia without changes in the FoxP2+ cell number. This increase in expression is accompanied by alterations in the intensity and distribution of markers related to the structure of the chromatin such as γH2A.X, H3K9ac or H3K27me3, which indicates alterations in the gene expression regulation.
These observations point to a possible role of FoxP2 in the regulation of thalamic nuclei and in the pathogenesis and/or pathophysiology of the psychotic phenotype.

Disclosures: B. Sanchez Moreno: None. **J.S. Nacher:** None. **J. Gilabert Juan:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.22

Topic: H.13. Schizophrenia

Support: NIMH K99MH129613-01

Title: Targeted modulation of MD to PFC thalamocortical circuits - a strategy to treat cognitive impairments in schizophrenia

Authors: *A. MUKHERJEE, M. HALASSA;
Brain and Cognitive Sci., MIT, Cambridge, MA

Abstract: Schizophrenia ranks within the top 15 leading causes of disability worldwide. Despite their side effects and treatment resistance, antipsychotic medications continue to be the primary treatment option for the disorder. To develop a new generation of targeted therapies, at par with modern medicine, we need to isolate discrete neural circuit deficits that underlie different symptoms of schizophrenia. In search of alternative therapeutic targets, the cognitive deficits in schizophrenia, which often emerge before the psychotic symptoms, have become a focus of recent scientific interest. While the cognitive deficits in schizophrenia have been linked to an aberrant engagement of the prefrontal cortex (PFC), recent studies have shown that the interactions of the PFC with its main thalamic partner, the medio dorsal thalamus (MD), is also perturbed. I have discovered genetically identifiable MD neurons that are essential for prefrontal control over flexible behavior. Specifically, one MD projection (MD_{D2}) that targets disinhibitory vasointestinal peptide (PFC_{VIP}) expressing prefrontal interneurons is required to amplify task relevant neural activity and maintain accurate performance in a cognitive task; while another MD projection (MD_{GRIK4}) that targets inhibitory parvalbumin (MD_{GRIK4} to PFC_{PV}) interneurons within the PFC suppresses task irrelevant activity to promote behavioral flexibility. Thus, rational targeting of MD-PFC sub circuits with neurostimulation might be a viable targeted therapeutics for schizophrenia and is the central aim of this project. Here using virus-based circuit tracing, *in vivo* electrophysiology and optogenetic manipulations I uncovered anatomical and functional perturbations in MD-PFC sub circuits in a mouse model that recapitulates a genetic deletion of schizophrenia predisposition in humans. Compared to wild types these mice showed a strong reduction in the suppression of $MDGRIK_4$ driven prefrontal spike rates. Furthermore, in a PFC dependent cognitive task that requires flexible switching of attentional control across task relevant inputs, these mice show delayed switching compared to wild types. Prior work had shown that MD driven suppression of PFC around the switch to a new cue set is causal to successful switching in this paradigm. In line with this data, I show that selective drive of the MD cell type that projects to PV neurons during switching rescued it to levels comparable with wild type mice. In conclusions, given the role of the MD as a primary source of information flow into the PFC, my work provides exciting evidence for the MD as a particularly attractive target for intervention in schizophrenia.

Disclosures: A. Mukherjee: None. **M. Halassa:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.23

Topic: H.13. Schizophrenia

Support: R56MH112642 from NIMH/NIH (to Uwe Rudolph)
R21MH104505 from NIMH/NIH (to Uwe Rudolph)
Harvard Brain Science Initiative Bipolar Disorder Seed Grant, supported by Ken and Liz Dauten (to Uwe Rudolph)
Stanley Center for Psychiatric Research, Broad Institute (to Uwe Rudolph)
K01MH107787 from NIMH/NIH (to Elif Engin)

Title: Increased copy number of the glycine decarboxylase (GLDC) gene differentially modulates hippocampal subregions in a mouse model of the pathophysiology of psychosis

Authors: *M. KAMBALI[1], Y. LI[2,4], P. UNICHENKO[5], J. FERIA[5], M. WANG[1,6], R. NAGARAJAN[1], J. LYU[1,6], J. LIU[3,4], V. VONGSOUTHI[7,8], C. J. JACKSON[7,8], E. ENGIN[3,4], G. E. HOMANICS[9], V. Y. BOLSHAKOV[2,4], C. HENNEBERGER[5], U. RUDOLPH[1,3,4];
[1]Dept. of Comparative Biosci, Col. of Vet. Med., Univ. of Illinois, Urbana-Campaign, Urbana, IL; [2]Cell. Neurobiol. Lab., [3]Lab. of Genet. Neuropharm, McLean Hosp., Belmont, MA; [4]Dept. of Psychiatry, Harvard Med. Sch., Boston, MA; [5]Inst. of Cell. Neurosciences, Univ. of Bonn, Bonn, Germany; [6]Neurosci. Program, Univ. of Illinois at Urbana-Champaign, Urbana, IL; [7]Res. Sch. of Chemistry,, [8]Australian Res. Council Ctr. of Excellence for Innovations in Peptide and Protein Sci., Australian Natl. Univ., Canberra, Australia; [9]Dept. of Anesthesiol. and Perioperative Med., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Genomic copy number variants (CNVs) have been implicated in the etiology of schizophrenia (SCZ) and bipolar disorder. A rare 9p24.1 CNV in two patients with psychosis that included multiple genes, including *GLDC* encoding the glycine-degrading enzyme glycine decarboxylase, has been found by others. We sought to determine whether this CNV is sufficient to induce biochemical or behavioral phenotypes in mice and which gene(s) would be underlying these phenotypes. We hypothesized that an increased copy number of *Gldc* would lead to increased degradation of glycine, eventually resulting in NMDA receptor hypofunction and schizophrenia-like phenotypes. To address this, we developed mouse models with a triplication (4 copies) of the 9p24.1 genes, or a triplication (4 copies) of Gldc alone (4cR) or of all other 9p24.1 genes (4cL). These mice were subjected to molecular (n=5 each genotype), biochemical (n=5 each genotype), immunofluorescence (n=5 each genotype), Seahorse assay (n=6 each genotype) and behavioral studies (n=10 each genotype) at 2-4-month of age. All genetic mouse models with increased copy numbers of *Gldc* showed increased GLDC protein in hippocampus (Hip), prefrontal cortex (PFC) and amygdala. Synaptoneurosomal fractions of hippocampus showed decreased BDNF levels and reduced activation of synaptic plasticity-related AKT-mTOR-CREB pathway in 4cR mice. Using the optical glycine FRET sensor GlyFS and two-photon excitation fluorescence microscopy, we found that in DG but not in CA1 of 4cR mice glycine levels are reduced compared to wild type controls. At the mPP-DG synapses, LTP was reduced in 4cR mice. The density of dendritic mushroom spines in DG was reduced in 4cR mice. In addition, the epigenetic marker of active gene promoters H3K4me3 was increased in CA1 and decreased in DG. A Seahorse mito stress test showed a deficit in mitochondrial function in DG but not in CA1. Furthermore, only in 4cR mice with an increased *Gldc* copy number, but not in 4cL mice with increased copy numbers of the other 9p24.1 genes, we found key behavioral deficits, i.e., startle habituation impairment, absence of latent inhibition to conditioned freezing, working memory deficits in both the Y- maze spontaneous alternation test and T- maze forced

alternation tests and sociability deficits, along with social novelty preference deficits. Thus, an increase in the copy number of the *Gldc* gene is sufficient to induce molecular, cellular and behavioral features consistent with a schizophrenia-like phenotype. Our results suggest that in the patients with the 9p24.1 duplication/triplication the increase in *GLDC* copy number may be an important contributing factor to pathophysiology.

Disclosures: **M. Kambali:** None. **Y. Li:** None. **P. Unichenko:** None. **J. Feria:** None. **M. Wang:** None. **R. Nagarajan:** None. **J. Lyu:** None. **J. Liu:** None. **V. Vongsouthi:** None. **C.J. Jackson:** None. **E. Engin:** None. **G.E. Homanics:** None. **V.Y. Bolshakov:** None. **C. Henneberger:** None. **U. Rudolph:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.24

Topic: H.13. Schizophrenia

Title: Thalamo-prefrontal circuit hypofunction mediates belief updating deficits in mice with schizophrenia-associated mutations

Authors: *T. ZHOU[1], Y. Y. HO[1], N. HARTLEY[2], A. FATH[1], M. M. HALASSA[1], G. FENG[1]; [1]Brain and Cognitive Sci., MIT, Cambridge, MA; [2]Broad Inst., Cambridge, MA

Abstract: Schizophrenia is a severe neuropsychiatric disorder that affects 1% of people worldwide. Delusion is one of the defining symptoms. Despite its importance, it has been difficult to directly study delusions in animals, presenting a major obstacle to understanding the underlying pathology of this severe disorder. Studies on patients have proposed that delusions can be explained by impaired belief updating in dynamic environments. Thus, we studied delusion by modeling belief updating in mice. Two synergistic approaches were applied. First, we developed a novel lever-pressing task for mice, which requires mice to make optimal decisions in a dynamic environment. By introducing variations of this task and building mathematical models, we identified parameters in the task indicating animals' belief updating rate and uncertainty levels. Second, we used CRISPR/Cas9 to generate a mouse model carrying high-risk rare variation in the gene Grin2a identified in schizophrenia patients by recent large scale exome sequencing studies. Mutant animals in our novel lever-pressing task showed slower belief updating rates and increased uncertainty levels. Using simultaneous in vivo single unit electrophysiological recordings in the prefrontal cortex (PFC) and medial dorsal thalamus (MD) in animals performing this task, we showed that PFC neurons encode values, essential for updating beliefs, while the MD encodes uncertainty in the lever-pressing task. Optogenetic inhibition of PFC in wild-type animals during the lever-pressing task dramatically decreased the belief updating rate while MD inhibition increased uncertainty, phenocopying the behavioral deficits in mutant animals. We further showed, using ex vivo electrophysiological recordings, the balance of excitatory/inhibitory (E/I) synaptic transmission on the MD-PFC circuit is impaired in

mutant animals. Last, we found that restoring the E/I balance on MD-PFC circuit by optogenetic approaches improved the behavioral performance of mutant mice in the lever pressing task. Overall, our data suggested that the thalamo-prefrontal circuit hypofunction mediates belief updating impairment in mice carrying the schizophrenia-associate mutations, paving the way for potential therapeutic circuit targets to treat this psychiatric disorder.

Disclosures: **T. Zhou:** None. **Y.Y. Ho:** None. **N. Hartley:** None. **A. Fath:** None. **M.M. Halassa:** None. **G. Feng:** None.

Poster

575. Schizophrenia

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 575.25

Topic: H.13. Schizophrenia

Support: Ministry of Health & Welfare (HU22C0150)
Ministry of Science & ICT (NRF-2022R1A2C3009749)
Ministry of Science & ICT (NRF-2017R1A5A1014708)
2022 Joint Research Project of Institutes of Science and Technology
GIST Research Institute (GRI) IIBR grant funded by the GIST in 202

Title: Effect of Vitamin D Deficiency on Perineuronal Nets and Gamma-Band Oscillations in mice

Authors: *S. YU, M. PARK, J. KANG, E. LEE, J. JUNG, T. KIM;
Gwangju Inst. of Sci. and Technol., Gwangju Inst. of Sci. and Tech., Gwangju, Korea, Republic of

Abstract: Vitamin D plays an essential role in cognitive functions as well as regulating calcium homeostasis and the immune system. Many epidemiological studies have also shown the close relationship between vitamin D deficiency (VDD) and the risk of schizophrenia. Cortical gamma-band oscillations (GBO) are associated with cognitive functions, such as attention and memory. Patients with schizophrenia show abnormal GBO with increased spontaneous GBO and decreased evoked GBO. However, the direct effect of VDD on GBO remains unknown. Parvalbumin interneurons, which predominantly contribute to the generation of GBO, are surrounded by perineuronal nets (PNN). We sought to investigate the associations among VDD, PNN, and GBO. Here, we injected a viral vector (AAV5-DIO-ChR2-eYFP) into the basal forebrain stereotaxically and implanted electrodes for electroencephalogram (EEG). At baseline, the evoked and spontaneous EEG power at the gamma frequency band was measured in 4-month-old male PV-Cre mice. After six and twenty weeks of vitamin D deficient food administration, the power of GBO was measured in the VDD condition. Next, we injected the chondroitinase ABC (ChABC) enzyme into the frontal cortex to eliminate PNN. We found that the VDD group showed decreased power of both optogenetically- and auditory-evoked GBO,

whereas the spontaneous GBO increased. Enzymatic digestion of PNN showed similar changes in GBO. Taken together, we suggest that VDD could result in decreased PNN and, consequently, increase the spontaneous GBO and decrease the evoked GBO, reminiscent of the aberrant GBO in schizophrenia. These results show that VDD might increase the risk of schizophrenia and aggravate the cognitive symptoms of schizophrenia.

Disclosures: **S. Yu:** None. **M. Park:** None. **J. Kang:** None. **E. Lee:** None. **J. Jung:** None. **T. Kim:** None.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.01

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH 1R01EB024261
HHMI
Lisa Yang
John Doerr
Schmidt Futures
Open Philanthropy
NIH 1R01MH123403

Title: Towards routine reconstruction of C. elegans connectomes, cell states, and cell types, through optimized expansion microscopy

Authors: *Y. LU[1], C. ZHANG[1], T. W. SHIN[1], C.-C. YU[1], M. A. SNEVE[1], B. AN[1], A. M. MAUERMANN[1], S. H. SHIM[1], E. S. BOYDEN[1,2];
[1]MIT, Cambridge, MA; [2]HHMI, Cambridge, MA

Abstract: Connectomes, complete wiring diagrams of entire brains, offer the opportunity to connect the detailed circuitry of the nervous system to emergent dynamics and consequent behavior. Serial section electron microscopy, the current strategy for mapping connectomes at nanoscopic level, has exquisite spatial resolution, but struggles with molecular identification, key to interpreting connectomes in terms of physiological properties; in addition, it requires expensive hardware and detailed expertise. A simple method for connectomics would enable routine analysis of cell connectivity, cell types, and cell states, in everyday neuroscience. By optimizing and applying our recent invention of expansion microscopy (ExM), which through physical magnification of biological specimens enables nanoimaging to be performed on ordinary microscopes (*Science, 347(6221), 543-548*), such inexpensive and simple molecularly-annotated connectomics may soon be possible. We are using the nematode *Caenorhabditis elegans*, the only species whose complete connectome has been mapped (by electron microscopy), as a testbed. *C. elegans* is surrounded by a tough cuticle; we developed a way of

embedding worms in a hydrogel equipped with proteases, to selectively remove the cuticle without affecting internal molecules. This allowed us to isotropically expand the worms to high expansion factors without losing molecular information. Both exogenously expressed and endogenous proteins, including synaptic proteins, could then be revealed at nanoscopic resolution by immunostaining. To trace neurons, we are distinguishing densely packed neurites by combining Brainbow-style color-coding and lipid staining. For color-coding, we generated worm strains that express fluorescent proteins (FPs) with cell-type specificity. These cytosolic FPs then differentiate neighboring neurites and encode cell types simultaneously. The color code helps physically segment the neurons, potentially facilitating tracing. For lipid staining, we devised a molecular tag that labels cell membranes and is compatible with immunostaining. Together, we are achieving delineation of neurites and annotation of synapses by expansion microscopy in a high throughput fashion. Therefore, our method is potentially capable of reconstructing complete connectomes in everyday neuroscience, revealing wiring, molecular composition, and thus cell types and states, after functional experiments are complete.

Disclosures: **Y. Lu:** None. **C. Zhang:** None. **T.W. Shin:** None. **C. Yu:** None. **M.A. Sneve:** None. **B. An:** None. **A.M. Mauermann:** None. **S.H. Shim:** None. **E.S. Boyden:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); ESB is an equity holder in a company in the space of commercial applications of expansion microscopy..

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.02

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: National Science Foundation Graduate Research Fellowship Grant No. 1122374
NIH 1R01EB024261
Lisa Yang
NIH 1R01AG070831
HHMI
NIH 1R01MH123403
ERC Synergy Grant No 835102

Title: Ultrastructural membrane expansion microscopy (umExM) for electron microscopy-like imaging of brain circuits

Authors: *T. W. SHIN[1,2], C. ZHANG[3], B. AN[3], Y. LU[3], B. GUNER-ATAMAN[3], C. M. MITCHELL[2], D. LEIBLE[2], H. WANG[2], C.-C. YU[2], L.-H. TSAI[2,4,5], E. S. BOYDEN[1,2,3,6]; [1]Dept. of Media Arts and Sci., [2]Dept. of Brain and Cognitive Sci., [3]McGovern Inst. for Brain Res., MIT, Cambridge, MA; [4]Picower Inst. for Learning and Memory, Cambridge, MA; [5]Broad

Inst. of Harvard and Massachusetts Inst. of Technol., Cambridge, MA; [6]Howard Hughes Med. Inst., Cambridge, MA

Abstract: Electron microscopy is the primary technology currently used to delineate the detailed shapes of neurons in brain circuits, down to synaptic resolution, important for connectomics. Ideally, however, such imaging would be possible on a conventional light microscope, so that anyone could delineate the detailed shapes of neurons, and how they are connected, in neural circuits. In addition, ideally, it would be possible to image such neural circuits at scale, and identify and localize biomolecules in the detailed ultrastructural context of neural circuits. Recently, building from our invention of expansion microscopy (ExM), we have shown that lipid stains can be constructed that enable electron microscopy-like images to be taken on ordinary light microscopes, and for biomolecules to be identified and localized through fluorescent antibody staining, but with a low resolution of ~80 nm or so (bioRxiv 829903). Here we report a new generation of lipid stains, as well as a newly optimized ExM protocol, that aim to achieve images with comparable resolution to electron microscopy - sufficient to delineate the shapes of neurons, with molecular contrast, and pointing the way towards the possibility of dense connectomics via light microscopy. In particular, we have now generated two lipid stains that, mixed together, delineate the boundaries of neurons with excellent contrast. We also have generated a novel lipid preservation process to preserve ultrastructure in the context of fixation and expansion steps, as well as an optimized gelation chemistry recipe with an 8x expansion factor. Finally, we optimize the entire procedure to maximize the lipid membrane integrity. We demonstrate the ability of this novel protocol, which we call ultrastructure membrane expansion microscopy (umExM), to map out the shapes of neurons and the locations of key biomolecules with ~50 nm resolution, sufficient to resolve axons, with ~20 nm resolution achieved through an iterative version of umExM (iumExM), which may be sufficient to facilitate connectomics. Combined with barcoding strategies also under development in our lab, we aim to share this protocol throughout neuroscience to assist with the democratization of neural morphology measurement and connectomics

Disclosures: T.W. Shin: None. **C. Zhang:** None. **B. An:** None. **Y. Lu:** None. **B. Guner-Ataman:** None. **C.M. Mitchell:** None. **D. Leible:** None. **H. Wang:** None. **C. Yu:** None. **L. Tsai:** None. **E.S. Boyden:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Expansion Technologies, Massachusetts Institute of Technology.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.03

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH 1R01EB024261
Lisa Yang

HHMI
John Doerr
Open Philanthropy

Title: Extracellular Space Staining for Expansion Microscopy

Authors: *A. EMENARI[1], E. BOYDEN[1,2];
[1]MIT, Cambridge, MA; [2]HHMI, Cambridge, MA

Abstract: Expansion microscopy (ExM) enables, through physical magnification, nanoimaging on conventional microscopes, and as such, is supporting the analysis of many cellular and tissue components, ranging from proteins to nucleic acids to lipids (Nature Methods 16.1 (2019): 33-41). Mapping the extracellular space (ECS) is important not only for the study of this key tissue structure, which contains various active components in its own right, and governs features like the flow of electric charge around cells. In connectomics, ECS mapping would offer the possibility of simple, high throughput mapping of neural shapes and wiring, by providing a fundamental contrast with the intracellular milieu. Here, we show a 20x ExM protocol and an ECS staining protocol that works on fixed tissues, and that is compatible with the expansion process. We term this process expansion microscopy with extracellular space staining (ExECS). We used hyaluron amine, equipped with polymer-anchorable side chains, a biotin moiety (for later fluorescent streptavidin staining), and click conjugation pairs that enable these ECS filling molecules to be iteratively added and grown into a network that fills the extracellular space. We validate the reagents and the procedure of administering them, and show the kinds of imaging that this labeling and expansion microscopy can support, in mouse brain circuitry.

Disclosures: **A. Emenari:** None. **E. Boyden:** Other; ESB is an equity holder in a company in the space of commercial applications of expansion microscopy..

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.04

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NSF GRFP Award #1745302 to MES
MathWorks Science Fellowship to MES
James and Patricia Poitras Center for Psychiatric Disorders Research at MIT
Hock E. Tan and K. Lisa Yang Center for Autism Research
Tom Stocky
NIH 1R01EB024261
Kathleen Octavio

Title: Multiplexed Expansion Revealing (multi-ExR): Development and Application to Mapping Nanostructures in Healthy and Alzheimer's Disease Brains

Authors: J. KANG[1], ***M. E. SCHROEDER**[2], M. ZENG[1], Y. LEE[1], K. L. TITTERTON[1], Z. PENG[2], L.-H. TSAI[3], G. FENG[1], E. BOYDEN[1,4];
[1]McGovern Inst. for Brain Res., [2]Brain and Cognitive Sci., [3]Picower Inst. for Learning and Memory, MIT, Cambridge, MA; [4]HHMI, Cambridge, MA

Abstract: The ability to study crowded and complex three-dimensional molecular assemblies in brain cells remains a challenge, as electron microscopy lacks molecular contrast, and the resolution of widely accessible conventional optical microscopy is limited. We recently developed expansion revealing (ExR; Sarkar et al., *Nature Biomedical Engineering*, accepted), a novel expansion-based technology that enables super-resolution imaging of brain tissue using conventional fluorescent antibodies, which are applied post-expansion, and thus benefit from decrowding of proteins from one another. The better labeling that results, in turn enables visualization of previously hidden brain nanostructures. Here, we validate and demonstrate multiplexed expansion revealing (multi-ExR), a new variant of ExR that enables immunostaining and thus visualization of up to 20 proteins in the same tissue sample using conventional antibodies, with ~20 nm resolution. We validate multi-ExR by staining for the same target synaptic proteins over seven rounds of staining, imaging, and stripping, showing negligible bleed-through between rounds, maintenance of high signal-to-noise ratio, and mean registration accuracy of ~20nm. Using this technology, we revealed the three-dimensional nanoscale organization of ten synaptic proteins at synapses in the mouse primary somatosensory cortex (**Fig 1a**). We further demonstrate multi-ExR in the 5xFAD mouse model of Alzheimer's disease, showing significantly decreased number and intensity of synaptic proteins (PSD95, Homer, SynGAP, NR1 and NR2B), down-regulated expression level of CaMKII, and thread-like beta-amyloid nanodomains in 5xFAD samples compared to wild-type controls (**Fig. 1b-e**). multi-ExR enables high-throughput, low-cost super-resolution imaging of up to twenty proteins in a single sample, thus enabling unprecedented access to molecular environments and facilitating the discovery of previously undiscovered biological structures. Such multiplexed nanoimaging may suggest novel protein-protein interactions.

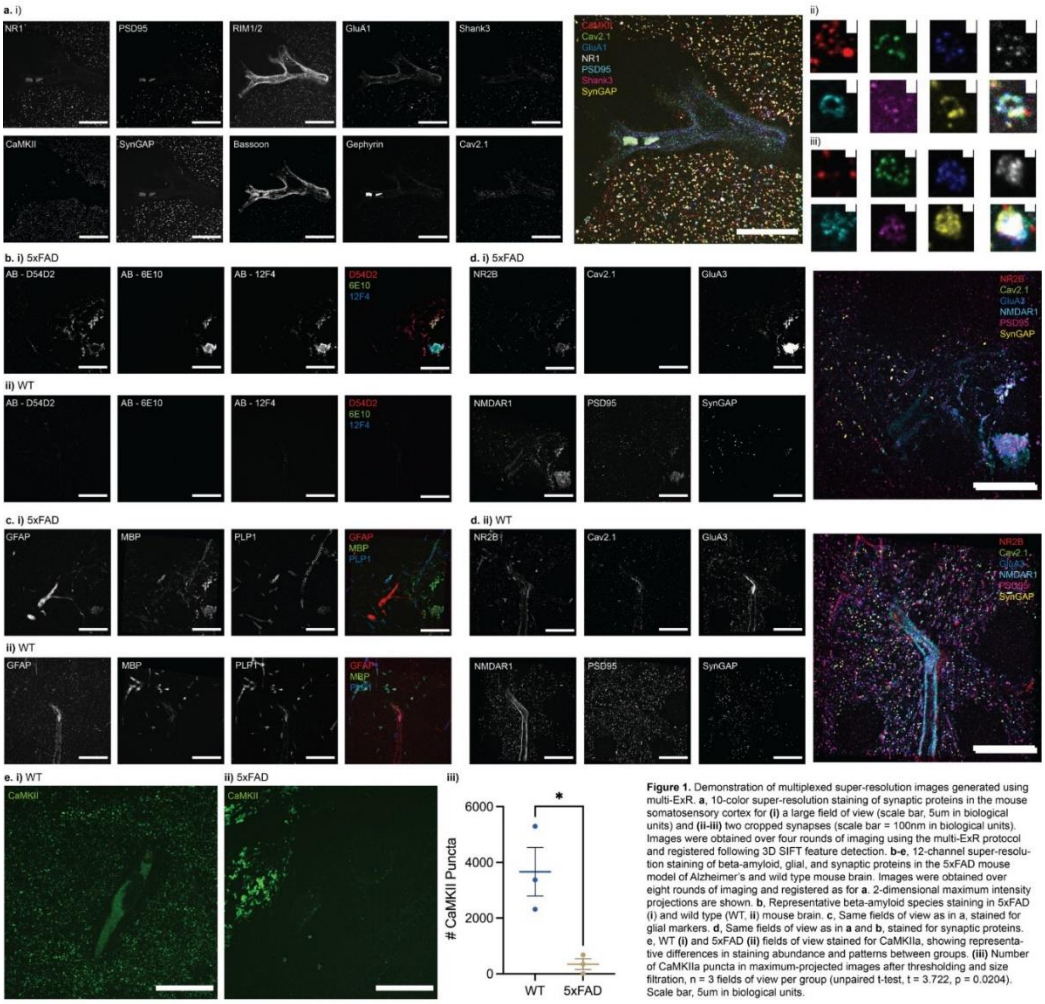


Figure 1. Demonstration of multiplexed super-resolution images generated using multi-ExR. **a**, 10-color super-resolution staining of synaptic proteins in the mouse somatosensory cortex for **(i)** a large field of view (scale bar, 5um in biological units) and **(ii-iii)** two cropped synapses (scale bar = 100nm in biological units). Images were obtained over four rounds of imaging using the multi-ExR protocol and registered following 3D SIFT feature detection. **b-e**, 12-channel super-resolution staining of beta-amyloid, glial, and synaptic proteins in the 5xFAD mouse model of Alzheimer's and wild type mouse brain. Images were obtained over eight rounds of imaging and registered as for **a**. 2-dimensional maximum intensity projections are shown. **b**, Representative beta-amyloid species staining in 5xFAD **(i)** and wild type (WT, **ii**) mouse brain. **c**, Same fields of view as in a, stained for glial markers. **d**, Same fields of view as in **a** and **b**, stained for synaptic proteins. **e**, WT **(i)** and 5xFAD **(ii)** fields of view stained for CaMKIIa, showing representative differences in staining abundance and patterns between groups. **(iii)** Number of CaMKIIa puncta in maximum-projected images after thresholding and size filtration, n = 3 fields of view per group (unpaired t-test, t = 3.722, p = 0.0204). Scale bar, 5um in biological units.

Disclosures: **J. Kang:** ; JK is a patent-filer for ExR and multi-ExR technology. **M.E. Schroeder:** ; MES is a patent-filer for multi-ExR technology.. **M. Zeng:** None. **Y. Lee:** None. **K.L. Titterton:** None. **Z. Peng:** None. **L. Tsai:** None. **G. Feng:** None. **E. Boyden:** ; ESB is an equity holder in a company in the space of commercial applications of expansion microscopy, and patent-filer for ExR and multi-ExR technology..

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.05

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: Koch Frontier Award
Center for Neurobiological Engineering at MIT

NIH 1R01EB024261
Tom Stocky
Kathleen Octavio
Jed McCaleb & James Fickel
Good Ventures

Title: Expansion Microscopy of Liquid Biopsies

Authors: *J. ARONSON[1,2], N. HAN[3], A. B. MILLER[4,5], C. BRAY[5], S. R. MANALIS[6,5], E. S. BOYDEN[1,2,7,5,8];
[1]Dept. of Brain & Cognitive Sciences, MIT, Cambridge, MA; [2]McGovern Inst. for Brain Research, MIT, Cambridge, MA; [3]Dept. of Electrical Engin. & Computer Science, MIT, Cambridge, MA; [4]Harvard-MIT Hlth. Sci. and Technol., Cambridge, MA; [5]Koch Inst. for Integrative Cancer Research, MIT, Cambridge, MA; [6]Dept. of Biol. Engineering, MIT, Cambridge, MA; [7]Media Arts and Sciences, MIT, Cambridge, MA; [8]Howard Hughes Med. Inst., Cambridge, MA

Abstract: Biology is based on nanoscale building blocks, biomolecules, which interact over nanoscale distances. Diseases, especially in their earliest stages, are associated with subtle changes in the presence and organization of biomolecules in cells and tissues. Expansion microscopy (ExM), a method of physical specimen expansion that preserves nanoinformation, and thus enables molecular mapping on conventional microscopes, is spreading rapidly through biology, with many hundreds of experimental papers and preprints to date. Some early studies have also shown ExM to physically magnify small changes found early in a disease, making them more obvious to clinical investigators. Here we ask whether this could be adapted to become a practical clinical diagnostic tool for early disease, by extending ExM to the imaging of liquid biopsies - easily obtained, minimally invasively extracted, specimens from patients (e.g., blood, saliva). For a proof of concept, we first adapted ExM to accelerate early detection of cancer in liquid biopsies, with a focus on detecting and characterizing circulating tumor cells (CTCs), with a hope to eventually use ExM of liquid biopsies (or liquidX for short) to detect brain diseases such as Alzheimer's. The first step was to adapt ExM to cells in suspension, beginning with dissociated cells from a fast growing, easy to culture cell line, HEK293 cells. We developed protocols for incorporating such cells into expandable hydrogels, followed by expansion and imaging. Specifically, we got cells to adhere directly to the bottom of wells treated to capture cells in suspension, with a ~70% yield. We also adapted the expansion microscopy protocol, tuning the composition of each recipe, to maximize expansion factor and minimize distortion and cracking. Having finished the initial protocol development, we are now validating the liquidX protocol on small cell lung carcinoma (SCLC) cells, showing that such cells, captured as described above, can be processed with many different ExM protocols. We have successfully visualized antibodies labeling Tomm20, α-tubulin, N-Cadherin, E-Cadherin, and EpCam. In the future, this protocol may be useful for the early detection of a variety of diseases, with a minimum of invasiveness. LiquidX may be an avenue for nanoscale exploration of other biological components of liquid biopsies, such as extracellular vesicles. Ultimately, this protocol holds exciting potential for non-invasive, longitudinal insight into early and subsequent changes in brain diseases.

Disclosures: J. Aronson: None. **N. Han:** None. **A.B. Miller:** None. **C. Bray:** None. **S.R. Manalis:** None. **E.S. Boyden:** E. Ownership Interest (stock, stock options, royalty, receipt of

intellectual property rights/patent holder, excluding diversified mutual funds); Expansion Technologies.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.06

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: Jed McCaleb, James Fickel
Lisa Yang
John Doerr
HHMI
NIH R01MH122971
NIH RF1NS113287
NIH 1R01MH123977

Title: Temporally multiplexed imaging for simultaneous observation of large numbers of fluorescent signals in living cells

Authors: *Y. QIAN[1], O. T. CELIKER[1,2], Z. WANG[1,3], E. S. BOYDEN[1,3,4];
[1]McGovern Inst. for Brain Res., [2]Dept. of Electrical Engin. and Computer Sci., [3]Dept. of Media Arts and Sci., MIT, Cambridge, MA; [4]Howard Hughes Med. Inst., Cambridge, MA

Abstract: Many processes in the brain, from learning and memory, to development and aging, to disease initiation and progression, involve dynamic changes in gene expression, as well as biomolecular interactions that occur in complex cascades, both within and between cells. Ideally one would be able to image many such molecular signals at once, in individual cells, to see how they interact. Without this ability, it is hard to determine the relationships between signals - for example, if when signal A is high in a given cell, signal B is low, and when signal A is low in a given cell, signal B is high, imaging of A and B in separate cells would miss out on this relationship. Seeing such relationships could lead to a better understanding of how signaling cascades and networks govern important biologic phenomena. Recently we published a spatial multiplexing method which clusters fluorescent reporters into puncta that we call signaling reporter islands (SiRIs), which enable large numbers of fluorescent signals to be imaged at once from different points within the same cell (Cell, 183(6), 1682-1698). However, SiRI technology relies on the availability of existing dynamic fluorescent indicators and cannot be used to monitor gene expression or biological functions for which no fluorescent reporter exists. Here, we present temporally multiplexed imaging (TMI), a strategy to use a set of reversibly photoswitchable fluorescent proteins (rsFPs), each with a different off-switching rate, to represent different biological signals. Due to their distinct clocklike properties, a movie recording the fluorescence fluctuations of the rsFPs in a cell can be linearly decomposed into a sum of the individual fluorescence fluctuation traces, each weighted by the brightness of its

corresponding fluorophore; the fluorophore brightness, thus extracted, can be then displayed on the computer for each fluorophore channel of the image. In this way, many different cellular signals can be imaged at the same time. We demonstrated that TMI allows up to six signals to be simultaneously imaged in live cells using a single optical channel of a standard epifluorescent or confocal microscope, with low (e.g., few %) crosstalk between the signals. We further show that TMI can be used in a wide diversity of biological contexts such as visualizing phases of the cell cycle, exploring the circadian rhythm, and measuring multiple kinase activities at once. TMI will help systematically explore dynamic changes in gene expression in the living brain over time, and many other important biological processes.

Disclosures: **Y. Qian:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); MIT. **O.T. Celiker:** None. **Z. Wang:** None. **E.S. Boyden:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); MIT.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.07

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH 1R01EB024261
Good Ventures
Lisa Yang
HHMI
NIH 1R01MH123403
NIH 1R56AG069192
NIH R01MH124606

Title: Next-generation expansion sequencing

Authors: *R. ZHANG[1], D. GHOSH[2], B. PRYOR[2], Y. CUI[2,1], S. ALON[2,1,7], D. GOODWIN[2,1], A. SINHA[2,3], A. WASSIE[2,1,4,5], F. CHEN[2,8], E. S. BOYDEN[2,1,6,5,9];
[1]Media Arts and Sci., [2]McGovern Inst., [3]Program in Hlth. Sci. and Technol., [4]Dept. of Biol. Engin., [5]Dept. of Brain and Cognitive Sci., [6]Koch Inst. for Integrative Cancer Res., MIT, Cambridge, MA; [7]Fac. of Engin., Bar-Ilan Univ., Ramat Gan, Tel Aviv District, Israel; [8]Broad Inst. of MIT and Harvard, Cambridge, MA; [9]Howard Hughes Med. Inst., Chevy Chase, MD

Abstract: Expansion sequencing (ExSeq, Science, 371 (6528), eaax2656 (2021)), integrates expansion microscopy (ExM) with *in situ* sequencing, allowing RNA transcripts to be identified and localized in physically expanded (~3-4X linear expansion) samples. This enables highly multiplexed mapping of RNAs with nanoscale precision throughout cells and tissues. The

targeted form of ExSeq enables hundreds of preselected RNAs to be mapped in the same sample. Previously, we reported that targeted ExSeq could be used to map the cell types of the mouse primary visual cortex, and study nanoscale RNA localization patterns in subcellular compartments in neurons across the mouse hippocampus, including dendrites and dendritic spines. We now have improved targeted ExSeq by (i) reducing cost; (ii) improving the sequencing chemistry; and (iii) reducing human labor. In more detail: we have developed a new small molecule anchor to tether RNAs to the expansion hydrogel, which reduces the cost of the procedure multifold. This new anchor binds to other biomolecule types, and in combination with optimized tissue softening methods, makes ExSeq compatible with techniques for labeling not only RNA, but also proteins and other biomolecule types simultaneously. Importantly, this method enables post-expansion staining to be performed in the context of ExSeq, useful because of the improved target access observed with post-expansion staining. In our current study, we have implemented a sequencing-by-ligation (SBL) chemistry that can be performed under isothermal conditions, helpful for simplifying the procedure, and facilitating the automation of round-to-round sequencing, which reduces human labor while increasing throughput. Such improvements hold the promise to open up avenues for nanoscale spatial mapping of brain circuits with higher precision, thus enabling a better understanding of brain function in health and disease.

Disclosures: **R. Zhang:** None. **D. Ghosh:** None. **B. Pryor:** None. **Y. Cui:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); U.S. Appl. No. 63/326,346. **S. Alon:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); US10995361B2, US10526649B2. **D. Goodwin:** None. **A. Sinha:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); US10995361B2, US10526649B2. **A. Wassie:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); US10995361B2, US10526649B2. **F. Chen:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); US10995361B2, US10526649B2. **E.S. Boyden:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Expansion Technologies, US10995361B2, US10526649B2.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.08

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

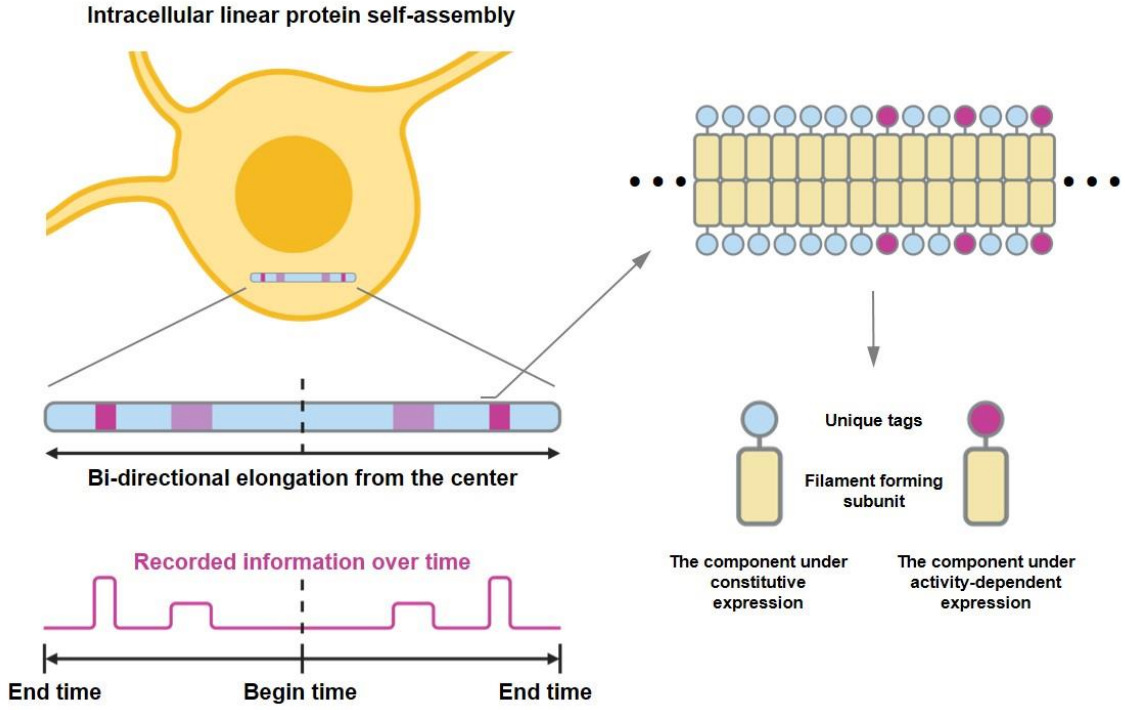
Support: J. Douglas Tan Postdoctoral Fellowship
NIH 1R01MH123977
NIH R01DA029639

NIH R01MH122971
NIH RF1NS113287
NSF 1848029
NIH UF1NS107697

Title: Recording of cellular physiological histories along optically readable self-assembling protein chains

Authors: *C. LINGHU[1], B. AN[1], M. SHPOKAYTE[2], O. T. CELIKER[1], N. SHMOEL[1], R. ZHANG[1], C. ZHANG[1], D. PARK[1], W. PARK[3], S. RAMIREZ[2], E. BOYDEN[1,4];
[1]MIT, Cambridge, MA; [2]Boston Univ., Boston, MA; [3]Kansas State Univ., Manhattan, KS;
[4]HHMI, Cambridge, MA

Abstract: Observing cellular physiological histories is key to understanding normal and disease-related processes, but longitudinal imaging is laborious and equipment-intensive. A tantalizing possibility is that cells could record such histories in the form of digital biological information within themselves, for later high-throughput readout. Here we show that this concept can be realized through information storage in the form of growing protein chains made out of multiple self-assembling subunits bearing different labels, each corresponding to a different cellular state or function, so that the physiological history of the cell can be visually read out along the chain of proteins. Conveniently, such protein chains are fully genetically encoded, and easily readable with simple, conventional optical microscopy techniques, compatible with visualization of cellular shape and molecular content. We use such expression recording islands (XRI) to record gene expression timecourse downstream of pharmacological and physiological stimuli, in cultured neurons and in living mouse brain.



Disclosures: **C. Linghu:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Changyang Linghu and Ed Boyden declare they applied for a US patent based on this work.. **B. An:** None. **M. Shpokayte:** None. **O.T. Celiker:** None. **N. Shmoel:** None. **R. Zhang:** None. **C. Zhang:** None. **D. Park:** None. **W. Park:** None. **S. Ramirez:** None. **E. Boyden:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Changyang Linghu and Ed Boyden declare they applied for a US patent based on this work..

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.09

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH 1R01EB024261
Lore McGovern
Good Ventures
Lisa Yang
NIH 1R01AG070831
HHMI
NIH 1R01MH123403

Title: Scalable sets of brainbow-like protein barcodes for scalable expansion microscopy-based neural morphology measurement and connectomics

Authors: *B. AN[1], S. G. RODRIQUES[2], K. LEUNG[1], A. PAYNE[3], T. XIAO[1], F. RIEGER[4], R. PARK[2], S. PANDIT[1], S. TRUCKENBRODT[3], M. SEARS[1], D. LEIBLE[1], J. KORNFELD[4], M. S. FEE[1], E. S. BOYDEN[1,5];
[1]McGovern Brain Inst., MIT, Cambridge, MA; [2]Francis Crick Inst., London, United Kingdom; [3]E11 Bio, Alameda, CA; [4]Max Planck Inst. for Bio. Int., Martinsried, Germany; [5]HHMI, Cambridge, MA

Abstract: Mapping neural circuits with synaptic resolution, ideally at a density sufficient to obtain detailed maps of local neural circuits, is challenging, with electron microscopy the dominant method currently used. However, electron microscopy is expensive, slow, and requires skill to perform, and struggles with the identification and localization of specific biomolecules in these ultrastructural contexts. Recently, the combination of expansion microscopy (ExM) and brainbow, the expression of multiple fluorophores in combinations in brain cells, has enabled visualization of small numbers of neurons (e.g., Nature Biotechnology, 34(9), 987-992), but the number of color combinations possible is limited. We here report our progress on developing a scalable set of protein epitopes that can be safely expressed in combinations in neurons, so that each cell gets a unique combination of epitopes, and can be distinguished during serial staining,

imaging, and washing steps. The technology is also compatible with immunostaining against synaptic and other proteins, for visualization of key biomolecules within and between neurons.We have now identified 10 epitope-labeled proteins that can be combinatorially expressed in random sets, e.g. via viral delivery, in mouse hippocampus and cortex. (In theory, n epitopes would enable 2^n different neurons to be distinguished, as an upper bound.) These epitopes uniformly label neurons throughout axons and dendrites, and are mutually orthogonally stainable with different commercially available antibodies. In more detail, we label each epitope with an antibody tagged with a DNA oligonucleotide, which is then anchored to a swellable ExM hydrogel. After expansion is complete, the DNA oligonucleotides can be visualized through rapid, highly multiplexed *in situ* hybridization, followed by imaging, and washing. With 4x-fold expansion, resulting in 70 nm resolution, and antibody staining against pre and post synaptic markers, we anticipate that this toolkit will be easily deployed in everyday neuroscience for the mapping of sparse connectomes in the short term, and in combination with optimized lipid stains being worked on by others in the group, may help with the obtaining of dense connectomes.

Disclosures: B. An: None. **S.G. Rodriques:** None. **K. Leung:** None. **A. Payne:** None. **T. Xiao:** None. **F. Rieger:** None. **R. Park:** None. **S. Pandit:** None. **S. Truckenbrodt:** None. **M. Sears:** None. **D. Leible:** None. **J. Kornfeld:** None. **M.S. Fee:** None. **E.S. Boyden:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); ESB is an equity holder in a company in the space of commercial applications of expansion microscopy.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.10

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: Jed McCaleb, James Fickel
Lisa Yang
NIH 1R01AG070831
HHMI
NIH 1U01NS120820
NIH 1R01MH123977
NIH R01DA029639

Title: Spatial multiplexing of fluorescent reporters for imaging signaling network dynamics in intact brain circuitry in vivo

Authors: *N. SHMOEL DAVID[1,2,3,4], C. LINGHU[1,2,3,4], K. R. JENKS[2], K. TSIMRING[2], M. SHPOKAYTE[6,7], O. T. CELIKER[1,2,3,4], J. F. NORMAN[8], S. RAMIREZ[6], M. SUR[2], E. S. BOYDEN[1,2,3,4,5];
[1]McGovern Inst., [2]Brain and Cognitive Sci., [3]Biol. Engin., [4]Media Arts and Sci., [5]Howard

Hughes Med. Inst., MIT, Cambridge, MA; [6]Psychological and Brain Sci., [7]Grad. Program for Neurosci., [8]Biomed. Engin., Boston Univ., Boston, MA

Abstract: Molecular signaling between and within cells plays a key role in normal physiology, converting cell inputs into outputs through detailed intracellular molecular networks, and goes awry in many diseases. Mapping such signal transduction networks *in vivo* is critical for understanding how cells generate appropriate responses to given input, identifying what pathways are impaired by disease, and subsequently identifying therapeutic targets. Traditionally, bioengineers have created fluorescent indicators of different colors to map the relationship between different signals in a living cell, but recently we revealed a different way to increase the multiplexing capacity of live imaging localizing distinct fluorescent reporters at different places in a cell, so that they can report signals simultaneously, from their unique positions (Cell, 183(6), 1682-1698). By fusing a fluorescent reporter to a pair of self-assembling peptides, they can be stably and safely clustered at different points in a cell; then, one can see a potentially arbitrary number of signals within a single living cell by imaging all the points within the cell with a conventional microscope. Each point reports a different "movie" of the signal being measured by the sensors at that point. The indicator clusters, which we call signaling reporter islands (SiRIs), can be identified post hoc through immunostaining against distinct immunoepitopes fused to each fluorescent reporter. In our earlier work, we performed in vitro imaging of SiRIs to map out relationships between Ca^{2+}, cAMP, and PKA signals; in the current study, we have focused on adapting the use of SiRIs *in vivo* in the mouse brain, key to the analysis of plasticity, learning, behavior and neurological illness. We have now derived protocols for expression, and imaging of SiRIs in awake mice. We first separately expressed SiRI indicators for Ca^{2+} and PKA in layer 2/3 neurons of the mouse visual cortex, and then chronically imaged these neurons in awake mice with two-photon microscopy to observe the activity and stability of SiRIs. We found that the resulting SiRIs displayed both spontaneous activity as well as physiologically evoked responses to visual stimuli (gratings or natural movies) and noradrenergic activation. We also found that SiRIs within single cells were stable over weeks, a critical point for accurate post hoc identification of puncta after chronic experiments. We then co-expressed Ca^{2+} and PKA SiRIs and found that individual SiRIs could be identified and displayed visually evoked activity. These experiments point the way towards the use of SiRIs to map multiple signaling pathways in a single cell simultaneously in a living brain.

Disclosures: N. Shmoel David: None. **C. Linghu:** None. **K.R. Jenks:** None. **K. Tsimring:** None. **M. Shpokayte:** None. **O.T. Celiker:** None. **J.F. Norman:** None. **S. Ramirez:** None. **M. Sur:** None. **E.S. Boyden:** None.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.11

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH R01 6942751
NIH R01 6946116
Bose 2612091
Tom Stocky
NIH 1R01EB024261
Kathleen Octavio
Lore McGovern

Title: 16-fold expansion microscopy (16exm) enables single-shot ultrahigh resolution imaging on conventional microscopes

Authors: *S. WANG[1,2], T. SHIN[1,3], C. ZHANG[1], H. YODER[1], K. LEUNG[1], N. HAN[1], Y. LIU[1], L. L. KIESSLING[2,4], E. S. BOYDEN[1,5];
[1]McGovern Inst. of Brain Res., [2]Dept. of Chem., [3]Media Arts and Sci., MIT, Cambridge, MA; [4]Broad Inst. of MIT and Harvard, Cambridge, MA; [5]HHMI, Cambridge, MA

Abstract: SW and TWS contributed equally to this work. Expansion microscopy (ExM) is in widespread use throughout biology because its isotropic physical magnification enables nanoimaging on conventional microscopes (Science, 347(6221), 543-548), with many hundreds of experimental papers and preprints appearing to date. To date, all methods either expand specimens to a limited range (typically ~4-10x in linear expansion factor, yielding ~30-70 nm resolution; the upper limit in expansion factor is limited by the lower gel density with bigger expansion factors, which in turn results in more fragile expanded samples), or achieve larger expansion factors through iterating the expansion factor over and over (typically ~15-20x in linear expansion factor, yielding ~20 nm resolution). In addition, recent expansion protocols such as expansion revealing (ExR; bioRxiv 2020.08.29.273540), which separate biomolecules from each other before staining, make target biomolecules more available and enable them to be better stained, in some cases converting invisible biomolecules into visible ones. We here present a new expansion protocol that supports post-expansion staining, and achieves ~16x expansion factor, sufficient to enable ~20-nm-resolution imaging on a conventional microscope, in a single expansion step. We show the utility of this protocol, which we call 16ExM, in mapping neural architecture in Thy1-YFP transgenic mice and other preparations and contexts of importance in neuroscience.

Disclosures: **S. Wang:** None. **T. Shin:** None. **C. Zhang:** None. **H. Yoder:** None. **K. Leung:** None. **N. Han:** None. **Y. Liu:** None. **L.L. Kiessling:** None. **E.S. Boyden:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); ESB is an equity holder in a company in the space of commercial applications of expansion microscopy.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.12

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: JSPS KAKENHI 1120626
Leon Levy Fellowship in Neuroscience
Howard Hughes Medical Institute
CHDI Foundation

Title: Multiplexed and scalable cellular phenotyping toward the standardized three-dimensional human neuropathology

Authors: *T. MURAKAMI, N. HEINTZ;
The Rockefeller Univ., The Rockefeller Univ., New York, NY

Abstract: Since the pioneering work of Ramón y Cajal, neurohistology has largely promoted the knowledge over cellular-level pathophysiology of our brains. This trend has recently been accelerated by the advent of three-dimensional histology. By combining mount-staining, tissue-clearing, and light-sheet microscopy, researchers expect to identify cytoarchitectural abnormalities in psychiatric disorders which have been undetectable or difficult to detect in two-dimensional histology. However, the large structural variability of human brains and difficulty in standardizing quantitative metrics in histology left challenges to know whether the histological differences are reflecting the genuine pathological/anatomical heterogeneity or methodological factors. Moreover, the lack of multiplexable and scalable mount-staining methods limit the assessment of selective cell vulnerability of diseases. In this study, we devised a rapid and scalable staining/imaging technique of a brain using multiplexed fluorescence in situ hybridization in 3D (mFISH3D). mFISH3D enables single-cell-resolution imaging of more than ten mRNAs in tissue with the size of a whole-mouse brain. By reconstructing a radial flow of cortical growth in a postmortem human brain, we developed a new approach to assess cellular heterogeneity over cortical tangential space. Using the workflow, biological heterogeneity among regions and individuals can be fairly assessed while minimizing artificial variabilities. Together with mFISH3D, I propose the standardized 3D histology with which the researchers can reproducibly inspect the selective cellular vulnerability in any neuropsychiatric disorders.

Disclosures: T. Murakami: None. **N. Heintz:** None.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.13

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: ERC Grant ADOS 339451
ERC Grant Dyn-Syn-Mem 787340
CIHR Grant 158090

Title: All-optical synaptic physiology in brain slices with Lattice Light Sheet Microscopy

Authors: *A. GETZ[1], M. DUCROS[2], D. CHOQUET[3];
[1]Interdisciplinary Inst. for Neurosci., CNRS, Bordeaux, France; [2]Bordeaux Imaging Ctr., Bordeaux, France; [3]UMR 5297 CNRS Univ. Bordeaux, UMR 5297 CNRS Univ. Bordeaux, Bordeaux Cedex, France

Abstract: The lattice light sheet microscope (LLSM) permits high-resolution imaging with low photobleaching and phototoxicity, which enables high-speed imaging of fast dynamic processes in live tissues. We added a photostimulation module (PSM) to the original Betzig LLSM layout that permits simultaneous imaging and 1- or 2-photon manipulation. Using a knock-in mouse model that allows for target-specific labeling of endogenous GluA2 subunits in intact tissue, we performed fluorescence recovery after photobleaching (FRAP) imaging of AMPA receptor surface diffusion to characterize the mobility endogenous AMPAR at dendritic spines in the *stratum radiatum* of organotypic hippocampal slices. We also used the genetically encoded calcium indicator GCaMP6f to monitor the amplitude of synaptic Ca^{2+} transients induced by 2-photon glutamate uncaging during high-frequency single plane time-lapse acquisitions. With the recently developed optogenetic tool photoactivatable CaMKII (paCaMKII), we used targeted 2-photon stimulation to induce structural plasticity and follow spine volume changes in 4D. Our combination of LLSM and PSM technology enables all-optical synaptic physiology experiments in live tissue preparations. These developments in advanced imaging techniques will allow further detailed study of the molecular mechanisms underlying the expression of synaptic plasticity in integrated neuronal networks with enhanced spatiotemporal resolution.

Disclosures: **A. Getz:** None. **M. Ducros:** None. **D. Choquet:** None.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.14

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: National Honor Scientist Program (NRF-2012R1A3A1050385) through the National Research Foundation of Korea.

Title: Visualization of astrocyte-neuron contact using eGRASP

Authors: *J. KIM, Y. SUNG, H. PARK, D. CHOI, J.-I. KIM, H. LEE, S. YE, B.-K. KAANG; Seoul Natl. Univ., Seoul-si, Korea, Republic of

Abstract: Astrocytes are known to contribute to synaptic functions and plasticity through tripartite synapses. Tripartite synapses are the site of communication among presynaptic, postsynaptic neurons and astrocyte by contact-dependent and diffusible factors. Therefore, identifying tripartite synapses and understanding its dynamics is important to understand the

synaptic functions and astrocytic contribution. However, current techniques to observe tripartite synapses are limited and laborious, hindering the research. Therefore, we developed specialized tool, Astrocyte-eGRASP (enhanced Green Fluorescent Protein Reconstitution Across Synaptic Partners), which can visualize astrocyte-neuron contact by fluorescence. By expressing pre-eGRASP and post-eGRASP in astrocyte and neuron, we could label neuron-astrocyte contact in vitro and in vivo. Astrocyte-eGRASP enabled us to analyze astrocyte-neuron contact in individual astrocyte, identify tripartite synapses in specific neurons, such as excitatory and inhibitory neurons. This study will provide the tool to expand the research on astrocytic participation in synaptic functions.

Disclosures: **J. Kim:** None. **Y. Sung:** None. **H. Park:** None. **D. Choi:** None. **J. Kim:** None. **H. Lee:** None. **S. Ye:** None. **B. Kaang:** None.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.15

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: Samsung Research Funding & Incubation Center for Future Technology (SRFC-IT1702-09)
National Research Foundation of Korea (NRF) (NRF-2021R1C1C1006642)
National Research Foundation of Korea (NRF) (NRF-2021M3A9I4026318)
NIH 1R01EB024261
NIH Director's Pioneer Award 1DP1NS087724
U. S. Army Research Laboratory and the U. S. Army Research Office W911NF1510548

Title: Super-resolution imaging of whole mouse embryos and neurological analysis using whole-body ExM

Authors: *C. PARK[1], J. SIM[1], I. CHO[1], K. MIN[2], M. EOM[1], S. HAN[1], H. JEON[3], H.-J. CHO[4], E.-S. CHO[1], A. KUMAR[1], Y. CHONG[5], J. KANG[6], K. D. PIATKEVICH[7], E. JUNG[8], D.-S. KANG[1], S.-K. KWON[3], J. KIM[3], K.-J. YOON[1], J.-S. LEE[4], E. S. BOYDEN[9], Y.-G. YOON[1], J.-B. CHANG[1];
[1]Korea Advanced Inst. of Sci. and Technol. (KAIST), Daejeon, Korea, Republic of; [2]Sungkyunkwan Univ., Suwon, Korea, Republic of; [3]Korea Inst. of Sci. and Technol. (KIST), Seoul, Korea, Republic of; [4]Korea Res. Inst. of Biosci. and Biotech. (KRIBB), Daejeon, Korea, Republic of; [5]The Catholic Univ. of Korea, Uijeongbu, Korea, Republic of; [6]Harvard Univ., Cambridge, MA; [7]Westlake Univ., Hangzhou, China; [8]MIE, Univ. of Illinois at Chicago, Chicago, IL; [9]MIT, Cambridge, MA

Abstract: Whole-body imaging of a vertebrate is essential for neurological studies. It is critical to observe the neuro-developmental process at super-resolution in order to understand various neurological diseases that occur as a result of abnormalities in fetal neuro-development. Although current technology allows for whole imaging of the fetus, it has a limitation in a nanoscale-resolution imaging. Recently developed whole-body ExM method provides a robust solution for such limitation. Through whole-body ExM, we were able to achieve super-resolution neuron tracing in transgenic mice, as well as visualize innervation and alterations in nerve endings that would have been difficult to discern with conventional imaging setup. Furthermore, we were able to see the differentiation process and alterations in the spinal cord and bone cells that were difficult to see with antibodies, allowing us to study neurological illnesses.

Disclosures: C. Park: None. **J. Sim:** None. **I. Cho:** None. **K. Min:** None. **M. Eom:** None. **S. Han:** None. **H. Jeon:** None. **H. Cho:** None. **E. Cho:** None. **A. Kumar:** None. **Y. Chong:** None. **J. Kang:** None. **K.D. Piatkevich:** None. **E. Jung:** None. **D. Kang:** None. **S. Kwon:** None. **J. Kim:** None. **K. Yoon:** None. **J. Lee:** None. **E.S. Boyden:** None. **Y. Yoon:** None. **J. Chang:** None.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.16

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: National Science Foundation (NeuroNex 1707352)
National Institutes of Health (R01NS120832)

Title: Interluminescence for selective control of synaptically connected pre-and postsynaptic neurons

Authors: *M. PRAKASH[1], M. TREE[1], K. M. RISELAY[1], N. C. SHANER[2], C. I. MOORE[3], D. LIPSCOMBE[4], U. HOCHGESCHWENDER[1];
[1]Central Michigan Univ., Mount Pleasant, MI; [2]Neurosciences, UCSD, La Jolla, CA; [3]Neurosci., [4]Carney Inst. for Brain Sci., Brown Univ., Providence, RI

Abstract: In BioLuminescent OptoGenetics, a genetically encoded light source, a luciferase, activates a light-sensing optogenetic element, a channelrhodopsin or a pump. Here, we leveraged this coupling strategy and developed a tool platform, Interluminescence, for experimental control of synaptic transmission between genetically defined neuronal partners by creating an optical synapse. ‘Interluminescence’ means ‘bioluminescent light in between’, here between a sender cell and a receiver cell. When the sender is a presynaptic neuron expressing luciferase and the receiver is its postsynaptic partner expressing opsin, we essentially create an optical synapse. Upon administration of the luciferase substrate, luciferin, bioluminescence emitted from a presynaptic neuron activates light-sensing opsins in a postsynaptic neuron. We previously

demonstrated the effects of Interluminescence electrophysiologically at postsynaptic population level both *in vitro* and *in vivo*. Here we tested Interluminescence by patch clamp recordings from individual postsynaptic neurons. We tested two different scenarios of optical synapse. First, when the presynaptic neuron expresses luciferase targeted to synaptic vesicles and second, when luciferase is tethered to the presynaptic membrane. The availability of luciferase in the former is dependent on presynaptic activity while the latter has persistent presence of luciferase in the cleft regardless of presynaptic activity. To test these conditions, rat cortex and striatum neurons were nucleofected with a luciferase-dTomato construct and an excitatory opsin-EYFP construct respectively, and were plated on glass coverslips as mixed culture. Synaptic pairs were located by visualizing the reporter expression for the luciferase (dTomato) on presynaptic cortical neurons and for the opsin (EYFP) on postsynaptic striatal neurons at the time of patch. Whole cell patch configuration was achieved on the opsin-expressing postsynaptic neurons and activity was recorded in continuous current clamp mode. Depolarization in postsynaptic neurons was robustly elicited with bioluminescence from presynaptic partners in the presence of synaptic blockers, and was likely due to trans-synaptic communication independent of traditional neurotransmission. We observed significant differences between traditional neurotransmission-induced versus Interluminescence-induced postsynaptic action potentials. The information gained about optical synapses at single neuron level with different luciferase and opsin combinations will be insightful in planning circuit-specific *in vivo* experiments.

Disclosures: M. Prakash: None. **M. Tree:** None. **K.M. Riselay:** None. **N.C. Shaner:** None. **C.I. Moore:** None. **D. Lipscombe:** None. **U. Hochgeschwender:** None.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.17

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NSF-1707316
NIH-1RF1MH120005
NIH-1RF1MH123402

Title: Trans-synaptic labeling and circuit reconstruction with single neuron resolution by light microscopy in the Drosophila brain

Authors: *Y. LI[1], K. I. ATHUKORALA[2], L. A. WALKER[3], D. CAI[1];
[1]Cell and Developmental Biol., [2]Neurosci. Undergraduate Program, [3]Biophysics, Univ. of Michigan, Ann Arbor, MI

Abstract: The precise functions of the nervous system rely on the proper formation of interconnected networks between individual neurons. Identifying related neurons in a circuit, and further resolving the connections between them are critical steps to understanding brain functions

in health and disease conditions. Progress in 3D electron microscopy (EM)-based circuit reconstruction sheds light on revealing the neural network structure with synapse resolution. However, EM's limited data volume acquisition and reconstruction throughput constrained its broader applications to a very small animal pool. Current studies lack quantification in animal-to-animal variations, which is particularly critical in investigating animals with genetic mutations. It is also difficult to retrieve molecular information of the reconstructed neurons, which makes light microscopy a crucial complementary modality for EM.
Recent genetic tools, including GRASP, trans-Tango, and TRACT permit identifying monosynaptic connections in the Drosophila nervous system. In particular, the latter two label the full morphology of connected neurons, which promises the potential of revealing the downstream neurons' identities and their projection patterns as ensembles. It is plausible to hypothesize that if individual neurons' morphology can be reconstructed, the labeled circuits may be mapped to the EM-based connectome for detailed analysis. Furthermore, the identity of the downstream neurons may be revealed by comparing their morphologies to numerous single neuron reconstructions from enhancer-GAL4 labeling using an informatics tool, e.g. NBLAST. However, current trans-synaptic tools use a single color reporter for labeling all downstream neurons, which prohibits the reconstruction of single neuron morphology.
To solve this problem, we created a new set of transgenic Bitbow flies to enable multispectral labeling with trans-Tango. Using sample expansion and a custom-built light microscope, we can image a whole fly brain with sub-hundred-nanometer resolution in a few hours. Combining with computer-aided neuronal tracing, we can reconstruct single neuron morphologies to build circuit consensus in a high-throughput manner. We envision that these new tools will greatly speed up our progress in brain circuit discovery.

Disclosures: **Y. Li:** None. **K.I. Athukorala:** None. **L.A. Walker:** None. **D. Cai:** None.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.18

Topic: H.08. Learning and Memory

Support: R21EY030727
DP2 EB030992
Research Council of Norway (223273, 248828, 283798)
NIH R21 EY029466
R21 EB026180
NSF ECCS-1752241
ECCS-2024776

Title: Functional connectivity revealed between cortical organoids and mouse visual cortex using transparent graphene electrodes and two-photon imaging

Authors: *M. WILSON[1], M. THUNEMANN[3], X. LIU[2], Y. LU[4], F. PUPPO[4], J. ADAMS[4], J.-H. KIM[5], D. P. PIZZO[7], S. DJUROVIC[8], O. A. ANDREASSEN[9], A. MANSOUR[10], F. H. GAGE[11], A. R. MUOTRI[6], A. DEVOR[3], D. KUZUM[1];
[1]Univ. of California San Diego, La Jolla, CA; [2]Univ. of California San Diego, San Diego, CA; [3]Biomed. Engin., Boston Univ., Boston, MA; [4]UC San Diego, La Jolla, CA; [5]Electrical and Computer Engin., [6]Pediatrics/Cellular Mol. Med., UCSD, La Jolla, CA; [7]Univ. of California, San Diego, CA; [8]Univ. of Oslo, Oslo, Norway; [9]Oslo Univ. Hosp. - Ulleval, Oslo, Norway; [11]LOG-G, [10]Salk Inst., La Jolla, CA

Abstract: Cortical organoids are rapidly advancing as models of human brain development and disease and offer promise as neural prosthetics to restore lost or degenerated brain regions. However, due to limitations in longitudinal recording technologies, organoids' beneficial abilities to functionally connect with host cortex upon implantation and respond to external sensory stimulation have yet to be demonstrated. Here, we establish a novel paradigm combining transparent graphene electrode arrays and two-photon microscopy for longitudinal, multimodal monitoring of organoids implanted in mice cortices. Over the course of eleven weeks, we recorded local field potentials and multi-unit activity (MUA) from organoids in response to visual stimulation and during spontaneous activity. Examining the electrophysiological responses to visual stimulation, we found that the evoked responses in organoids matched that of the surrounding cortex, suggesting functional connectivity had formed between organoids and mouse tissue. In further support of functional connectivity, we observed increases in gamma and MUA power and phase locking of MUA to local field potentials following visual stimulation. Two-photon microscopy confirmed functional vascularization of the organoids. Using post-mortem immunohistochemistry, we observed synaptic connectivity and morphological fusion between organoids and host mouse cortices. Our novel, multimodal recording setup revealed the first demonstration of organoids functionally connecting to mouse visual cortical networks and offers a unique platform through which researchers can study the potential of organoids to restore damaged brain regions upon integration.

Disclosures: **M. Wilson:** None. **M. Thunemann:** None. **X. Liu:** None. **Y. Lu:** None. **F. Puppo:** None. **J. Adams:** None. **J. Kim:** None. **D.P. Pizzo:** None. **S. Djurovic:** None. **O.A. Andreassen:** None. **A. Mansour:** None. **F.H. Gage:** None. **A.R. Muotri:** Other; Co-founder and has equity interest in TISMOO. Terms have been reviewed and approved by UCSD in accordance with its conflict of interest policies.. **A. Devor:** None. **D. Kuzum:** None.

Poster

576. Visualizaing Neural Connectivity With High Resolution

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 576.19

Topic: I.03. Anatomical Methods

Support: FWF der Wissenschaftsfonds
MolTag Molecular Drug Targets
EU H2020 Marie Skłodowska-Curie

Title: A versatile toolbox for the comprehensive analysis of nervous tissue organization with light microscopy

Authors: *J. M. MICHALSKA[1], J. LYUDCHIK[1], P. VELICKY[1], H. KORINKOVA[1], J. F. WATSON[1], A. CENAMERI[1], C. SOMMER[1], A. VENTURINO[3], K. ROESSLER[4], T. CZECH[4], S. SIEGERT[5], G. NOVARINO[1], P. JONAS[2], J. G. DANZL[1];
[2]Neurosci., [1]Inst. of Sci. and Technol. Austria, Klosterneuburg, Austria; [3]Inst. of Sci. and Technol. IST Austria, Klosterneuburg, Austria; [4]Dept. of Neurosurg., Med. Univ. of Vienna, Vienna, Austria; [5]Insitute of Sci. and Technol. Austria, Klosterneuburg, Austria

Abstract: The brain is an exceptionally sophisticated organ consisting of billions of cells and trillions of connections that orchestrate our cognition and behavior. To decode its complex connectivity, it is pivotal to disentangle its intricate architecture spanning from cm-sized circuits down to tens of nm-small synapses.To achieve this goal, we have developed CATS - Comprehensive Analysis of nervous Tissue across Scales - a toolbox for obtaining a holistic view of nervous tissue context with fluorescence microscopy. CATS provides rich ultrastructural context by creating contrast between the intra- and extracellular space in a variety of samples types, including slice cultures, perfused brain tissue and clinical samples. It is compatible with common (super-resolution) fluorescence imaging techniques, such as stimulated emission depletion (STED) and expansion microscopy, as well as labeling of molecular markers. We interface this toolbox with segmentation of cellular architecture and a state-of-a-art machine-learning based analysis pipeline for annotation of synaptic transmission sites.
CATS enables the analysis of key features of nervous tissue connectivity across scales, ranging from whole tissue organization down to synapse architecture. We present the potential of this novel toolbox by mapping the local synaptic input structure of a CA3 pyramidal neuron, as well as the synaptic output structure of a DG granule cell - a feat that has thus far only been achieved by electron microscopy. To characterize the DG granule cell-CA3 pyramidal neuron synapse in more depth, we reconstruct hippocampal mossy fiber boutons with their synaptic transmission topology in near-natively preserved brain.Further, we combine CATS with electrophysiological recordings to enable investigating structure-function relationships in the brain, apply it to human clinical samples to study tissue context and fully annotate a piece of cerebral organoid, thereby paving the way towards light microscopy-based saturated reconstruction of nervous tissue.

Disclosures: J.M. Michalska: None. **J. Lyudchik:** None. **P. Velicky:** None. **H. Korinkova:** None. **J.F. Watson:** None. **A. Cenameri:** None. **C. Sommer:** None. **A. Venturino:** None. **K. Roessler:** None. **T. Czech:** None. **S. Siegert:** None. **G. Novarino:** None. **P. Jonas:** None. **J.G. Danzl:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.01

Topic: I.04. Physiological Methods

Title: Developing an Implantable Device to Monitor Optical Neuronal Activity

Authors: *T. BEN ULIEL[1,2], Y. LIN[3], G.-L. ALBANO[4], S. POZZI[1,2], R. PLEN[1,2], E. SEGAL[3], O. SHEFI[1,2];
[1]Fac. of Engin., Bar-Ilan Univ., Ramat Gan, Israel; [2]Bar-Ilan Inst. of Nanotechnologies and Advanced Materials, Ramat Gan, Israel; [3]Dept. of Biotech. and Food Engin., Technion - Israel Inst. of Technol., Haifa, Israel; [4]Tech. Univ. of Braunschweig, Braunschweig, Germany

Abstract: The ability to record neuronal activity over time is of great importance in research and therapy. The conventional electrophysiological recording methods of cellular activity require wiring, hence enabling only a limited number of recording sites. In addition, they are relatively harmful to the cells, restricting the recording time duration. A comprehensive approach for neuronal detection that is based on optical detection may overcome these challenges. Lately, calcium imaging of fluorescent calcium indicators has emerged as a dominant alternative for neural recording. The technique measures fluctuations in intracellular calcium concentrations, offering an indirect indication of neural activity. To date, there are genetic and chemical calcium indicators that offer tradeoffs; however, chemical indicators are advantageous due to sensitivity, light throughput, signal-to-noise ratio, and their dynamic range. However, chemical calcium indicators are limited by their rapid extrusion from cells, which prevents prolonged monitoring in neuronal populations. We develop novel responsive platforms for wireless and noninvasive long-term monitoring of neuronal activity and viability via calcium signals. Our platform is based on a silicon nanostructured chip - porous silicon (PSi) - which is biocompatible and biodegradable. In the experiment, we absorb calcium indicators into the PSi, which are spontaneously released from it. Release and calcium imaging experiments were conducted with mice cortical neurons. We have shown that the platforms release the indicators with diverse durations and doses, from days to weeks, based on the chemical functionalization we are performing. Our PSi device is used as a reservoir of calcium indicators for prolonged optical detection of neuronal activity by calcium imaging. The ability to control the release kinetic offers the device usage for monitoring neuronal activity under varying conditions. Furthermore, to enable pulsatile and a release based on the neuronal pathology, we are currently developing an additional layer that can be triggered by external stimulation and act in a biofeedback manner.

Disclosures: **T. Ben Uliel:** Other; the council of higher education. **Y. Lin:** None. **G. Albano:** None. **S. Pozzi:** None. **R. Plen:** None. **E. Segal:** None. **O. Shefi:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.02

Topic: I.04. Physiological Methods

Support: ONR N00014161253
ONR N000142012405
NSF ECCS-2024776
NSF ECCS-1752241
NIH R21 EY029466
NIH R21 EB026180
NIH DP2 EB030992

Title: Predicting Calcium Activity at Depth from Surface Potentials Recorded by High-Density Transparent Graphene Arrays

Authors: *M. RAMEZANI[1], J.-H. KIM[1], C. REN[3], X. LIU[1], M. WILSON[1], T. KOMIYAMA[2], D. KUZUM[1];
[2]Ctr. for Neural Circuits and Behavior Rm 304, [1]Univ. of California San Diego, La Jolla, CA; [3]Div. of Biol. Sci., UCSD, San Diego, CA

Abstract: Optically transparent neural microelectrodes have been widely used in multimodal experiments to record the brain's electrophysiological activity and simultaneously perform optical imaging or stimulation. Here, we fabricated a high-density transparent graphene array and implanted it over the visual cortex of a transgenic mouse expressing GCaMP6s in most cortical excitatory neurons. We recorded surface potentials from the 64 channels of the graphene array and used two-photon microscopy to image neurons underneath the transparent microelectrodes at two different depths, 50 and 225 μm, corresponding to layer 1 and layer 2/3, respectively. The animal was presented with drifting gratings (8 directions) as visual stimulation. The field of view (FoV) for calcium imaging was 960 μm × 960 μm and covered nine electrodes. By examining the visually evoked signals, we found that the high-frequency bands are spatially localized to the source of the response (V1) while low-frequency bands propagate further to PPC, S1, and RSC. We also found that the MUA power of channels in the FoV is highly correlated with the average calcium activity of L2/3 neurons. Given this association between the MUA and the cellular calcium signals, we constructed a simple neural network model to predict brain activity at deep layers using surface potentials. The model successfully predicted the averaged calcium activity of L1 and L2/3 using the signal powers at different frequency bands (δ, θ, α, β, γ, H-γ, and MUA) as input features. Next, we investigated the contribution of different channels and frequency bands to the prediction performance. We found that different channels provide complementary information, and high-frequency bands (γ, H-γ, and MUA) are more informative than low-frequency bands (δ, θ, α, β). Our high-density, fully transparent graphene electrode enables us to study cortical dynamics in greater detail.

Disclosures: M. Ramezani: None. **J. Kim:** None. **C. Ren:** None. **X. Liu:** None. **M. Wilson:** None. **T. Komiyama:** None. **D. Kuzum:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.03

Topic: I.04. Physiological Methods

Support: BBRF NARSAD Young Investigator Award
NIMH R01 MH124695-01

Title: A case of spurious correlation: Pitfalls of normalizing dual-color $GRAB_{DA}$ and RCaMP fiber photometry by a 410 nm isosbestic wavelength

Authors: *M. JANECEK, R. PEIXOTO;
Dept. of Psychiatry, Ctr. for Neurosci., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Fiber photometry measures bulk fluorescence signal of neural activity and neurotransmitter dynamics even in deep brain tissue. However, when used in freely moving animals, optic fiber and animal motion can generate artifacts that confound activity-dependent changes in fluorescence. As a solution, many photometry protocols include a near-ultraviolet isosbestic wavelength excitation (405-415 nm) in response to which green fluorescent protein (GFP)-based sensors exhibit negligible changes in activity-dependent fluorescence. An isosbestic wavelength channel should therefore contain only fluorescence unrelated to sensor activity, serving as a control for GFP-based calcium and neurotransmitter sensors, such as the dopamine 2 receptor-based $GRAB_{DA2m.}$ Since isosbestic excitation causes photobleaching over time, it can de-trend the $GRAB_{DA2m}$ fluorescence activity trace and provide a baseline (*F0*) for calculating relative change in fluorescence. Several protocols have further extended the use of a near-ultraviolet isosbestic wavelength to normalize signals of red-shifted sensors, such as jRCaMP1b. However, applying GFP-specific isosbestic excitation to red-shifted sensors requires further validation. Here, we controlled both $GRAB_{DA2m}$ and jRCaMP1b fluorescence using a 410 nm isosbestic channel, recorded concurrently in the striatum of freely moving mice. To normalize both signals, we linearly fitted isosbestic channel fluorescence against each sensor to obtain a scaled *F0*. Unexpectedly, the isosbestic channel proved sensitive to $GRAB_{DA2m}$ activity, and, critically, isosbestic normalization of fluorescence channel sampling jRCaMP1b introduced spurious transients into the red-shifted channel in animals only expressing $GRAB_{DA2m.}$ Finally, spurious noise introduced by isosbestic normalization inflated the dual-channel correlation of relative signal up to 6-fold in animals lacking any sensor. Therefore, correcting dual-color photometry using a 410 nm isosbestic control could impede interpretation of neural activity or, at worst, conjure a relationship that is not of biological origin. We conclude by presenting an alternative analysis pipeline and a discussion of a theoretical isosbestic wavelength tailored for red-shifted sensors.

Disclosures: M. Janecek: None. **R. Peixoto:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.04

Topic: I.04. Physiological Methods
MH115215 (AD)
MH111703 (VPF)

Title: A genetically encoded sensor reveals the dynamics of cholinergic signaling during goal-directed behavior in non-human primates.

Authors: *M. BOMPOLAKI[1,2], F. A. MUNOZ[3], A. DRANOVSKY[2,1], V. P. FERRERA[3]; [1]Systems Neurosci., New York State Psychiatric Inst., New York, NY; [2]Psychiatry, [3]Neurosci., Columbia Univ., New York, NY

Abstract: The use of genetic tools to study circuit function in non-human primates has been technically challenging due to often low and transient transgene expression. Genetically encoded fluorescent sensors of neurotransmitter activity offer an opportunity to tamper the impact of these obstacles. Specifically, measuring fluctuations in local neurotransmitter release can provide valuable information about neuromodulation without the need for directing reporter expression to specific cells of interest or requiring single cell resolution for signal analysis. Thus, sparser viral transduction is more likely to lead to informative results. Here, we describe the use of GRAB-ACh3.0 to examine the effect of cholinergic modulation on signal detection in V1 of a rhesus macaque. The cholinergic system is known to orchestrate task engagement. However, we lack a detailed understanding of how transient shifts in performance may be influenced by rapid changes of acetylcholine (ACh) levels. We used fiber photometry to measure GRAB-ACh3.0 activity with simultaneous single cell electrophysiology, pupillometry, and eye tracking while the animal was engaged in a smooth pursuit eye movement task. This study was designed to test the hypothesis that fluctuations in task-engagement correlate with rapid changes in ACh levels. Systemic acetylcholine is thought to be associated with changes in pupil size and smooth pursuit gain. Our initial studies revealed that ACh signal was significantly increased during on-task compared to off-task trials, supporting the notion that momentary lapses of attention occur during periods of low ACh levels and therefore cholinergic compensation could improve performance. To our knowledge, this is the first evidence of rapid neurotransmitter changes correlating with moment-to-moment fluctuations in behavioral output in primates. These results have important implications for understanding the neurochemical underpinnings of sustained attention and goal-directed performance and define a pharmaceutically targetable substrate to manage conditions like attention deficit disorder.

Disclosures: M. Bompolaki: None. **F.A. Munoz:** None. **A. Dranovsky:** None. **V.P. Ferrera:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.05

Topic: I.04. Physiological Methods

Support: NIH Intramural Funding

Title: Systematic comparison of three in vivo fiber photometry recording methods

Authors: *K. D. STEVANOVIC, L. R. WILSON, A. C. LETSINGER, J. A. CUSHMAN; Neurobio. Labs., Natl. Inst. of Envrn. Hlth. Sci., Research Triangle Park, NC

Abstract: Systematic comparison of three in vivo fiber photometry recording methods
Stevanovic, K., Wilson, L., Letsinger, L., Cushman, J.
The use of *in vivo* fiber photometry has rapidly expanded in recent years as technical advances in commercially available and custom-built recording systems have made it more accessible. Increasingly sophisticated fluorescent sensors combined with viral and transgenic targeting strategies have made photometry an indispensable tool for connecting behavior to the neural circuitry that underlies it. Multiple different photometry recording approaches have been developed, with each approach likely to have specific strengths, weaknesses, and caveats. It is also not clear whether these different approaches produce quantitatively or qualitatively similar results, which makes comparison across studies difficult. In order to systematically investigate this we conducted a comparison of the three most commonly used approaches: a spectrally-resolved fiber photometry system, a camera-based multi-wavelength photometry system and a lock-in demodulation photometry system. We systematically rotated three groups of male C57Bl6/J mice through each photometry system in a counterbalanced manner and recorded a stimulus evoked GCaMP response in the locus coeruleus (LC). The presentation of an LED light elicits a strong and consistent transient response in the LC, which allowed for a systematic comparison of each system. Here, we summarize the data processing pipeline for each system and systematically compare the results. These findings provide important quantitative comparisons that will aid in interpretation across studies utilizing these different methods and help future researchers determine the best *in vivo* fiber photometry system to use for their specific experimental needs.

Disclosures: K.D. Stevanovic: None. **L.R. Wilson:** None. **A.C. Letsinger:** None. **J.A. Cushman:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.06

Topic: I.04. Physiological Methods

Support: U01-NS103518
U01-NS122123

Title: Multi-regional functional two photon calcium imaging in an awake behaving rhesus macaque

Authors: *K. WINGEL[1], J. CHOI[1], J. HAGERTY[1], M. CHOUDHURY[1], A. CHARLES[2], H. HAFIZI[1], A. DUBEY[1], R. BAKHSHI[1], B. PESARAN[1];
[1]New York Univ. Ctr. For Neural Sci., new york, NY; [2]Johns Hopkins, Baltimore, MD

Abstract: In the non-human primate (NHP) brain, multiregional communication between cortical areas supports sensation, movement, attention, thought, and other distributed brain functions. As a result, when studying brain function, it is important to think of different cortical regions not as being involved in individual processes, but as a multi-regional network. A major challenge involves gaining access across brain areas to perform multiregional two photon (2P) imaging. Here, we performed functional 2P imaging using a robotic microscope to study the arm movement network in motor (M1), premotor dorsal (PMd), and premotor ventral (PMv) in an awake rhesus macaque (*Macaca mulatta*). We engineered and implanted a 2x4 cm subdural craniotomy durotomy window. Three anatomical markers were identified in the vasculature as a roadmap to navigate the cortical window. This allows for multiregional 2P imaging across an entire hemisphere comprising prefrontal cortex (PFC) to posterior parietal cortex (PPC). To enable 2P imaging of neural activity, expression of a genetically-encoded calcium indicator, GCaMP, was virally-induced. Four rounds of viral injections were made with the first being under anesthesia via stereotax protocol. The subsequent injections were performed awake with the use of a custom Convection Enhanced Delivery (CED) microdrive. The virus pgP-AAV-syn-jGCaMP8m-WPRE was injected in the PMd, M1, and PFC. Tandem dimer tomato (tdtomato) a nonfunctional vector was placed in PFC, PMd, and PMV to label histological sites concurrently. GCaMP expression was initially observed between 4-8 weeks following injection and subsequently observed for at least 5 months. During imaging sessions, the macaque performed a center-out reach from a central start position to one of seven peripheral targets. Reaching behavior was measured and processed by a DeepLabCut pipeline. Deeplabcut was trained for object recognition and semantic segmentation to allow the trained algorithm to detect the onset and trajectory of each reach. 2P imaging demonstrated functional cellular responses around the start of the reaching movement in areas M1, PMd, and PMV up to a depth of about 400 um. We will discuss future implications of this capability to perform functional multiregional imaging including the simultaneous use of bidirectional electrophysiology devices for stimulating and recording neural activity. This work demonstrates the use of multiregional cellular imaging in the NHP to better understand the circuitry of the brain through behavior.

Disclosures: K. Wingel: None. **J. Choi:** None. **J. Hagerty:** None. **M. Choudhury:** None. **A. Charles:** None. **H. Hafizi:** None. **A. Dubey:** None. **R. Bakhshi:** None. **B. Pesaran:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.07

Topic: I.04. Physiological Methods

Support: R01NS078168

Title: Understanding the nature and drivers of brain motion in behaving mice

Authors: *C. S. GARBORG[1], Q. ZHANG[2], K. L. TURNER[1], Y.-J. KIM[1], F. COSTANZO[2], P. J. DREW[2];
[1]Dept. of Biomed. Engin., [2]Dept. of Engin. Sci. and Mechanics, The Pennsylvania State Univ., University Park, PA

Abstract: The brain shifts within the skull during behavior. However, the drivers and physiological impact of this motion are not known. It is important to understand the origins and nature of this movement as it is a ubiquitous confound for neurological imaging in both humans and animals. This motion may also help remove metabolic waste from the brain by displacing and mixing cerebrospinal fluid (CSF) as the brain shifts within the skull. The nature and drivers of brain motion are surprisingly not well understood given the relevance it has to many aspects of brain function and neuroscience as a whole. We quantified brain motion in head-fixed awake mice using high-speed, multi-plane two-photon microscopy during free locomotion on a spherical treadmill. Electromyography (EMG) electrodes were implanted into the oblique abdominal muscles to quantify muscle activity, as well as heartbeat and respiratory activity. We found significant abdominal muscle contraction and robust rostral displacement of the brain during voluntary locomotion, along with smaller lateral shifts. These movements are highly correlated with abdominal contractions and poorly coupled to respiratory and cardiac pulsations. Our results suggest that abdominal muscle contractions are responsible in part for brain motion within the skull. This may be explained by abdominal muscle activity causing abdominal pressure increases that are transmitted to the spinal cord and then on into the brain.

Disclosures: C.S. Garborg: None. **Q. Zhang:** None. **K.L. Turner:** None. **Y. Kim:** None. **F. Costanzo:** None. **P.J. Drew:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.08

Topic: I.04. Physiological Methods

Title: In vivo evaluation of the temperature rise induced locally by deep brain illumination using MR-thermometry.

Authors: J. MOLET, P. BLEUET, C. CHABROL, T. COSTECALDE, N. AUBERT, N. TORRES, V. AUBOIROUX;
Univ. Grenoble Alpes, CEA, LETI, Clinatec, Grenoble, France

Abstract: Rationale: Several studies have highlighted the potential effect of light therapy applied through the skull to the brain. Photobiomodulation (PBM) using red to near infrared (NIR) light seems to be a promising treatment for various pathologies and neurodegenerative diseases such as Parkinson's disease. This new therapeutic approach leads to the development of implanted brain optical fiber devices. However, light can generate heat, which increases tissue temperature (TEMP) and might be able to damage tissues. Therefore, device characterization requires a precise evaluation of the TEMP at the implantation site. High resolution MR-thermometry allows quantification of thermal variations in animal brain with large spatial coverage compared to thermocouple-based measurements. Similar to studies performed for laser-induced thermotherapy (LITT), we present a high spatial resolution, high precision characterization of the thermal effects, in order to assess the safety of active implanted optical fiber devices. The objective was to determine optical powers compatible with a TEMP increase of less than 2°C (Requirements for active implantable medical devices EN 45502-1_17.1.). **Methods:** We tested in rat and monkey brains the 670 nm wavelength using by a device specifically designed for deep brain PBM delivery based on optical fibers and in rat brain the 960 nm wavelength commonly used for example in Optogenetics. Animals were anesthetized and stereotaxically implanted with one optical fiber per hemisphere into regions of interest. Increasing light power at the fiber tip has been assessed using proton resonance frequency shift (PRFS)-based MR-thermometry (4.7 T Bruker imager). High-resolution (0.3 x 0.3 x 0.1 mm^3) thermal maps has been acquired during an *in vivo* illumination in rat and monkey brains. **Results:** We evaluated, in continuous illumination mode, the TEMP increase induced by optical powers ranging from 5 to 100 mW at the end of an optical fiber implanted in different brain regions. Our data revealed neither brain region effect nor species effect but a significant main effect of optical power. At 670 nm, an optical power greater than 30 mW is required to induce a TEMP rise of 2°C, whereas greater than 66 mW is necessary at 960 nm. Interestingly, the heat spot at the fiber tip showed a Full Width at Half Maximum (FWHM) of about 2.4 mm and 3.9 mm for 670 nm and 960 nm respectively. **Conclusions:** MR-thermometry is an adequate method to accurately measure *in vivo* deep brain thermal variations and allows thermal characterization of chronic implantable medical devices. The heat increase pattern was correlated both to the light power delivered and to the wavelength.

Disclosures: J. Molet: None. **P. Bleuet:** None. **C. Chabrol:** None. **T. Costecalde:** None. **N. Aubert:** None. **N. Torres:** None. **V. Auboiroux:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.09

Topic: I.04. Physiological Methods

Support: NSERC CGS D- 534171- 2019
FRQNT- 275075
NSERC DG 2017-04730

CIHR PG 156223
Healthy Brains for Healthy Lives
McGill University Max E Binz Fellowship

Title: 2-photon optogenetic mapping of visual cortex microcircuits

Authors: *C. Y. C. CHOU[1], H.-W. WONG[2], K. E. BOUKOULOU[1], T. A. LIANG[1], C. GUO[1], P. J. SJOSTROM[3];
[1]Ctr. for Res. in Neurosci., The Res. Inst. of the McGill Univ. Hlth. Ctr., Montreal, QC, Canada; [2]Dept. of Medicine, Dept. of Neurol. and Neurosurg., [3]Dept of Med., McGill Univ., Montreal, QC, Canada

Abstract: Neuronal connectivity determines information processing in local cortical circuits. For example, in primary visual cortex (V1) layer 2/3 (L2/3), pyramidal cells (PCs) that respond to similar visual stimuli connect more frequently. Inhibitory interneurons (INs) play a key role in maintaining stability and sensitivity in neuronal circuits and have been shown to follow specific connectivity patterns and synaptic dynamics according to cell type. Yet cell-type-specific IN connectivity remains poorly explored. This may be due to the experimental challenges inherent in probing cell-type-specific connectivity. Multiple patch clamp is the state-of-the-art technique, but it is technically challenging and yields only a few connections per day. Therefore, we developed optomapping, a high-throughput connectivity mapping method that combines 2-photon (2P) optogenetics and patch-clamp electrophysiology.
We targeted expression of the soma-targeted opsin ChroME to PCs by injecting AAV9-CAG-DIO-STChroME-P2A-mRuby in neonatal Emx1-Cre mice. In P18-P25 acute slices, we spiral scanned ChroME-expressing PCs in V1 with 1040-nm Ti:Sa laser to elicit action potentials with single-cell spatial and millisecond temporal resolution. By spiral-scanning hundreds of candidate input PCs while whole-cell recording from PCs, basket cells (BCs), or Martinotti cells (MCs), we were able to, in a single experiment, record dozens of laser-evoked EPSPs from input cells hundreds of microns away, across all cortical layers. We verified cell type of the patched neuron by spike pattern and morphology.
Within 100 µm, L5 PC-PC (17/196 = 9%; 0.51 ± 0.09 mV, n = 6 cells) and PC-BC connectivity 85/206 = 41.3%; 2.30 ± 0.42 mV, n = 6 cells) qualitatively matched paired recordings (7%; 0.87 ± 0.09 mV, n = 162 resp.; 100/299 = 33%; 2.10 ± 0.29 mV, n = 100), thus validating optomapping. We next mapped excitatory inputs onto PCs, BCs, and MCs in layers 2, 3, 5, and 6 of V1, which revealed connectivity rates as well as synaptic dynamics, e.g., L2/3 BCs received stronger local
excitation (54/233 = 23.2%; 1.13 ± 0.19 mV, n = 6 cells) than did L2/3 PCs (66/356 = 18.5%; 0.46 ± 0.07 mV, n = 8 cells), which likely ensures stability. We could also verify the canonical circuit, and additionally show e.g., that BCs receive cross-layer excitation and that short-term dynamics of excitatory inputs onto MCs depends on the layer of origin.
In conclusion, optomapping enables large-scale connectivity mapping to reveal principles of local connectivity, e.g., we verified the V1 canonical circuit but could additionally reveal strong, long-range excitation of BCs.

Disclosures: C.Y.C. Chou: None. **H. Wong:** None. **K.E. Boukoulou:** None. **T.A. Liang:** None. **C. Guo:** None. **P.J. Sjostrom:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.10

Topic: I.04. Physiological Methods

Support: AFSOR FA9550-20-1-0061

Title: Evaluation of spontaneous activity in adrenal chromaffin cells in slices derived from mice expressing genetically-encoded calcium indicators

Authors: N. M. PROCACCI[1], T. W. GOULD[2];
[1]Dept. of Pharmacol., [2]Dept. of Physiol. and Cell Biol., Univ. of Nevada, Reno, Reno, NV

Abstract: Adrenal chromaffin cells (ACC) are excitable neuroendocrine cells that secrete catecholamines into the blood in times of stress, triggering the "fight or flight" response. The ability to measure and modulate this response could ameliorate health conditions in which it is dysfunctional, as well as enhance physical and cognitive performance. Dissociated ACC, as well as ACC in acutely prepared slices of adrenal gland, exhibit spontaneously generated action potentials mediated by the activities of Na^+, Ca^+ and K^+ channels. Whether all ACC in slices exhibit such spontaneous activity, and whether it is caused by transmitter released from distal ends of the sympathetic splanchnic nerve, has not rigorously been examined. To address these questions, we took advantage of *Sox10-Cre* and tyrosine hydroxylase *(TH)-GCaMP6f* mice to drive the expression of the genetically-encoded calcium indicator GCaMP6f in mouse ACC. The principal advantage of this technique is the simultaneous capture of the activity of large populations of ACC. We used male and female 2-4 month old mice. The morphology of the clusters in which ACC reside was more distinct in slices imaged at low magnification (20X) from *Sox10-GCaMP6f* mice. In these samples, large ~0.17 Hz waves of calcium activity spread between clusters in sequential patterns that occasionally reversed direction. In contrast, owing to incomplete recombination and/or recombination exclusively in ACC and not satellite glial cells, the morphology and activity patterns of individual ACC were clearer in slices imaged at high magnification (60X) in *TH-GCaMP6f* mice. The frequency of this activity showed no difference between sexes, but varied between slices even from the same mouse on the same day, ranging between 0.2 and 0.9 Hz. To minimize variability, experiments focused on the effects of a) time of tissue incubation prior to imaging b) temperature during imaging and c) duration and number of imaging sessions. We ran 3-way ANOVAs comparing incubation time (0.5 hours vs 2.5 hours) X temperature (23°C X 35°C) X session duration (every 15 minutes for 2 hours). While the amount of incubation time prior to imaging was not significant, frequencies were consistently higher and more variable when recorded at 23°C vs. 35°C (0.62 vs. 0.38 Hz; $F(1) = 1245$, $p < 0.0001$). Frequencies at 23°C were reduced in the second hour of imaging, whereas those at 35°C remained similar $F(7) = 31.49$, $p < 0.0001$. Future studies will assess the nature of these spontaneous activity patterns (i.e., bursting vs. tonic), their dependence on splanchnic nerve input, the underlying molecular mechanisms, and the ability to modulate this activity by electrical stimulation.

Disclosures: **N.M. Procacci:** None. **T.W. Gould:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.11

Topic: I.04. Physiological Methods

Support: CIHR FDN-143209

Title: Dual brain cortical calcium imaging reveals interaction-specific correlated activity in mice.

Authors: *N. MICHELSON, F. BOLANOS, L. BOLANOS, M. BALBI, J. LEDUE, T. MURPHY;
Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Social behavior involves many circuits throughout the brain. Widefield cortical calcium imaging provides an opportunity to observe neural activity across the entire dorsal cortex in vivo, but its application to social neuroscience is largely unexplored. We employed widefield cortical calcium-imaging to observe brain activity in two head-fixed mice during a staged social touch-like interaction. Imaging interactions in head-restrained mice controls the timing and duration of interaction events, and limits behaviors that may impede neuronal data acquisition. Mice are brought together using a motorized rail system until the whiskers of each mouse can make contact. Mesoscale cortical calcium signals were acquired from both mice before, during, and after the social contact period. During the social contact period, bouts of mutual whisking and inter-animal correlated cortical activity across cortex were observed. Correlations were not observed after trial-shuffling mouse pairs, suggesting that correlated activity was specific to individual interactions. Whisking-related GCAMP6s signals during the social contact period were observed in vibrissae cortex. We present an open-source platform to investigate the dorsal cortical neurobiology of social interaction, including mechanical drawings and software for constructing cost-effective Raspberry-pi-based imaging systems. Extension of this head-fixed interaction paradigm to other imaging modalities such as fiber photometry or 2-photon imaging can provide further insight into the physiology underlying social interaction.

Disclosures: **N. Michelson:** None. **F. Bolanos:** None. **L. Bolanos:** None. **M. Balbi:** None. **J. Ledue:** None. **T. Murphy:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.12

Topic: I.04. Physiological Methods

Support: Leon Levy Fellowship in Neuroscience
BBRF Young Investigator Grant
Rockefeller Kavli Neural Systems Institute Pilot Grant
NIH NEI R21 Grant EY031486

Title: Calcium imaging of marmoset cortical neurons toward examining face-selective cells

Authors: *D. G. C. HILDEBRAND[1], S. OTERO CORONEL[1], J. DEMAS[1], S. GRØDEM[2], K. K. LENSJØ[2], G. H. VATNE[2], B. CHEN[1], F. TEJERA[1], S. WEISENBURGER[1], M. FYHN[3], A. VAZIRI[1], W. A. FREIWALD[1];
[1]The Rockefeller Univ., New York, NY; [2]Univ. of Oslo, Oslo, Norway; [3]Dept. of Biosci., Oslo, Norway

Abstract: A major challenge to uncovering the computations and circuits that enable face perception is the disparity in the scales at which the face-processing system has been studied. We know a great deal about the broad properties of face areas from fMRI and individual 'face cell' tuning properties from single-cell electrophysiology. However, it remains difficult to investigate how populations of face cells work together to represent faces without simultaneous activity measurements from many individual face cells. To fill this gap, we are developing approaches to record calcium dynamics from populations of face cells within the posterior dorsal (PD) face area of awake, head-restrained marmosets using two-photon microscopy. Toward this goal, we have localized PD with intrinsic signal optical imaging, optimized imaging chambers, and performed preliminary experiments using awake calcium imaging. We have also tested different virus delivery systems and calcium indicators, including ribosome-linked indicators for soma targeting that reduce background and signal cross-contamination. In the near future, we aim to combine these developments capture dynamics from large populations of face cells.

Disclosures: D.G.C. Hildebrand: None. **S. Otero Coronel:** None. **J. Demas:** None. **S. Grødem:** None. **K.K. Lensjø:** None. **G.H. Vatne:** None. **B. Chen:** None. **F. Tejera:** None. **S. Weisenburger:** None. **M. Fyhn:** None. **A. Vaziri:** None. **W.A. Freiwald:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.13

Topic: I.04. Physiological Methods

Support: Lundbeck foundation R266-2017-4331
Lundbeck foundation R276-2018-792
Lundbeck foundation R230-2016-3154

Lundbeck foundation R181-2014-3090
Lundbeck foundation R231-2016-2481-5
Independent Research Fund Denmark – 7016-00325B

Title: Behavioral encoding across timescales by region-specific dopamine dynamics

Authors: *S. H. JØRGENSEN[1], A. L. EJDRUP[1], M. D. LYCAS[2], L. P. POSSELT[4], K. L. MADSEN[5], L. TIAN[6], J. K. DREYER[7], F. HERBORG[1], A. T. SÖRENSEN[8], U. GETHER[3]; [1]Dept. of Neurosci., Univ. of Copenhagen, Copenhagen N, Denmark; [2]Dept. of Neurosci. and Pharmacol., Univ. of Copenhagen, Cobenhagen N, Denmark; [3]Univ. of Copenhagen, Copenhagen N, Denmark; [4]Dept. of Neurosci., Univ. of Copenhagen, Dept. of Neurosci., Copenhagen N, Denmark; [5]Dept. of Neurosci., Univ. of Copenhagen, Fac. of Hlth. and Me, Copenhagen, Denmark; [6]Biochem. and Mol. Med., Univ. of California, Davis, Davis, CA; [7]Dept. of Bioinformatics, H Lundbeck A/S, Valby, Denmark; [8]Ctr. for Neurosci., Copenhagen Univ., Kobenhavn N, Denmark

Abstract: Behavioral encoding across timescales by region-specific dopamine dynamicsSøren H. Jørgensen[1,*], Aske L. Ejdrup[1,*], Matthew D. Lycas[1] , Leonie P. Posselt[1] , Kenneth L. Madsen[1] , Lin Tian[2] , Jakob K. Dreyer[3] , Freja Herborg[1] , Andreas T. Sørensen[1] and Ulrik Gether[1]
[1]Department of Neuroscience, Faculty of Health and Medical Sciences, Maersk Tower 7.5, University of Copenhagen, DK-2200 Copenhagen, Denmark [2]Center for Neuroscience, School of Medicine, UC Davis, USA [3]Department of Bioinformatics, H Lundbeck A/S, Valby, Denmark. *These authors contributed equally to the work
The dorsal (DS) and ventral striatum (VS) receive dense dopaminergic projections controlling motor functions and reward-related behavior. However, it remains poorly understood how dopamine release dynamics is coupled in these regions across temporal scales to modulation of behavioral outcomes. Here, we address this question by probing extracellular dopamine dynamics in freely moving mice concomitantly in DS and VS using the genetically encoded dopamine sensor dLight1.3b. Our data reveal highly different dopamine dynamics in the two regions with a rapidly fluctuating signal in the DS, carrying information across dopamine levels, and a much slower fluctuating signal in the VS, consisting mainly of slow-paced transients. Using machine learning, we correlate these dynamics to behavioral syllables across timescales and observe striking coordination of the dopamine signals between the two regions during exploratory behavior. Disruption of dopamine dynamics with cocaine imposes drastic changes that leads to randomization of action selection sequencing and disturbance of DS-VS coordination. The data indicate that coordinated, distinct dopamine dynamics of DS and VS, on a sub-second to minutes timescale, orchestrate behavioral sequences where the DS signal modulates the stringing together of actions and the VS signal provides the motivation to initiate and sustain the selected action.

Disclosures: S.H. Jørgensen: None. **A.L. Ejdrup:** None. **M.D. Lycas:** None. **L.P. Posselt:** None. **K.L. Madsen:** None. **L. Tian:** None. **J.K. Dreyer:** None. **F. Herborg:** None. **A.T. Sörensen:** None. **U. Gether:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.14

Topic: I.04. Physiological Methods

Title: Mapping and localizing neurons using a robotic multiphoton microscope in NHP

Authors: *J. HAGGERTY[1], J. CHOI[3], M. W. CHOUDHURY[2], K. WINGEL[5], B. PESARAN[4], A. CHARLES[6];
[1]New York Univ., New York, NY; [2]New York Univ., Bronx, NY; [3]NYU, New York, NY; [4]Ctr. for Neural Sci., NYU, New York City, NY; [5]New York Univ. Ctr. For Neural Sci., New York, NY; [6]Johns Hopkins Univ. - Homewood Campus, Baltimore, MD

Abstract: Analyzing multiregional networks requires simultaneous access to large expanses of the brain such that we can interrogate the activity of the neurons, their locations, and their biological properties such as projections targets, cell types, and functional roles. Recently pioneered imaging methods have enabled such experiments in NHP by developing a "brain observatory": a large window for optical access across many brain areas combined with a robotically mounted microscope that can flexibly reposition to image across the window. This access creates a fundamental problem of targeting micron scale recordings in centimeter-sized expanses. Building individualized brain maps, i.e., subject specific, that can be used to localize imaging targets within the accessible fields-of-view is a critical need. Such a map would let us navigate to known or iteratively discovered target populations on-line during behavior.
We thus present a solution that involves first mapping a subject's cortical surface, and then localizing a recording session either within the same session or on subsequent imaging days. Mapping is based on stitching vascular images across the brain surface into a single map. The vascular template, computed offline, defines brain coordinates for the subject. These brain coordinates, unlike chamber coordinates obtained by measuring microscope position relative to the window, are based on the brain anatomy and consequently enable alignment between days robust to changes in brain volume and position in the cranium. Mapping typically requires X images, and requires Y min of imaging to complete. Localization can then be performed online, given the map, and situates any individual imaging session in brain coordinates by matching vascular features. We have implemented our mapping and localization approach on data from an awake, behaving primate, and find a high correlation with the microscope positioning in single days (NUMBER HERE), while noting a displacement between days indicative of shift between the chamber and the brain.

Disclosures: J. Haggerty: None. **J. Choi:** None. **M.W. Choudhury:** None. **K. Wingel:** None. **B. Pesaran:** None. **A. Charles:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.15

Topic: I.04. Physiological Methods

Support: MRC MR/T022922/1
Wellcome Trust 201225/Z/16/Z
Wellcome Trust 224668/Z/21/Z

Title: Probing long-range functional connectivity using all-optical interrogation combined with Neuropixels recordings from downstream brain areas

Authors: *E. BAUMLER[1], N. T. ROBINSON[1], M. BUCHHOLZ[2], D. NITU[1], S. BROWN[2], M. HAUSSER[3];
[2]Wolfson Inst. for Biomed. Res., [1]Univ. Col. London, London, United Kingdom; [3]UCL, London, United Kingdom

Abstract: Understanding how information is transmitted between brain areas is a fundamental problem in neuroscience. Addressing this problem requires a method for measuring functional connectivity between specific ensembles of neurons in one brain area and their downstream targets in other brain areas. This would allow us to understand how specific activity patterns are decoded and transformed by downstream neurons to guide behaviour.
To achieve this, we have developed a novel approach which combines all-optical interrogation (2-photon holographic optogenetics) with simultaneous Neuropixels probe recordings in downstream brain areas. This allows us to target functionally defined cell ensembles and photostimulate them in specific temporal and spatial patterns, while reading out the downstream impact of those manipulations with single-spike resolution. To demonstrate the utility of this approach we investigate how activity of distinct hippocampal place cell assemblies is differentially read out in a wide range of known target regions including the entorhinal cortex, septum and subiculum.
We first used 2-photon calcium imaging to identify the active place cell population in hippocampal CA1 of head-fixed mice navigating in a virtual reality environment. Next, we selectively photostimulated ensembles of place cells that were co-tuned to behaviourally salient locations (start zone vs. reward zone) and systematically varied the number and tuning coherence of stimulated neurons while recording from a range of potential downstream target areas using Neuropixels. Our preliminary results demonstrate that photostimulation of functionally characterized CA1 neurons produces either excitation or inhibition in follower neurons, with the response amplitude dependent on the number of stimulated upstream neurons. Importantly, we observed differential responses to stimulation of size-matched start vs reward cell ensembles. Finally, we interpret these responses in the context of the behavioural tuning and temporal spiking properties of the downstream neurons.
This unprecedented combination of all-optical interrogation and Neuropixels recording provides a novel way to map functional connectivity between brain areas with single neuron resolution. This will enable new experiments to probe the principles governing brain-wide information processing.

Disclosures: **E. Baumler:** None. **N.T. Robinson:** None. **M. Buchholz:** None. **D. Nitu:** None. **S. Brown:** None. **M. Hausser:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.16

Title: WITHDRAWN

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.17

Topic: I.04. Physiological Methods

Title: Tools for acquisition and analysis of simultaneous neural activity and vascular dynamics in freely behaving animals

Authors: *D. R. OLLERENSHAW[1], K. ZITELLI[2], S. GULATI[3], N. ADIL[4], J. H. FORD[5], A. H. CHANG[5], J. PAZ[5], S. Q. NEUFELD[4], J. J. NASSI[1];
[2]Sci., [1]Inscopix, Inc., Mountain View, CA; [3]Inscopix, Moutain View, CA; [4]Inscopix, Inc, Mountain View, CA; [5]Dept. of Neurol., Gladstone Institutes; Univ. of California San Francisco, San Francisco, CA

Abstract: The intricate link between neural activity and local changes in vascular dynamics is critical for brain function and prone to disruption in pathological conditions. For example, changes in vessel diameter and red blood cell velocity can be important contributing factors in neurodegenerative and neurovascular (e.g. migraine, stroke, or epilepsy) diseases. We have developed a dual color miniscope, the nVue system, that allows for imaging of neural activity and blood flow in freely behaving animals in both cortical and deep brain structures. The nVue system acquires dual color data by multiplexing two LEDs at up to 100 Hz, enabling direct visualization of both blood flow and calcium activity. To facilitate analysis, we have extended the Inscopix Data Processing Software package to include a module that provides time-varying estimates of both vascular diameter and red blood cell (RBC) velocity in vessels of interest. Diameter is measured as the full-width at half max of a Lorentzian function fit to a vessel cross section. RBC velocity is measured by calculating time-lagged cross correlations between a seed pixel at each location of interest and all pixels in a neighboring region. The distances between correlation peaks are then tracked as a function of time. The algorithms were benchmarked against ground truth manual annotation data. To our knowledge this represents the first

demonstration of simultaneous acquisition of high resolution vascular dynamics and neural circuit activity signals in freely moving animals which, coupled with out of the box analysis solutions, provides important new multi-system capabilities for basic and translational neuroscience research and preclinical therapeutic development.

Disclosures: D.R. Ollerenshaw: None. **K. Zitelli:** None. **S. Gulati:** None. **N. Adil:** None. **J.H. Ford:** None. **A.H. Chang:** None. **J. Paz:** None. **S.Q. Neufeld:** None. **J.J. Nassi:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.18

Topic: I.04. Physiological Methods

Support: NIH grant 1R01NS123681

Title: Voltage imaging of miniature EPSP and development of a new GEVI screening system

Authors: *S. LEE[1], Y. A. HAO[2], D. JIANG[1], M. Z. LIN[1,2];
[1]Dept. of Neurobio., [2]Dept. of Bioengineering, Stanford Univ., Stanford, CA

Abstract: Genetically encoded voltage indicators (GEVIs) are becoming a tool of choice for various neuroscience experiments. Recently, GEVIs have been demonstrated in the olfactory bulb, hippocampus, striatum, and somatosensory cortex of living rodents as well as in zebrafish larvae and fruit flies. With a few exceptions, however, these examples mainly recorded suprathreshold spikes. One of the most crucial advantages of GEVIs would be the ability to report subthreshold activities at the cellular level where it is difficult, if not impossible, to investigate by using calcium indicators or multi-electrode arrays. Homeostatic synaptic plasticity is a form of non-Hebbian plasticity that helps neurons maintain stable synaptic strength. Quantification of the synaptic strength is typically done by recording miniature excitatory postsynaptic potential or current under a whole-cell patch-clamp mode. We sought to try voltage imaging of the miniature events from rat hippocampal neurons with ASAPs, a leading series of GEVIs (ASAP1-ASAP4) that the Lin lab has developed. Using one of the ASAPs with a high dynamic range and fast response, miniature EPSPs with amplitudes of only a few millivolts could be visualized reliably. We will also present our new strategy to develop the next-generation GEVI screening system that involves hydrogel bioelectronics. This system uses a polyacrylamide gel array as a platform for cellular imaging and manipulation of membrane potential. Its capacity to screen 65,000+ mutants in a single run will enable a 100-fold improvement compared to our current system. We will discuss the idea behind the strategy and preliminary results.

Disclosures: S. Lee: None. **Y.A. Hao:** None. **D. Jiang:** None. **M.Z. Lin:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.19

Topic: I.04. Physiological Methods

Support: NIMH Grant 1R44MH129023-01

Title: Scanned line angular projection two photon laser scanning (SLAP2) microscopy for real-time volumetric in vivo imaging at subcellular resolution

Authors: *J. R. GLASER[1], B. KIMMEL[2], G. JAINDL[2], P. ANGSTMAN[1], K. PODGORSKI[3], D. A. FLICKINGER[4], N. ROUSSEL[1], J. KING[2], N. J. O'CONNOR[1];
[1]MBF Biosci., Williston, VT; [2]MBF Biosci., Ashburn, VA; [3]Allen Inst., Seattle, WA; [4]Inst. Design & Fab, Howard Hughes Med. Inst., Ashburn, VA

Abstract: Optical imaging of in vivo neuronal populations using fluorescent voltage and neurotransmitter indicators is an emerging frontier that requires microscopic imaging of brain volumes at kilohertz rates in highly scattering, live brains. SLAP2 uses a novel and unique scan technology where a vertical laser line is horizontally swept nearly 10,000 times per second across the surface of a Digital Micromirror Device (DMD) that projects excitation light to random regions of interesting (ROI) of a specimen. This approach enables scanning multiple cell-sized volumetric ROIs at kilohertz rates. The DMD can switch “on" and “off" individual pixels in a field of view (FOV) at full scan rates, allowing random-access imaging of ROIs with minimal mechanical movement overhead associated with other random-access imaging technologies. MBF Bioscience is continuing to work with Dr. Kaspar Podgorski to further improve the SLAP2, including developing software that improves the reliability and performance of the original prototype system. These improvements include software user experience optimizations for controlling the specialized hardware required for rapid imaging of multiple ROIs. Key features include automated calibration tools for tuning acquisition parameters, and real-time display of their impacts on acquired images. Experimental parameters are also saved for experimental reproducibility. We are currently developing tools in SLAP2 that allow researchers to easily configure complex imaging experimental workflows comprised of both structural and functional activity imaging of multiple neurons, without requiring computer programming expertise. SLAP2 is capable of imaging in vivo mouse brain and in vitro brain slices at subcellular spatial resolution, and with a temporal resolution of milliseconds (i.e., 1000 Hz and higher). We believe that SLAP2 will be an asset to many experimental paradigms focused on imaging of living neuronal circuits. Characterizing physiological activity in three-dimensional culture and organoid models will all benefit from this fast, large-scale microscopic imaging advancement.

Disclosures: **J.R. Glaser:** A. Employment/Salary (full or part-time):; MBF Bioscience. **B. Kimmel:** A. Employment/Salary (full or part-time):; MBF Bioscience. **G. Jaindl:** A. Employment/Salary (full or part-time):; MBF Bioscience. **P. Angstman:** A. Employment/Salary (full or part-time):; MBF Bioscience. **K. Podgorski:** None. **D.A. Flickinger:** None. **N. Roussel:**

A. Employment/Salary (full or part-time):; MBF Bioscience. **J. King:** A. Employment/Salary (full or part-time):; MBF Bioscience. **N.J. O'Connor:** A. Employment/Salary (full or part-time):; MBF Bioscience.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.20

Topic: I.04. Physiological Methods

Support: DFG CRC 854 TP Z01
BMBF 13N15790
IB Saxony-Anhalt 1904/00045 (ZS/2018/10/95184)

Title: Imaging neural activity-linked metabolic dynamics by label-free wide-field FLIM

Authors: *W. ZUSCHRATTER[1], A. WEBER[1], E. ALTUN[2], A. WETZEL[2], A. BIKBAEV[3], A. LUARTE[4], R. HERRERA-MOLINA[1];
[1]Leibniz Inst. for Neurobio., Magdeburg, Germany; [2]Otto von Guericke Univ., Magdeburg, Germany; [3]Johannes Gutenberg-University, Mainz, Germany; [4]Univ. de Los Andes, Santiago, Chile

Abstract: We describe an imaging method that can follow small metabolic changes associated with the activity of brain cells over long periods under physiological conditions without invasive staining procedures and with minimal cell damage. Fluorescence lifetime imaging microscopy (FLIM) enables the determination of energy-related redox and protein-bound states in a label-free manner by analyzing the autofluorescence of metabolically relevant molecules such as NAD(P)H and FAD. As such, the fluorescence lifetime of these intrinsic metabolites provides essential information about the composition and conformation of molecules, which can be used to monitor activity- or pharmacologically-induced changes in neuronal cell cultures. However, observing such small changes in active cells is challenging due to these metabolic molecules' low quantum yield. Furthermore, correct FLIM measurements require robust statistics based on a sufficient number of photons collected.For this reason, a reliable FLIM system must meet the following criteria: (1) high sensitivity, (2) high signal-to-noise ratio, (3) temporal resolution in picoseconds, and (4) a detection range equivalent to the entire field of view.Here we present a novel wide-field FLIM method for NAD(P)H/FAD molecule detection using an innovative, commercially available camera system (LINCam, Photonscore GmbH, Germany) based on time-correlated single-photon counting (TCSPC). The camera features a uniquely high signal-to-noise ratio, high temporal resolution (< 50 ps), and a high sensitivity, enabling it to operate under low light conditions (< 30 mW/cm^2 on average). We show that the LINCam-based FLIM system can measure changes in metabolic activity of electrically stimulated cultured neurons. These experiments reveal a close correlation between neuronal activity and the dynamic changes of the observed metabolites. We show that the high sensitivity of the LINCam enables significant

spatial scalability and high temporal resolution to resolve fluctuations in the molecular states of NAD(P)H/FAD during single-neuron imaging. The recorded live-cell data can be merged with images from immunostained samples after fixation to characterize the subcellular source of the measured metabolic activity. In another study, we examined retinal cell metabolism under normal and pathophysiological conditions. Collectively, this novel technology may contribute to the generation of diagnostic tools based on minimally invasive imaging of patients with several pathological conditions.

Disclosures: **W. Zuschratter:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Photonscore GmbH. **A. Weber:** A. Employment/Salary (full or part-time):; Photonscore GmbH. **E. Altun:** None. **A. Wetzel:** None. **A. Bikbaev:** None. **A. Luarte:** None. **R. Herrera-Molina:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.21

Topic: I.04. Physiological Methods

Title: Brain micro-anatomy revealed by 2-photon shadow imaging in vivo

Authors: *Y. DEMBITSKAYA, G. LE BOURDELLES, S. BANCELIN, J. GIRARD, M. SATO-FITOUSSI, U. NÄGERL;
Interdisciplinary Inst. for Neuroscience, Univ. of Bordeaux/CNRS, Bordeaux, France

Abstract: Getting an accurate, detailed and physiologically relevant view of brain structure and neuronal circuits is a major goal of modern neuroscience. Current large-scale connectomics efforts rely either on EM or MRI, which are either incompatible with live conditions or do not offer cellular resolution. Fluorescence microscopy allows for live imaging with cellular resolution *in vivo*, but has relied on positively labeling of a sparse set of cells, giving an incomplete and biased view of the anatomical organization of brain tissue. Breaking this impasse, super-resolution shadow imaging (SUSHI) established a new paradigm to visualize tissue anatomy in brain slices with nanoscale resolution in an all-encompassing and panoramic way, based on fluorescence labeling of the ACSF and 3D-STED microscopy. Because of the stringent optical demands of super-resolution microscopy, however, the approach has only been applied to living organotypic brain slices so far. We have now extended the shadow imaging concept to the mouse brain *in vivo*, based on 2-photon shadow imaging (TUSHI) and labeling of the cerebrospinal fluid with a fluorescent membrane-impermeant dye. We present the optical details of the microscope, the labeling strategy for sufficiently bright and homogeneous inverted cellular contrast, as well as the cranial window technique and anesthesia formula for optically clear and mechanically stable access to superficial layers of the cerebral cortex. Despite the diffraction-limited resolution, the new approach opens a stunning window on the micro-anatomical organization of the brain *in vivo*, where cell bodies, dendritic branches of neurons,

perivascular spaces and spatial heterogeneities in the extracellular space become visible. By adding a second fluorescence channel, the shadow imaging approach reveals the diverse and complex anatomical context of positively labeled neurons, astrocytes, microglia, and tumor cells. In summary, our work demonstrates the feasibility of TUSHI *in vivo* to visualize brain structure and context with subcellular resolution. It provides a powerful new investigative tool to monitor dynamical changes of brain structures *in vivo* under various (patho-physiological) conditions, such as experience-dependent neuronal plasticity, sleep, aging, stroke, tumor invasion & proliferation.

Disclosures: Y. Dembitskaya: None. **G. Le Bourdelles:** None. **S. Bancelin:** None. **J. Girard:** None. **M. Sato-Fitoussi:** None. **U. Nägerl:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.22

Topic: I.04. Physiological Methods

Support: NIH Grant U01NS103517
NIH Grant U01NS120822
NIH Grant UF1NS107610
NIH Grant U19NS104590
NSF NeuroNex DBI-1707261
DARPA Grant N6600117C4012 (NESD)
DARPA Grant N6600119C4020 (N3)

Title: Dual-polarity voltage imaging reveals functional interactions between genetically identified cell classes during behavioral state transitions

Authors: *M. KANNAN[1,2], G. VASAN[1,2], S. HAZIZA[3,5], M. J. SCHNITZER[5,6,4], V. A. PIERIBONE[1,2];
[1]Yale Univ., Yale Univ., New Haven, CT; [2]The John B Pierce Lab., New Haven, CT; [3]Biol., [4]CNC Program, Stanford Univ., Palo Alto, CA; [5]James H. Clark Ctr., Stanford, CA; [6]Howard Hughes Med. Inst., Stanford, CA

Abstract: The dynamic interplay between multiple excitatory and inhibitory cell classes lies at the core of neural signal processing underlying animal perception and behavior. Genetically encoded fluorescent voltage indicators (GEVIs) are uniquely poised to uncover the pertinent millisecond-scale interactions *between* targeted cell classes. We describe a suite of four mutually compatible GEVIs, comprising the second-generation FRET-opsin indicators Ace-mNeon2 and VARNAM2 as well as their respective reverse response polarity variants pAce and pAceR. The new indicators enable single-neuron, single-spike resolution voltage recordings from large genetically identified ensembles in head-restrained, running mice in high-speed (0.4-1 kHz)

epifluorescence microscopy. The studies revealed cell class-specific modulations of spiking patterns during behavioral state transitions in the primary visual cortex. Transmembrane voltage recordings from ~1200 cells showed that a major fraction of all recorded cell classes, namely, pyramidal neurons (PNs) and interneurons expressing neuron-derived neurotrophic factor (NDNF), somatostatin (SST), or vasoactive intestinal peptide (VIP), increased their spontaneous firing rates during a rest-to-arousal transition. However, SST-neurons exhibited the most profound state-dependent activation, with 78% of cells increasing their firing rates, closely followed by NDNF-neurons. By contrast, only half of the VIP-neurons and PNs elevated their spontaneous activity whereas a third of these populations exhibited diminished spiking in aroused states. To further dissect the fine-scale distinctions in the cell class-specific activation patterns, we performed simultaneous dual-population recordings using dual-polarity multiplexed voltage imaging (DUPLEX). By targeting the green indicators with opposite signaling polarities to distinct cell classes, we obtained single-channel, paired voltage recordings from NDNF- & VIP-neurons, SST- & VIP-neurons, or VIP- neurons & PNs, in which we inferred each cell's class by the directionality of its optical spike waveform. Our results revealed that the voltage dynamics of neighboring pairs of NDNF- & VIP-neurons, and of VIP- and PN-neurons are uncorrelated under baseline conditions with no change during arousal. In contrast, DUPLEX uncovered strong negatively correlated dynamics between SST- and VIP-neuron pairs under baseline conditions and a significant rise in this anti-correlation during arousal. Together, our work showcases multipopulation voltage imaging for investigations of the fine-scale dynamics of interclass interactions in behaving animals.

Disclosures: **M. Kannan:** None. **G. Vasan:** None. **S. Haziza:** None. **M.J. Schnitzer:** None. **V.A. Pieribone:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.23

Topic: I.04. Physiological Methods

Support: Business Finland Grant 42539/21/2020
State research funding Grant K33755

Title: Functional near-infrared spectroscopy reveals evident water and hemodynamic responses in human brain during clinical radiotherapy

Authors: ***V. O. KORHONEN**[1], T. MYLLYLÄ[3], P. KARTHIKEYAN[3], U. HONKA[3], J. LOHELA[2], K. INGET[2], H. FERDINANDO[3], M. JÄRVELÄ[3], J. KANANEN[3], S. KARHULA[2], V. KIVINIEMI[1], J. NIKKINEN[2];
[1]Diagnostics, [2]Oncology, Oulu Univ. Hosp., Oulu, Finland; [3]Univ. of Oulu, Oulu, Finland

Abstract: Introduction Radiotherapy is a cornerstone of clinical management of brain tumors. Radiation increases the free oxygen radical concentration by ionizing water molecules in tissue, and the effectiveness of radiotherapy depends on the amount of these radicals. However, to the best of our knowledge, no method to measure direct human brain tissue effects in real-time during radiation exists. Thus, our aim is to develop non-invasive, real-time method to measure the effect of radiotherapy during the treatment. **Methods** The functional near-infrared spectroscopy (fNIRS) device used in this study utilizes frequency-coding technique that is based on modulating the illuminating light, each wavelength at a specific frequency, and demodulating these after receiving. The patient is positioned in a supine position in the medical linear accelerator and two fNIRS channels with source detection distance of 3 cm are attached on their forehead above the mask for measuring the radiotherapy response in real-time. Ionizing radiation inside the radiotherapy room poses high demands on the devices used. E.g. with fNIRS device, all optical fibers and optodes must be compatible with radiotherapy to avoid interfering the treatment procedure. In our setup, we placed only optical fibres and optodes in the treatment room and the fNIRS device was placed in the control room by a fiber entry. In this study, we performed a total of 48 measurements from 14 different brain tumor patients (age: 65.0 +- 9.7 years, 6 females) with fNIRS during whole brain radiation therapy. Every treatment plan consisted of left and right side radiation fields shaped with multileaf collimator. Measured raw fNIRS time courses (sampling rate 1 kHz) were converted into time courses representing temporal changes in cerebral water (H_2O), oxygenated (HbO), deoxygenated (Hb) and total hemoglobin (HbT) concentrations using the modified Beer-Lambert law. **Results** We observed that all hemoglobin related concentrations (HbO, HbR, HbT) increased and, in contrast, H_2O concentration decreased in both fNIRS channels during the both radiation fields. **Conclusion** Our results display instant cerebral H_2O and hemodynamic responses recorded via non-invasive fNIRS during clinical radiotherapy. Furthermore, our study demonstrate the feasibility of monitoring the immediate radiation induced changes of H_2O, HbO, HbR and HbT and in the human brain in real-time. We suggest that the decreased H_2O and increased hemoglobin related changes are direct effects of radiation and thus the fNIRS device may potentially be used for monitoring cerebral hemoglobin and water concentration changes during radiation therapy.

Disclosures: V.O. Korhonen: None. **T. Myllylä:** None. **P. Karthikeyan:** None. **U. Honka:** None. **J. Lohela:** None. **K. Inget:** None. **H. Ferdinando:** None. **M. Järvelä:** None. **J. Kananen:** None. **S. Karhula:** None. **V. Kiviniemi:** None. **J. Nikkinen:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.24

Topic: I.04. Physiological Methods

Support: US NIH Grant U01EB027005 (J.P.C.)
US NIH Grant F31NS110261 (Z.E.M.)
US NIH Grant R21MH109775 (A.T.E.)

US NIH Grant K01MH103594 (A.T.E.)
Facebook Sponsored Academic Research Agreement (J.P.C.)

Title: Decoding the identities of naturalistic movies from human brain hemodynamics using high-density diffuse optical tomography

Authors: *Z. E. MARKOW[1], K. TRIPATHY[2], J. W. TROBAUGH[3], A. M. SVOBODA[7], M. L. SCHROEDER[8], S. M. RAFFERTY[4], E. J. RICHTER[3], A. T. EGGEBRECHT[5], M. A. ANASTASIO[9], J. P. CULVER[6];
[1]Biomed. Engin., [2]Neurosci., [3]Electrical and Systems Engin., [4]Radiology, [5]Radiology, Biomed. Engin., [6]Radiology, Biomed. Engineering, Physics, Washington Univ. In St. Louis, Saint Louis, MO; [7]Col. of Med., Univ. of Cincinnati, Cincinnati, OH; [8]Speech, Language, and Hearing Sci., Purdue Univ., West Lafayette, IN; [9]Bioengineering, Univ. of Illinois at Urbana-Champaign, Urbana, IL

Abstract: In the last decade, functional MRI (fMRI) has achieved remarkable success at decoding the identity and content of a wide range of stimuli experienced by human subjects, using their brain activity and machine learning methods. Unfortunately, fMRI employs bulky equipment that cannot be used at the bedside, cannot image many patients with metal implants, and is difficult in young children. More-lightweight non-invasive tools have not accomplished the decoding performance of fMRI. High-density diffuse optical tomography (HD-DOT) non-invasively images blood oxygen fluctuations related to brain activity, like fMRI, but with near-infrared light. HD-DOT is an alternative in fMRI-incompatible patients and settings, and has spatial resolution between fMRI and more-lightweight non-invasive technologies such as NIRS and EEG. To evaluate feasibility of decoding naturalistic movie stimuli with HD-DOT, we first evaluated how well an HD-DOT system could identify which of four 90-second movie clips was being viewed based on imaged brain dynamics. During each session, each clip was viewed 4-8 times (trials) total. For the first half of each session (training set), the brain responses to each clip were averaged to form a "template" response for each clip. For each remaining trial (testing set), our simple decoder guessed that the clip shown was the clip whose template had the highest correlation with the measured response. These guesses were 93% accurate on average across 16 sessions from 4 subjects, well above chance (25%) in terms of effect size and statistical significance ($p<0.001$). Next, we attempted to decode which clips were shown in one session using the template responses from a different session in the same subject. These guesses were 86% accurate on average across those 16 sessions, still well above chance ($p<0.001$). Finally, we attempted to identify clips outside a decoder's training set. To do this, we relabeled two clips and their responses as training trials, and we used these to fit a linear model for predicting the imaged response from wavelet motion energy features in any clip. The model was used to predict the responses to the other two (test) clips and to two extra "lure" clips that subjects had never viewed. This decoder guessed that the clip viewed in each trial was the clip whose predicted response had the highest correlation with the measured data. Across subjects and sessions, average accuracy exceeded 60% and statistically exceeded chance (25%, $p<0.05$). Accuracy remained above chance even with up to 40 lure clips. Future work will attempt to decode more types of stimuli.

Disclosures: Z.E. Markow: None. **K. Tripathy:** None. **J.W. Trobaugh:** None. **A.M. Svoboda:** None. **M.L. Schroeder:** None. **S.M. Rafferty:** None. **E.J. Richter:** None. **A.T. Eggebrecht:** None. **M.A. Anastasio:** None. **J.P. Culver:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.25

Topic: I.04. Physiological Methods

Support: NIH R44OD024879
NSF 2036439

Title: Using gigapixel Multi-Camera Array Microscopy (MCAM) imaging to predict zebrafish visuomotor performance

Authors: J. P. BECHTEL[1], M. LORING[2], K. FOUKE[3], J. EFROMSON[1], T. J. J. DOMAN[1], P. REMEAY[1], S. A. CARPENTER[3], P. BOKADIA[1], G. HORSTMEYER[1], E. A. NAUMANN[4], ***M. HARFOUCHE**[5];
[1]Ramona Optics, Durham, NC; [2]Neurobio., Duke Univ. Hlth. Syst., Durham, NC; [3]Duke Univ., Durham, NC; [4]Neurobio., Sch. of Med., Durham, NC; [5]Ramona Optics Inc, Durham, NC

Abstract: The zebrafish (Danio rerio) is a popular model in the study of vertebrate genes, neural function, and behavior due in part to their small size, transparency and large number of offspring (up to 200 in a clutch). However, not all of these individual zebrafish perform equally well in neural imaging and behavioral experiments, requiring researchers to go through multiple pre-screening steps. In a typical workflow, only a few healthy zebrafish are selected at random from a clutch for in-depth examination in behavioral or calcium imaging assays. The selection criteria, often loosely documented, can vary from scientist to scientist and reduce the reproducibility of these experiments. Here, we show how one can use a novel gigapixel scale multi-camera array microscope (MCAM) to simultaneously monitor the development of hundreds of zebrafish from 24-hours post fertilization through 5 days post fertilization to reduce sampling bias and improve pre-screening standardization in behavioral and neuroimaging experiments. After an entire clutch is plated in several 96 well plates, each individual plate is synchronously imaged at high resolution under the MCAM at various spatial resolution scales (10µm - 30µm / pixel) and temporal scales (up to 120 frames per second). The MCAM simultaneously records the activity of the larvae under infrared illumination (850 nm) and stimulates the zebrafish with repeated light and vibrational stimuli. Each recording results in a quantitative metric relating to the activity of the larvae at that age. We then correlate these metrics with behavior observed in a freely swimming closed loop behavior setup, with a 2-hour observation session per individual zebrafish. We show that these early metrics can be used as an indicator for the future visuomotor behavioral performance of the zebrafish at 6-7 dpf in expanded free-swimming optomotor response assays. Together, these experiments demonstrate how simultaneous large scale, high-resolution behavioral imaging with the MCAM provides a rigorous, unbiased method to standardize sample selection.

Disclosures: **J.P. Bechtel:** None. **M. Loring:** None. **K. Fouke:** None. **J. Efromson:** None. **T.J.J. Doman:** None. **P. Remeay:** None. **S.A. Carpenter:** A. Employment/Salary (full or part-time):; Ramona Optics. **P. Bokadia:** None. **G. Horstmeyer:** None. **E.A. Naumann:** None. **M. Harfouche:** None.

Poster

577. Optical Methodology: Application

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 577.26

Topic: I.04. Physiological Methods

Support: ERC 692943 (BrainBIT)
H2020 N. 945539 (HBP-SGA3)

Title: Optical perturbational approach to study information integration among distributed cortical regions in mice

Authors: ***F. RESTA**[1], A. SCAGLIONE[1], E. CONTI[2], G. MAZZAMUTO[3], A. ALLEGRA MASCARO[4], F. S. PAVONE[1];
[1]LENS European Lab. for Non-Linear Spectroscopy, Univ. of Florence, Sesto Fiorentino, Italy; [2]European Lab. for Non-linear Spectroscopy, Sesto Fiorentino, Italy; [3]Consiglio Nazionale delle Ricerche, Sesto Fiorentino, Italy; [4]Neurosci. Inst., Natl. Res. Council, Pisa, Italy

Abstract: Brain responsiveness and its activation complexity are linked to the level of consciousness. However, how these features change across brain states is still not clear. The combination of Transcranial Magnetic Stimulation (TMS) and hd-EEG recordings represents the gold standard method to address this issue in humans. A preclinical analogous in lab animals would provide novel mechanistic insights into the brain-state-dependent complexity of the brain. A promising non-invasive approach to simultaneously record and stimulate neuronal activity in mice is the use of all-optical neurophysiological methods. Here we established a crosstalk-free large-scale all-optical method combining wide-field fluorescence imaging of the red-shifted calcium indicator jRCaMP1b and transcranial optogenetic stimulation of Channelrhodopsin-2 (ChR2). To achieve a cortex-wide expression we took advantage of the recently developed viral technology AAV.PHP.eB. This led to a uniform expression of the functional indicator and the opsin in the whole cortex, raising the possibility of causally investigating the whole dorsal cortical mantle in wild-type mice. Results show that in awake mice, optogenetic stimulations at increasing laser power (0.1 - 15 mW) evoked a distributed cortical response in several areas in the two hemispheres, whereas, during anesthesia, stimulation led to a localized response limited in space and time. These results suggest that response complexity decrease with the levels of consciousness, as observed in pathological patients affected by disorders of consciousness.

Disclosures: **F. Resta:** None. **A. Scaglione:** None. **E. Conti:** None. **G. Mazzamuto:** None. **A. Allegra Mascaro:** None. **F.S. Pavone:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.01

Topic: I.06. Computation, Modeling, and Simulation

Support: Canadian Institute for Advanced Research Azrieli Global Scholars Program

Title: Low-dimensional task-relevant information extraction in fMRI data using Task Relevant Autoencoder via Classifier Enhancement (TRACE) model

Authors: *S. OROUJI[1], V. TASCHEREAU-DUMOUCHEL[2], A. CORTESE[3], B. ODEGAARD[4], C. CUSHING[5], M. CHERKAOUI[5], M. KAWATO[6], H. LAU[7], M. PETERS[1]; [1]Cognitive Sci., Univ. of California-Irvine, IRVINE, CA; [2]Psychiatry and addictology, Univ. de Montréal, Montreal, QC, Canada; [3]Cognitive Mechanisms Laboratories, Dep. of Decoded Neurofeedback, ATR Inst. Intl., Kyoto, Japan; [4]Psychology, Univ. of Florida, Gainesville, FL; [5]Psychology, UCLA, Los Angeles, CA; [6]ATR BICR, Kyoto, Japan; [7]RIKEN Ctr. for Brain Sci., Wako, Japan

Abstract: Functional magnetic resonance imaging (fMRI) is a widely used neuroimaging technique to explore and characterize neural representations. However, fMRI data is contaminated by noise and task-irrelevant information, due to factors such as attention and the arousal level of participants. Additionally, fMRI data often suffer from extreme sparsity of sample size: with thousands of voxels (features) but often only hundreds of presentations of a given stimulus (samples). Thus, fMRI data typically possess extremely poor samples-to-features ratios. As a consequence, state-of-the-art deep learning models often tend to overfit on these small-scale datasets. Therefore, there is a critical need for developing assumption-free models that not only are able to find a low-dimensional task-relevant representation of fMRI data but also can do so within the constraints of data paucity. To address these issues, we propose the Task Relevant Autoencoder via Classifier Enhancement (TRACE) model. TRACE's architecture is similar to a standard autoencoder (AE), except a classifier is attached to the bottleneck to ensure the low dimensional representations are in fact task-relevant. We benchmarked TRACE against a standard AE using the MNIST and Fashion MNIST datasets, including under extreme data paucity. We then compared the task-relevancy of the representations extracted by TRACE and AE in a real-world setting by applying them to a fMRI dataset from 60 individuals who viewed pictures of animals and objects while fMRI data from their ventral temporal cortex (VTC) was collected. Our results showed that TRACE drastically outperforms AE in extracting more task-relevant information and exhibits substantially more robust behavior in the presence of data paucity. TRACE is able to achieve these successes in an assumption-free manner, and further can project the extracted low-dimensional representations back into the original functional and anatomical brain space in ways which were found to be cleaner and more categorically informative even than the input data itself. Our findings suggest exciting

possibilities to use these more canonical reconstructed inputs as category representation in decoded neurofeedback studies.

Disclosures: S. Orouji: None. **V. Taschereau-Dumouchel:** None. **A. Cortese:** None. **B. Odegaard:** None. **C. Cushing:** None. **M. Cherkaoui:** None. **M. Kawato:** None. **H. Lau:** None. **M. Peters:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.02

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH R01NS117405

Title: Computational modeling of corticospinal waves evoked by transcranial magnetic stimulation across multiple subjects

Authors: *G. J. YU[1], K. KUMARAVELU[1], F. RANIERI[2], V. DI LAZZARO[3], M. A. SOMMER[1], A. V. PETERCHEV[1], W. M. GRILL[1];
[1]Duke Univ., Duke Univ., Durham, NC; [2]Univ. of Verona, Verona, Italy; [3]Campus Bio Medico Univ. of Rome, Rome, Italy

Abstract: Transcranial magnetic stimulation (TMS) is a non-invasive neuromodulation modality with myriad diagnostic and therapeutic applications. However, it is still unknown precisely which mechanisms contribute to the neuronal network response to TMS. When stimulating the motor cortex with TMS, corticospinal recordings can provide information about evoked activity in layer 5 of the motor cortex. Indirect (I) waves can be isolated within the corticospinal response and are the result of a series of transsynaptic activations caused by the initial stimulation, i.e., they are representative of the network response. Therefore, corticospinal I-waves are an important readout of cortical circuit activity for investigating the response to TMS. In this work, epidural recordings of corticospinal I-waves from the cervical spinal cord of human subjects in response to a single TMS pulse were used to constrain a spiking neuronal network model of motor cortex. The neuronal network model was comprised of leaky-integrate-and-fire point neurons that represented layers 2/3, 5, and 6 with excitatory and inhibitory cell types for each layer. The parameters optimized included the synaptic strengths (network parameters) and the proportions of neurons activated by the TMS pulse (activation parameters). Particle swarm optimization was used to identify model parameters that minimized the error of the magnitudes and timings of the I-wave peaks and the baseline firing rates of the cell types. Additional constraints included dose-response curves relating TMS stimulation intensity to the cortical silent period and the amplitude of motor evoked potentials. Subject-specific models yielded the best fits with different combinations of network and activation parameters for each subject. Optimization using all subject data simultaneously yielded an all-subject model with shared

network parameters but different activation parameters. The all-subject model exhibited poorer fits on a per-subject basis but better generalizability across subjects. A sensitivity analysis was performed by applying a regularized multilinear regression on the parameters explored by the optimization process. The regularization eliminated the parameters that contributed least to the I-wave properties. Across all properties, the sensitivity analysis reduced the number of relevant parameters by an average of 18%. The results predicted that the peaks and timings of corticospinal waves are most sensitive to layer 2/3 and layer 5 pyramidal cells, though the projections of both layer 2/3 and layer 5 to layer 6 pyramidal cells are also important.

Disclosures: **G.J. Yu:** None. **K. Kumaravelu:** None. **F. Ranieri:** None. **V. Di Lazzaro:** None. **M.A. Sommer:** None. **A.V. Peterchev:** None. **W.M. Grill:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.03

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH MH122023
NSF OAC-1730655

Title: Intrinsic oscillatory dynamics in a model neocortical circuit

Authors: *S. S. NAIR[1], **Z. CHEN**[2], D. FAGUE[3];
[1]Univ. of Missouri Columbia, Univ. of Missouri, Columbia, MO; [2]Electrical Engin. and Computer Sci., Univ. of Missouri, Columbia, Columbia, MO; [3]Electrical Engin. & Computer Sci., Univ. of Missouri, Columbia, Fulton, MO

Abstract: Rhythmic activity is closely linked to neuronal processing within and across brain regions. The microcircuit mechanisms that subserve such rhythms via intrinsic and extrinsic inputs are being studied actively. Neocortical circuits exhibit both beta and gamma rhythms which are through to be caused by distinct subcircuits involving different type of interneurons. Another approach to study the microcircuit mechanisms is via population vectors. However, it is not clear how rhythmically oscillating cell assemblies and population vectors are related, and how they influence each other. We report a 1000-cell biophysical computational model of a neocortical circuit to investigate how different patterns of population activity recruit specific beta and gamma generating micro-circuits. Our preliminary results with a 1000-cell network model indicated that stable afferent ensembles recruit beta while varying ensembles recruit gamma oscillations. Previous work on gamma and beta oscillations have emphasized the resonant properties of interneuron networks as being instrumental in rhythm generation. Our preliminary model indicated that this is only part of the reason for the emergence of brain rhythms. The frequency of the rhythm is largely determined by the resonance of the recurrent principal cells/interneuron network. However, whether that network is activated, and the persistence of

that activity, depends upon the slower temporal dynamics of the principal cell ensembles that supply the excitation to fuel the rhythm. Our preliminary model did not have specific layers and had only one type of pyramidal cells. The revised network model includes all pertinent layers in the neocortex with their predominant cell types. Model single cell characteristics as well as network features such as cell numbers, connectivity, synaptic and short- and long-term plasticity characteristics were calibrated using biological reports. We explored how random extrinsic drive engages the population vectors by varying either the drive intensity or the ensemble recruited. We also explored activation of different ensembles and how the population vector trajectory transitions on the low dimensional manifold. On-going work explores how the connectivity between model cell types determines the population vector trajectory and studies the dynamic of the transition between gamma and beta oscillation.

Disclosures: **S.S. Nair:** None. **Z. Chen:** None. **D. Fague:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.04

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH NS109552-01
Craig H. Neilsen Foundation 599050
NIH NS125863-01A1
NSF HRD-1547723

Title: Supercomputer simulation-based reverse engineering of motoneuron firing patterns

Authors: M. K. CHARDON[1], ***C. WANG**[2], M. GARCIA[3], R. K. POWERS[4], C. HECKMAN[1]; [1]Neurosci., Northwestern Univ., Chicago, IL; [2]Electrical and Computer Engin., California State University, Los Angeles, Los Angeles, CA; [3]Argonne Natl. Lab., Lemont, IL; [4]Physiol. & Biophysics, Univ. of Washington, Seattle, WA

Abstract: Massively parallel computation via supercomputers offers the possibility of remarkably extensive parameter searches, a capacity well matched to the multitude of possible inputs driving a particular set of neurons. Our recent studies combine 3 advances: 1) novel array electrodes to measure firing patterns of populations of motoneurons in humans, via motoneuron connections to muscle fibers, 2) highly realistic computer simulations of these motoneurons and 3) implementation of these models using supercomputers at the Argonne Laboratory Computing Resource Center. The huge advantage of the supercomputer approach is that their massive parallelism allows thousands (soon millions) of simulations to be carried out simultaneously. In this particular application, we use this computational power as the basis of a brute force approach to reverse engineer motoneuron firing patterns to identify the organization of their synaptic inputs. More specifically our goal is to identify the patterns of excitatory, inhibitory and

neuromodulatory inputs. Our initial simulations (solving 7M+ ODE models) show that, although a given motor output pattern could potentially be generated by a huge number of combinations of these three types of input, neuromodulatory input makes motoneuron input-output properties so nonlinear that the effective “solution space” is restricted. These high levels of neuromodulatory inputs are coupled to strong inhibitory inputs, with interaction between these two input types providing the “amplifier” upon which excitation acts to execute the pattern of the movement. This is a novel insight into the synaptic organization of motor commands in humans. Overall, our results show that this reverse engineering approach can achieve deep insights about the organization of synaptic inputs that drive a set of neuronal firing patterns. Identifying the separate patterns of excitatory and inhibitory inputs appears to require highly nonlinear processing within the neurons.

Disclosures: **M.K. Chardon:** None. **C. Wang:** None. **M. Garcia:** None. **R.K. Powers:** None. **C. Heckman:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.05

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH Award R43 NS117226

Title: Application of Connectome Fingerprinting to predict functional organization of motor cortex in individual glioma patients

Authors: *V. TRIPATHI[1], B. M. HAGER[2], L. RIGOLO[3], B. K. BRACKEN[2], A. J. GOLBY[3], Y. TIE[3], D. C. SOMERS[1];
[1]Dept. of Psychological and Brain Sci., Boston Univ., Boston, MA; [2]Sensing, Perception, and Applied Robotics (SPAR) Div., Charles River Analytics, Cambridge, MA; [3]Dept. of Neurosurg., Brigham and Women's Hosp., Brookline, MA

Abstract: Neurosurgery is a key part of the treatment for brain tumors, and individual functional brain mapping plays an important role in the prevention of surgery-induced permanent deficits while achieving a maximal resection to increase survival. Task-based functional MRI (fMRI) is the method of choice for non-invasive, individualized functional brain mapping to inform presurgical planning and intraoperative decision-making (Silva et al, 2018; Golby et al., 2020). However, many patients are unable to reliably perform tasks during fMRI, and a sizable battery of tasks may need to be administered. Connectome Fingerprinting (CF) is a non-invasive precision brain mapping approach that can accurately predict individual functional brain organization from resting-state fMRI (rs-fMRI). Rs-fMRI eliminates subject task performance concerns, and one set of rs-fMRI data yields multi-battery task predictions. CF constructs high resolution models of connectome - task fMRI activation relationships (e.g., Tobyne et al, 2018).

Here, we examined the feasibility of applying CF methods to predict motor system functional organization in 6 glioma patients (3 low grade, 3 high grade, aged 24-47 years), 15 healthy control subjects acquired on the same MR scanner, and 69 healthy control subjects from the Human Connectome Project (HCP-YA). Rs-fMRI and motor task fMRI was collected in all subjects, permitting validation of CF predictions. A CF model was constructed from motor task fMRI (hand, foot, lip/tongue) and rs-fMRI collected in 50 other HCP-YA subjects; a separate CF model was constructed from the local controls with cross validation. CF accuracy for each participant was assessed by Pearson correlation of the predicted and actual spatial activation patterns. Individual HCP-YA predictions from 60 mins of rs-fMRI strongly correlated with task fMRI: LH foot (r=0.63), LH hand (r=0.66), RH foot (r=0.63), RH hand (r=0.67), and bilateral lip (r=0.68). With only 4 minutes of rs-MRI, prediction accuracy fell but remained robust (r=0.46 to 0.53). Within scanner predictions for local controls was stronger (r=0.23-0.51) than cross-scanner predictions (r=0.14-0.39), highlighting the need for cross-scanner harmonization. The CF model trained on local control subjects (r=0.31-0.42) outperformed the HCP-YA model (r=0.07) in predictions of hand task activations in glioma patients. Patients with LGG had better predictions (r=0.31-0.42) as compared to patients with HGG (r=0.03-0.11). This work reveals both the promising feasibility of and some challenges in applying CF in the creation of individual, precision functional brain maps in individual subjects including glioma patients.

Disclosures: **V. Tripathi:** None. **B.M. Hager:** None. **L. Rigolo:** None. **B.K. Bracken:** None. **A.J. Golby:** None. **Y. Tie:** None. **D.C. Somers:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.06

Topic: I.06. Computation, Modeling, and Simulation

Support: AdERC grant GA-340063
F.I.R.S.T. grant no. 1075/2013 from the Israel Science Foundation

Title: Using Markov Decision Processes to benchmark the performance of artificial and biological agents, and study the neural representation of the task

Authors: *A. KAZAKOV[1], M. M. JANKOWSKI[2], A. POLTEROVICH[3], J. NIEDIEK[3], I. NELKEN[4];
[1]ELSC - Edmond & Lily Safra Ctr. For Brain Scienc, ELSC - Edmond & Lily Safra Ctr. For Brain Scienc, Jerusalem, Israel; [2]Edmond and Lily Safra Ctr. for Brain Sci., The Hebrew Univ. of Jerusalem, Jerusalem, Israel; [3]Edmond and Lily Safra Ctr. for Brain Sci., Hebrew Univ. of Jerusalem, Jerusalem, Israel; [4]Hebrew Univ., Jerusalem, Israel

Abstract: When an animal is trained on a complex task, different task parts may be learned at different rates. Since reward is provided usually only at the end of each trial, it cannot be used to

infer within-trial learning trends. Behavioral features such as speed or trial duration capture trends in the animal's decision-making, but do not necessarily indicate that the animal is getting better at the task. We propose here an approach for studying learning of sub-parts of the task by a fine-level analysis of animal behavior. The task is modeled as a Markov Decision Process (MDP), making it possible to assign a value to each and every movement the rat performs while solving the task, and to determine the best decision the rat can take at each moment in time. By comparing the value of the best decision with the value of the action actually taken by the rat we get a time-resolved evaluation of rat behavior.We applied this approach to rats performing a sound localization task. In the behavior of real rats, we observed that (1) Rat behavior approached the optimal policy gradually throughout training; (2) most of the policy refinement occurred at a specific, short (<1s) segment of the trial; (3) the first trials of each day showed sub-optimal performance that improved during the session. Lastly, we modeled the rat using artificial agents guided by a deep neural network. We observed similar features of learning in the artificial agents as in the real rats. We then investigated how the task is encoded by the agent's deep neural network (DNN).Preliminary results indicate that the strongest connections between neurons were crucial for the precise network activity: The action accuracy of the network dropped by 50% when the strongest 5% of the weights are erased. However, the strongest weights were not sufficient for precise actions, since removing 40% of the smallest weights reduced accuracy by 20%. Interistingly, invariance to the rotational symmetry of the arena in which the rats were trained emergedin the DNN only in the last 2 layers. Further study of the information processing along the layers of the DNN will shed light on learning principles in artificial agents, and may be used to generate working hypotheses for learning in biological brains as well.

Disclosures: **A. Kazakov:** None. **M.M. Jankowski:** None. **A. Polterovich:** None. **J. Niediek:** None. **I. Nelken:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.07

Topic: I.06. Computation, Modeling, and Simulation

Support: NIMH F32 MH107159
NIMH R01-MH102840
DOD ARO W911NF-15-1-0426

Title: Estimating counterfactual spike trains under optogenetic interventions

Authors: *Z. SACCOMANO[1], S. A. MCKENZIE[2], D. F. ENGLISH[3], A. AMARASINGHAM[4];
[1]City Univ. of New York, New York, NY; [2]Neurosciences, UNM HSC, Albuquerque, NM; [3]Neurosci., Virginia Tech., Blacksburg, VA; [4]Mathematics, City Col. CUNY, New York, NY

Abstract: *Numerous studies - including ones using causal perturbations - have indicated that certain classes of monosynaptic interactions produce a precise, short-latency postsynaptic response. However, some distinct experimental studies suggest that in some circuits common input may be very fine timescale as well, raising the danger that fine-timescale correlations originating from common input may still masquerade as 'monosynaptic.'*
Here we examine the mechanisms of fine timescale synaptic interactions in biophysical models, fit models to data, and develop a method for inference, including estimators and confidence bounds for excitatory, inhibitory, and common cause inputs. The approach combines a timescale-based latent variable model with a causal potential outcome framework. Finally, we entertain the thought experiment that there are no temporal features distinguishing synaptic and common cause inputs in observational data, but show how the model gives distinct predictions for how correlations would change under distinct neural interventions (e.g., optogenetic silencing of common inputs versus silencing the synaptic type of a presumed direct connection). We note how this can, in turn, provide feedback to the model.

Disclosures: Z. Saccomano: None. **S.A. McKenzie:** None. **D.F. English:** None. **A. Amarasingham:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.08

Topic: I.06. Computation, Modeling, and Simulation

Support: NBRC Flagship program
BT/MEDIII/NBRC/Flagship/Program/2019:Comparative mapping of common mental disorders(CMD)over lifespan

Title: Using Mahalanobis distance as functional integration metric among brain network communities in Resting-state fMRI of patients with common mental health disorders.

Authors: *N. C. SASTRY[1], A. BANERJEE[2];
[1]Computat. Neurosci., Natl. Brain Res. Ctr., Gurugram, India; [2]Computat., Natl. Brain Res. Ctr., Gurgaon, India

Abstract: Evidence from neuroimaging studies have implicated altered functional connectivity and abnormal brain network topology measures in Schizophrenia. Mechanistically this may reflect an imbalance of functional integration and segregation which is increasingly agreed to be essential for the operation of distributed brain networks underlying cognitive function. In this study, we aim to characterise global functional integration of brain network communities, using Mahalanobis distance, a multivariate Euclidean distance metric, on fMRI dataset of 72 patients with schizophrenia and 74 healthy controls (COBRE Dataset). First, mean BOLD timeseries were extracted using a 400 region, 17 resting state networks (RSNs) Schaefer parcellation.

Functional connectivity (FC) was estimated by calculating Pearson correlation between brain regions and were subsequently r-to-z transformed. Subject-wise undirected, signed, weighted adjacency matrix was estimated considering brain regions as nodes and FC as edges. For each subject, in HC and SZ, we employ a data-driven generative model-based community detection approach called the weighted stochastic block model (WSBM) to cluster brain networks into communities. First, we systematically vary the number of communities detected from K=3-13. We select K=7 based on log-likelihood analysis. Next, we use an iterative consensus partitioning procedure, to derive representative communities at the group level. The iterative step is run for n=146 times, once for each subject and the algorithm converged at K=4 communities at the group level. Finally, we estimate global functional integration of brain network communities as Mahalanobis distance between communities. In the FC landscape, regions that are highly correlated can be visualised as hillocks and low-correlated regions as valleys. We calculate Mahalanobis distance between each brain parcel in one community with the distribution of brain parcels in other community, essentially calculating distance between hills and valleys. Low Mahalanobis distance indicates higher functional integration among brain network communities. Results show significant lower Mahalanobis distance between communities in SZ compared to HC, indicating an overall higher global functional integration in SZ. We use permutation-based t-test (B=10,000 permutations) to identify significant between-group differences. We compared Mahalanobis distance with other metrics used to characterise between-community interactions which further validates the efficacy. Thus, our study establishes the prospective use of Mahalanobis distance in identifying various disorders of mental health.

Disclosures: **N.C. Sastry:** None. **A. Banerjee:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.09

Topic: I.06. Computation, Modeling, and Simulation

Support: Subaward from the Vermont Biomedical Research Network under NIH award P20GM103449.

Title: Mathematical modeling approach to investigating inhibitory plasticity in the developing visual cortex

Authors: *J. CRODELLE;
Middlebury Col., Middlebury, VT

Abstract: The mammalian primary visual cortex (V1) contains neurons that respond preferentially to oriented visual stimuli. In some mammals such as rodents, these orientation-preferring neurons are scattered throughout V1 in a "salt and pepper" orientation-preference (OP) map, while the OP map is much more ordered in mammals such as the monkey and cat.

Underlying the formation of this OP map is the plasticity of synaptic connections between V1 cells, together with feedforward synapses from the thalamus. Disruptions in the activation of neurons along this pathway during development can modify the plasticity underlying the formation of the synapses that determine OP, potentially leading to abnormal sensory processing that occurs in some neurodevelopmental disorders. Our prior work has shown that gap-junction coupling among excitatory neurons in V1 can affect the plasticity of feedforward synapses into V1 and, consequently, the organization of the resulting OP map.
In our current work, we focus on the role of inhibition in forming the OP map. Inhibition is a crucial component of healthy neuronal networks; however, its spike-timing-dependent plasticity (iSTDP) is rarely treated realistically in computational models of synaptic development. This is due, in part, to the difficulties associated with measuring changes in inhibitory synaptic strengths and the vast number of different inhibitory interneuron populations. As a result of these difficulties, several different descriptions of iSTDP onto excitatory cells across the cortex have arisen, both from experiments and theoretical modeling work. We use a biologically-motivated mathematical model of the developing visual cortex to characterize the effect of these different iSTDP descriptions on the formation of feedforward synapses onto V1 cells, i.e., on the formation of its OP. In particular, we analyze two plasticity rules, one symmetric and one asymmetric, that give rise to different synaptic structures in the adult V1 network. This study will lend itself to advances toward a better understanding of the role of inhibitory plasticity in the formation of neuronal circuitries underlying computation in the cortex.

Disclosures: J. Crodelle: None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.10

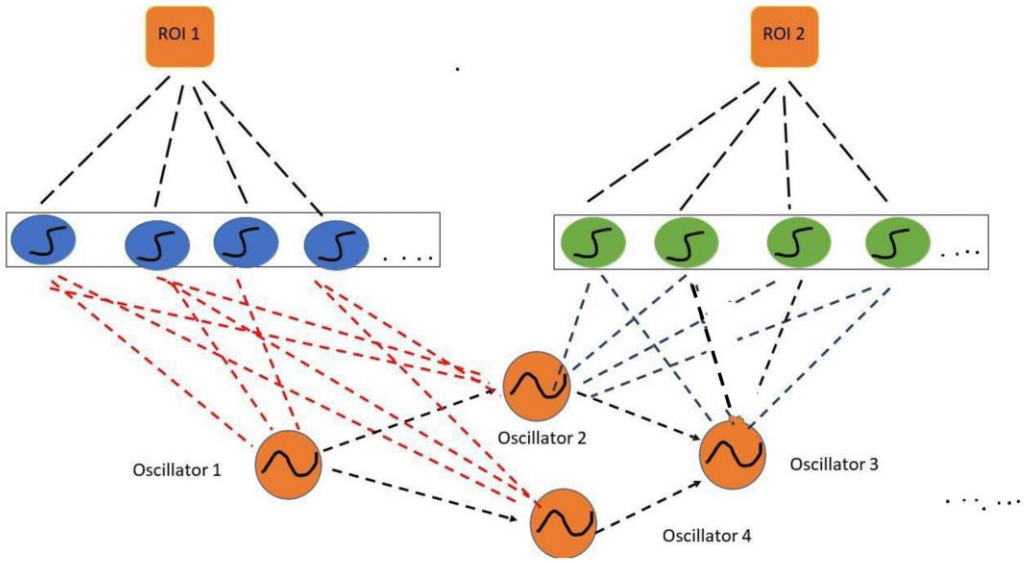
Topic: I.06. Computation, Modeling, and Simulation

Title: A trainable oscillatory neural network for modelling BOLD signals

Authors: *A. BANDYOPADHYAY[1], S. GHOSH[1], D. BISWAS[1], B. S[2], S. V. CHAKRAVARTHY[1];
[1]Dept. of Biotech., IIT Madras, Chennai, India; [2]IIIT Hyderabad, Hyderabad, India

Abstract: We propose a trainable oscillatory neural network for modelling Brain Oxygen Level Dependent (BOLD) signals from resting-state functional fMRI. In this network, a single oscillator represents a single Region of Interest (ROI). Similarly, in the output layer, there is a single output neuron for each ROI. The network has two components: a network of oscillators with internal connections and a feedforward network. Training is performed in 2 stages: in the 1st stage, the intrinsic frequencies of the individual oscillators and the coupling weights among them are trained. In the 2nd stage, the feedforward network consisting of two complex-valued weight stages with a hidden layer of complex sigmoidal neurons are trained using backpropagation of

complex-valued training error. Connections among the oscillators of the oscillatory layers are determined by the structural connectivity among the ROIs taken from diffusion MRI. Each oscillator along with its neighbours is connected to its corresponding output neuron via a hidden layer with 30 hidden neurons. Important to note that the amplitude of the complex coupling is fixed equal to the structural connectivity measurement, and the angle of the coupling is the trained angle from the first stage of learning. The experimental dataset is selected from an openly available resource [https://dx.doi.org/10.6084/m9.figshare.3749595.]. We only take the first participant's 1st session data of 1196 time points of approximately 15 minutes among the available 40 participants from the Human Connectome Project (HCP). BOLD signal of 160 ROIs is reconstructed with high accuracy after 5000 epochs with a mean Root mean square error (mean RMSE) of 0.035 and 0.0033 standard deviation. Another way to define the model's competency is to check the Pearson's Correlation Coefficient between the functional connectivity matrix from the model and the empirical data. After 5000 epochs of training, it is 0.97.
However, our ultimate motive is to generate a more biologically plausible model without shunning the accuracy achieved.



Disclosures: **A. Bandyopadhyay:** None. **S. Ghosh:** None. **D. Biswas:** None. **B. S:** None. **S.V. Chakravarthy:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.11

Topic: I.06. Computation, Modeling, and Simulation

Support: NHMRC Investigator Award 1196855

Title: Criticality in the developing zebrafish brain

Authors: M. H. MCCULLOUGH[1], Z. PUJIC[1], R. WONG[2], B. SUN[1], ***G. J. GOODHILL**[2,1]; [1]The Univ. of Queensland, Brisbane, Australia; [2]Washington Univ. in St Louis, SAINT LOUIS, MO

Abstract: It has been hypothesized that the brain may sometimes operate in a critical state, where the activation of one neuron leads on average to the activation of one other neuron. In principle such neural criticality would confer computational advantages including optimization of dynamic range, information transmission and storage capacity. There is evidence for criticality from several different neural systems and species, including the larval zebrafish. Here we used volumetric 2-photon calcium imaging in zebrafish larvae at a range of ages to demonstrate the following. (i) Markers of criticality in neural dynamics change over development. (ii) Criticality markers are region-specific, so that criticality at the whole-brain level may not be a reliable guide to behavior at a regional level. (iii) Criticality markers are altered in fish mutant for fmr1, mutations in which are the most common inherited form of autism-spectrum disorder in humans. Together these results suggest that neural criticality may be better understood at a regional rather than whole-brain scale and that stage of development plays a significant role. However, the finding that markers of criticality were altered in an autism mutant supports the hypothesis that criticality may be important for brain function.

Disclosures: M.H. McCullough: None. **Z. Pujic:** None. **R. Wong:** None. **B. Sun:** None. **G.J. Goodhill:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.12

Topic: I.06. Computation, Modeling, and Simulation

Title: Estimating peak isometric muscle force in elbow muscles via hierarchical Bayesian modeling

Authors: *R. JOHNSON, Y. DARMON, R. KUAN, D. YE, J. FINLEY, N. SCHWEIGHOFER;
USC, USC, Los Angeles, CA

Abstract: Musculoskeletal models are useful to estimate individual muscle forces, as these forces are impossible to measure empirically. These models have been used to understand the cause of motor impairments in clinical populations. However, the validity of muscle force estimates depends on accurate model parameters such as peak isometric muscle force (F_0). Previous studies have used Maximum Likelihood Estimation (MLE) or muscle imaging to estimate model parameters. However, because these methods return point estimates, parameter uncertainty is unknown, and it is unclear how to incorporate previous knowledge in the model. Here, we propose a hierarchical Bayesian modeling approach to estimate muscle parameters of individual subjects. Unlike MLE, Bayesian modeling assumes prior parameter distributions and updates these priors into posterior distributions based on the data with Bayes' Rule. In addition, the hierarchical structure allows "borrowing" of knowledge between participants, which allows for faster and more accurate parameter estimation. First, we tested the feasibility of using Hierarchical Bayesian modeling to estimate F_0 during a simulated upper limb isometric strength assessment using a model of the elbow joint with five muscle actuators (3 flexors; 2 extensors) with a Markov Chain Monte Carlo (MCMC) algorithm (implemented in JAGS). We simulated participants with a range of F_0, tendon stiffness, and anthropometry parameters generated from normal distributions around population means. We then solved an optimization procedure to compute muscle activations during a series of isometric elbow conditions. Then, using a hierarchical Bayesian model, we sought to recapture the F_0 for each simulated participant by using muscle activations with realistic signal-dependent noise as an input and elbow torque as the output. The model was able to accurately recapture F_0 for each participant and the population (isometric torque RMSE < 1 Nm). We then applied the same hierarchical model to estimate F_0 in a set of experimental data from 15 young, healthy participants. We collected isometric torque and associated electromyography data from a series of elbow angles and torque magnitudes. Although the model fit had greater errors (RMSE = 2-6 Nm) and the posterior distributions were wider than in simulations, the estimated F_0 was physiologically plausible. In current work, we are developing more complex models to better predict the isometric torque. In the future, we intend to apply this workflow to estimate the muscle properties in people post-stroke, which will help us understand the source of motor impairments and track their recovery.

Disclosures: R. Johnson: None. **Y. Darmon:** None. **R. Kuan:** None. **D. Ye:** None. **J. Finley:** None. **N. Schweighofer:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.13

Topic: I.06. Computation, Modeling, and Simulation

Support: Cognitive Sciences at the University of California, Irvine
Canadian Institute for Advanced Research Azrieli Global Scholars Fellowship in Brain, Mind, & Consciousness

Title: Cograph: exploring the latent semantic structure and conceptual evolution of the neurosciences, cognitive sciences, and ai

Authors: *A. G. HANSEN[1], A. PRADESH[2], J. VANDEKERCKHOVE[1], M. A. K. PETERS[1]; [1]Cognitive Sci., Univ. of California - Irvine, Irvine, CA; [2]Electrical Engin., Indian Inst. of Technol. - Palakkad, Kozhippara, India

Abstract: The scientific ecosystem that spans the rapidly coevolving and dynamically interacting fields of Neuroscience, Cognitive Science, and AI has generated unprecedented advances over the past several decades. These have come not only in the form of enormous leaps in applications and globally disruptive technologies, but (repeatedly) in terms of how we interrelate the core concepts which constitute the body of knowledge itself. Acknowledging this, there is substantial motivation to develop techniques which allow for robust inference from the generating dynamics underlying such advances, in order to 1) make predictions about future states of research, 2) reliably identify areas with high-impact potential, and 3) understand the evolution of conceptual frameworks. We present CoGraph, a data- and knowledge base constructed from proceedings of many leading conferences in Neuroscience, Cognitive Science, and AI. CoGraph provides a basis to generate temporal co-occurrence networks and temporal knowledge graphs, and address these three aims. It spans over four decades of research, collectively encompassing tens of thousands of conference publications including from COSYNE, the Cognitive Science Society, CCN, NeuroIPS, MathPsych, and many others. Furthermore, the corpus includes conference, publication, author, and affiliation data, as well as titles, keywords, abstracts, and extracted textual entities. Given that conferences embody bleeding edge trends in research, serve as public forums for the multidisciplinary exchange of ideas, and do so with relatively low latency, we believe that they represent ideal candidates of study. We highlight the scope and utility of CoGraph, by demonstrating the breadth and depth of the temporal co-occurrence networks and temporal knowledge graphs generated from the data therein, thousands of nodes and millions of edges, which allow for the tracking of neurocentric trends, and co-occurring concepts and conceptual relations related to brain and mind. We then compare and contrast subgraphs of each, which are relevant to understanding important trends in the temporal evolution of concepts in neuroanatomy, neurophysiology, neurohistology, neurocomputation, cognitive neuroscience, as well as related methodologies and techniques. Our exploratory work provides a basis for studying the coevolving structures underlying the collective emergence of novel ideas and research trajectories, and for assessing the merit of scientific contributions to an ever-broadening ontological horizon—both to explore the space of conceptual content itself and to analyze its convergence and divergence across time.

Disclosures: **A.G. Hansen:** None. **A. Pradesh:** None. **J. Vandekerckhove:** None. **M.A.K. Peters:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.14

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH R01 NS120581
NSF DMS-1951165
NIH R01 EB022862
NSF DMS-1951599

Title: Sequence control of quadruped gaits using combinatorial threshold-linear networks

Authors: *J. LONDONO ALVAREZ[1], K. MORRISON[2], C. P. CURTO[1];
[1]Mathematics, The Pennsylvania State Univ., University Park, PA; [2]Sch. of Mathematical Sci., Univ. of Northern Colorado, Greeley, CO

Abstract: Neural integration and sequential pattern generation underlie many complex behaviors, and various architectures have been proposed to model them. Still, two challenges remain: robustness to noise (particularly for neural integrators) and coexistence of different attractors in the same circuit (e.g., to model multiple locomotive gaits). Here we model these disparate neural functions with Combinatorial Threshold Linear Networks (CTLNs), which are firing rate models with binary synapses and simple perceptron-like neurons. The dynamics of CTLNs are controlled solely by the structure of a directed graph, thus isolating the role of connectivity. Moreover, CTLNs are piecewise-linear, providing a mathematically tractable framework well suited for engineering circuits with prescribed attractors. We present three examples of neural integration and pattern generation. First, we propose a CTLN counter that counts the number of input pulses via the position of an attractor in a linear chain; a small variation on this network yields a signed counter that tracks the total number of signed (L/R) pulses on a number line. Simulations reveal that these counters are robust to noise. Next, we present a CTLN capable of reproducing five quadrupedal gaits: bound, pace, trot, pronk, and walk. These gaits coexist as distinct limit cycle attractors in the same network without changing parameters, allowing rapid transitions between gaits by stimulating single neurons. As with all CTLNs, the neurons do not intrinsically oscillate and so the periodic behavior of limit cycles is a result of connectivity alone. Finally, to illustrate a more general construction of a circuit controlling a sequence of attractors, we combine the basic counter with the 5-gait network in a layered CTLN (Fig. 1A-B). A sequence of gaits is encoded in the wiring between the counter and the 5-gait network, and each new incoming pulse advances the counter and activates the next gait in the sequence (Fig. 1C). While the timing of the sequence is determined by external pulses, the sequence itself is fully encoded in the network.

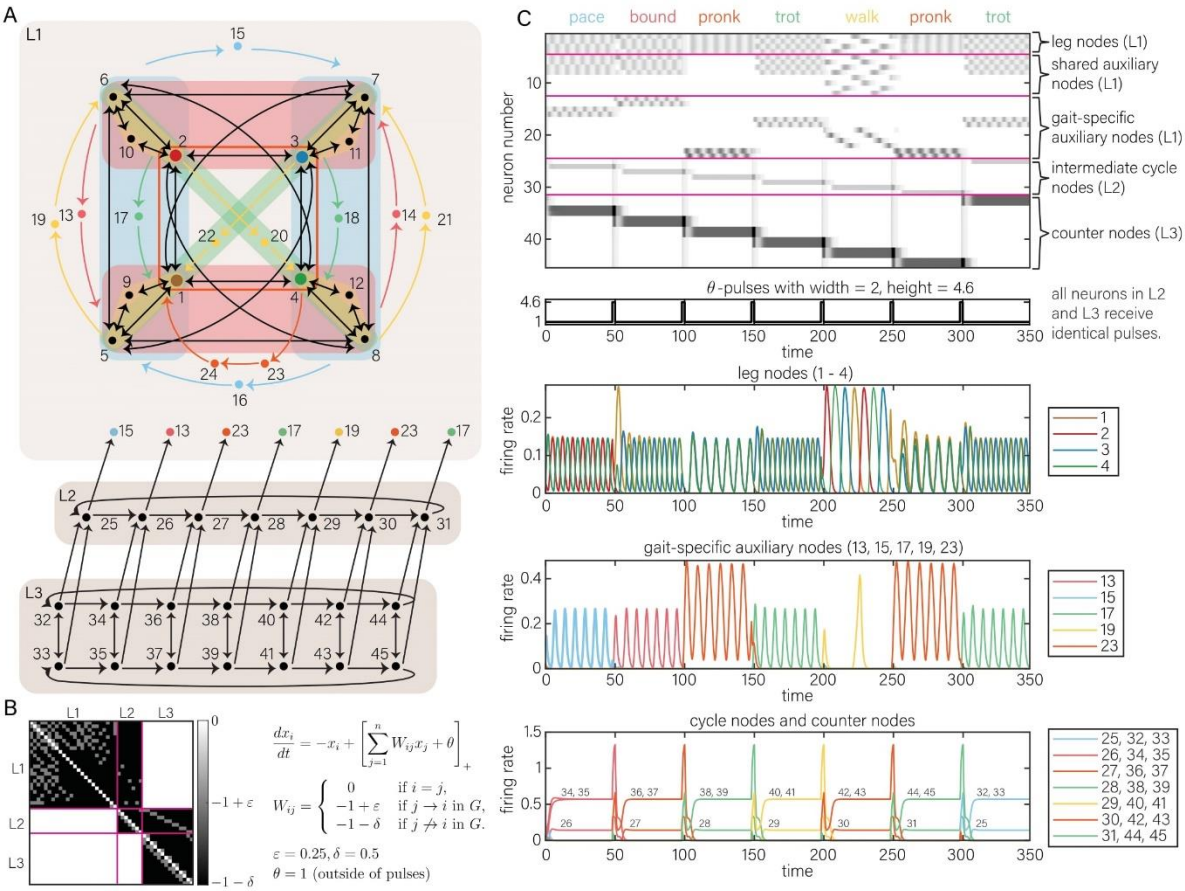


Figure 1. (A) Layered network for sequential control. L1 consists of the 5-gait network, auxiliary nodes corresponding to different gaits are redrawn on the bottom; the order in which they are drawn specifies the order in which each gait will be accessed. L2 is an intermediate layer consisting of a 5-cycle. L3 is the unsigned counter network. (B) Equations of CTLNs and the matrix W used for the layered network in panel A. The only inhibition between layers is from L2 to L1 and from L3 to L2. (C) Greyscale, pulses and rate curves for network in panel A. On top of the greyscale is the order in which gaits are accessed, matching those specified in panel A.

Disclosures: **J. Londono Alvarez:** None. **K. Morrison:** None. **C.P. Curto:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.15

Topic: I.06. Computation, Modeling, and Simulation

Support: NIHM Grant R01 MH121009
NIMH Grant 1R56 MH11568
NIH Grant T32 GM132008
NSF Award DMS-1937229

Title: Context-dependent covariance patterns reveal subnetworks in the amygdala

Authors: *A. N. AUCOIN[1], A. B. MARTIN[2], K. K. LIN[3,1], K. M. GOTHARD[4];
[1]Program in Applied Math, [2]Physiol., [3]Mathematics, [4]Col. of Med., Univ. of Arizona, Tucson, AZ

Abstract: The amygdala is known to be responsible for extracting from external stimuli their social and emotional significance. Context during stimulus delivery, which changes over longer timescales than rapidly changing stimuli, is crucial to determining the social importance of the stimuli. One way the amygdala might encode these persistent states is through changes in context-dependent coactivity patterns between populations of cells within and between amygdala nuclei. To explore this possibility, we analyzed local field potential (LFP) data recorded from the amygdala of three macaque monkeys presented with alternating blocks of social and non-social tactile stimuli. The social tactile stimulus, designed to mimic grooming, was delivered by a trusted human handler using gentle sweeps across the muzzle or brow of the monkey. The non-social tactile stimulus was a machine-delivered gentle air puff which targeted the same locations of the skin. We recorded LFP data from linear arrays with 32 contacts inserted into the amygdala and analyzed the data using Generalized Eigen-Decomposition (GED), a robust framework useful for dimension reduction and source separation in multivariate electrophysiological data. GED provides a set of weights, i.e., a spatial filter, for the recorded channels that maximizes the difference in coactivity patterns between two specified states, in this case the difference between social versus non-social baseline (pre-stimulus) LFP signals. The GED analysis yielded spatial filters that extracted subnetworks of similarly covarying channels corresponding to nuclei in amygdala. Although the GED analysis was blind to information about the amygdala anatomy, the subnetworks identified by the algorithm aligned well with anatomical boundaries in the amygdala as reconstructed from structural MRI. Our results indicate that the organization of these subnetworks is context-dependent, with statistically separable spatial filters that changed between social and non-social blocks, suggesting that the amygdala can represent persistent states through context-dependent organization of coactivity across its nuclei.

Disclosures: **A.N. Aucoin:** None. **A.B. Martin:** None. **K.K. Lin:** None. **K.M. Gothard:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.16

Topic: I.06. Computation, Modeling, and Simulation

Support: NIDCD R01DC012947
NIBIB U24EB028998
NIMH P50MH109429
NIBIB U01EB017695
NYS DOH01-C32250GG-3450000
NSF 190444
Army Research Office W911NF-19-1-0402

Title: Analyzing the origin of cortical oscillation events in a data-driven biophysical model of the primate auditory thalamocortical system

Authors: *E. Y. GRIFFITH[1,3], S. DURA-BERNAL[2,3], A. BARCZAK[3], M. N. O'CONNELL[3], T. M. MCGINNIS[3], C. E. SCHROEDER[4,3], W. W. LYTTON[2,5], P. LAKATOS[3,6], S. A. NEYMOTIN[3,6];
[1]Neural & Behavioral Sci., [2]Physiol. and Pharmacol., SUNY Downstate Hlth. Sci. Univ., Brooklyn, NY; [3]Ctr. for Biomed. Imaging and Neuromodulation, Nathan S. Kline Inst. for Psychiatric Res., Orangeburg, NY; [4]Depts. Psychiatry and Neurol., Columbia Univ. Col. of Physicians and Surgeons, New York, NY; [5]Neurol., Kings County Hosp. Ctr., Brooklyn, NY; [6]Psychiatry, NYU Grossman Sch. of Med., New York, NY

Abstract: We developed a biophysically-detailed model of the macaque auditory thalamocortical pathway, with medial geniculate body (MGB), thalamic reticular nuclei (TRN), and a column of primary auditory cortex (A1). This model used the NEURON simulator and NetPyNE modeling tool to integrate information at the subcellular, cellular, and circuit-level scales, from synapse characteristics to cell electrophysiology to long-range, local and dendritic connectivity. We used this model to reproduce cortical oscillations observed in macaque non-human primate (NHP) A1. We found that oscillations emerged spontaneously in the model and were comparable to those recorded in vivo. Individual oscillation events were detected in current source density (CSD) data from in silico and in vivo resting state recordings. These events were then classified by laminar region (supragranular, granular, infragranular) and frequency band (delta, theta, alpha, beta, gamma). To see if we reproduced physiologically realistic oscillation events in silico, we compared the duration, number of cycles, and peak frequency of oscillation events in the model and NHP datasets. These properties showed similar average values and overlapping distributions across frequency bands and laminar regions. We also compiled several examples of individual oscillation events from model and NHP which matched across all of these features. Having reproduced realistic oscillation events in silico, we used a supragranular theta oscillation event to demonstrate that the model can decipher the contributions of distinct neuronal populations to the overall CSD signal. This analysis revealed that the layer 4 spiny stellate and pyramidal tract cells, and the layer 5A intratelencephalic cells (IT5A), made the strongest contributions to the CSD signal during the oscillation. The contribution of IT5A was particularly interesting, since IT5A cell bodies were located 350-650 um below the electrode where the oscillation was recorded, suggesting that IT5A apical dendrite currents generated a substantial component of the detected oscillation. We also examined the corresponding spiking activity in these populations, and observed gamma band activity in the spike rate spectrograms, demonstrating a cross-frequency interaction often observed in oscillations, but with the added benefit of cell-type specificity. Overall, our model provides a valuable framework for integrating and reproducing experimental data in auditory circuits. Here we demonstrate this with respect to cortical oscillation events, and highlight how the model's biological detail can be used to examine the origins of complex cortical activity.

Disclosures: E.Y. Griffith: None. **S. Dura-Bernal:** None. **A. Barczak:** None. **M.N. O'Connell:** None. **T.M. McGinnis:** None. **C.E. Schroeder:** None. **W.W. Lytton:** None. **P. Lakatos:** None. **S.A. Neymotin:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.17

Topic: I.06. Computation, Modeling, and Simulation

Support: John Templeton Foundation #61283
Fetzer Institute, Fetzer Memorial Trust #4189

Title: An investigation of the relationship between fMRI and MEG functional connectivity using data from the Human Connectome Project

Authors: *D. LIANG[1], M. LANDRY[2], U. MAOZ[1], A. SCHURGER[1];
[1]Brain Inst., Chapman Univ., Orange, CA; [2]McGill Univ., Montréal, QC, Canada

Abstract: Neuroscience research is increasingly focused on understanding how networks of regions underpin cognitive processes rather than focusing on the role of specific brain regions. These networks are often studied using functional connectivity, a technique that investigates the relationships between neural time-series. Magnetoencephalography (MEG) and functional magnetic resonance imaging (fMRI) are amongst the most well-established non-invasive neuroimaging approaches to derive functional connectivity. These modalities are complementary to each other (Hall et al., 2014). Therefore, combining them holds the promise of utilizing both the high temporal resolution of MEG and the high spatial resolution of fMRI, thereby opening new research avenues. In this regard, evidence suggests that the two modalities measure similar brain activity—the local field potential (LFP). However, a direct comparison of MEG and fMRI data is difficult, largely due to the multifaceted nature of the MEG signal (Hall et al., 2014). Indeed, the possibility to decompose MEG across time and frequency domains allows the exploration of multiple features. Moreover, the analysis of MEG signals in source space is proven to be reliable, which allows for the comparison of MEG and fMRI. It nevertheless remains poorly understood how and to what extent (if any) the MEG signal matches up with the corresponding fMRI signal. Here we used both resting-state MEG and fMRI data of 89 human subjects from the Human Connectome Project (HCP) to investigate the relation of the functional connectivity metrics between the two modalities. We compared the MEG source-localized ROI-to-ROI amplitude envelope correlation (AEC), weighted phase lock index (wPLI) and phase lock value (PLV) with the fMRI functional connectivity. Our preliminary results with 10 subjects show that the PLV (zero-lag non-adjusted) reflects similar structures (e.g., the cross-hemispheric connectivity) to the fMRI functional connectivity, whereas wPLI (zero-lag adjusted) and AEC (band-passed instantaneous power) show significantly different patterns. This result suggests that zero-lag phase synchrony, often considered as volume conduction artifacts in MEG, may contain some information about neural resting-state activity captured by fMRI. We thus provided empirical evidence that MEG connectivity metrics and fMRI functional connectivity have some similarities; however, one cannot fully explain the other and the result might heavily depend on the MEG analysis method.

Disclosures: **D. Liang:** None. **M. Landry:** None. **U. Maoz:** None. **A. Schurger:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.18

Topic: I.06. Computation, Modeling, and Simulation

Support: R01MH122622
R01MH110212

Title: Principal component and network analyses reveal relationships between sex, social status, oxytocin receptor, vasopressin V1a receptor, and serotonin 1A receptor densities across the social decision-making network

Authors: *J. H. TAYLOR[1], Z. A. GRIEB[1], A. P. ROSS[1], A. NORVELLE[1], V. MICHOPOULOS[2], K. L. HUHMAN[1], H. E. ALBERS[1];
[1]Georgia State Univ. Neurosci. Inst., Georgia State Univ. Neurosci. Inst., Atlanta, GA; [2]Emory Univ., Decatur, GA

Abstract: The social decision-making network (SDMN) has been a useful concept for examining the roles of interconnected nodes in the expression of social behavior. In order to understand these brain networks, it is necessary to describe the relationships between nodes and to relate connections and patterns within the network to distinct social behavioral states, such as sex or dominance status. In this study, we used graph theory network analysis (NA) and principal component analysis (PCA) to analyze oxytocin (OTR), vasopressin (V1a), and serotonin 5HT1a receptor binding data from 14 regions across the SDMN with the purpose of elucidating novel receptor expression networks and relationships. To investigate differences based on sex and social status (dominant, subordinate, nonsocial control) we extracted PCA scores and performed 2(sex) x 3(social status) ANOVAs using these data as dependent variables. Three PCA components accounted for nearly 50% of the variance. Component 1 was dominated by positive loadings from OTR nodes and V1a nodes within the mesolimbic dopamine system. Component 2 was more heterogenous, and was marked by strong loadings from V1a and 5HT1a in the AH and the MPOA. Males loaded significantly more highly than females on this component. Component 3 was dominated by V1a nodes, particularly those within the mesolimbic dopamine system. Our NA revealed similar and complementary results. OTR nodes represented 60% of the top 25% of nodes. The three most central nodes were OTR in the paraventricular nucleus, the bed nucleus of the stria terminalis, and the medial prefrontal cortex (mPFC). Despite the high centrality of the mPFC with regard to OTR expression, the nodes representing V1a and 5HT1a in the mPFC were among the least central. The NA between males and females showed similar patterns of centrality among nodes. Notable differences include OTR in the anterior hypothalamus, which was the ninth-most central node for females, but 22nd for males, and OTR in the medial preoptic area, which was the 12th-most central node for females, but 25th for males. Node centralities were largely similar between dominants and subordinates. Notable exceptions were V1a in the central amygdala, which was the 35th-most central node for dominants, but was 21st for subordinates.

These data show that, in Syrian hamsters, OTR expression in nodes in the SDMN are tightly coupled, and V1a and 5HT1a expression in these nodes differ between males and females but not between hamsters with differing dominance statuses. Supported by R01MH122622 and R01MH110212 to HEA and KLH. The content is solely the responsibility of the authors and does not necessarily represent the official views of the NIH or GSU.

Disclosures: **J.H. Taylor:** None. **Z.A. Grieb:** None. **A.P. Ross:** None. **A. Norvelle:** None. **V. Michopoulos:** None. **K.L. Huhman:** None. **H.E. Albers:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.19

Topic: I.06. Computation, Modeling, and Simulation

Support: The Post-K Exploratory Challenge 4–1 and Program for Promoting Research on the Supercomputer Fugaku (hp200139).
Program for Promoting Researches on the Supercomputer Fugaku (hp210169)
Grant-in-Aid for Transformative Research Areas (JP21H05137)

Title: A Spiking Neural Network Simulation of Layered Sheet of Cortico-Cerebello-Thalamic Circuit at Human-Scale

Authors: *J. IGARASHI[1], T. YAMAZAKI[3], H. YAMAURA[3], K. NOMURA[4], Z. SUN[2], C. GUTIERREZ[5], K. DOYA[5];
[1]Ctr. for Computat. Sci., [2]Ctr. for Advanced Photonics, RIKEN, Wako city, Japan; [3]Grad. Sch. of Informatics and Engin., The Univ. of Electro-Communications, Chofu city, Japan; [4]Ctr. for Planetary Sci., Kobe Univ., Kobe, Japan; [5]Neural Computation Unit, Okinawa Inst. of Sci. and Technol., Onna Village, Japan

Abstract: Human-scale whole-brain simulation is the ultimate tool for understanding the mechanisms of information processing and disorders in the human brain. However, a human-scale brain simulation containing major brain regions of the cerebral cortex and cerebellum has not been achieved due to insufficient computational resources and adequate parallelization methods.
To overcome the issues, we tested the effectiveness of exascale computing using the Supercomputer Fugaku for simulations of a cortico-cerebello-thalamic circuit.
The supercomputer Fugaku has 158,976 ARM-based CPUs and 4.85 petabytes of memory. The CPU has 48 compute cores, each of which has two SIMD units of 512-bit length. The theoretical performance is about one exaFLOPS of in single precision.
We used our in-house spiking neural network MONET simulator that performs a tile partitioning method and communication at an interval of half signal transmission delay. We introduced a new data structure to it to utilize the many-core architecture and SIMD computing.

We evaluated the performance using spiking neural network models of the cerebral cortex [1], cerebellum [2], and thalamus that we have developed based on anatomical and physiological studies. The numbers of layers and neuron types were 5 and 18 for the cerebral cortex and 7 and 9 for the cerebellum, respectively. We used a leaky integrate-and-fire neuron model for all neuron models.
We examined weak scaling performance for the three models of the cerebral cortex, cerebellum, and cortico-cerebello-thalamic circuit using 1024 to 150,544 compute nodes of the Fugaku. At the maximum scales of models using 150,544 compute nodes, the Fugaku achieved simulations of the cerebral cortex with 24.5 billion neurons, the cerebellum with 124.8 billion neurons, and the cortico-cerebello-thalamic circuit with 44.5 billion neurons. In the three models, the elapsed times for one second of biological time at 150,544 compute nodes were 2.9, 13.1, and 11.8 seconds which increased by less than 21 % compared to those of 1,024 compute nodes. The results demonstrated the scaling-up of the models with limited overheads.
The Fugaku performed simulations of the cerebral cortex and cerebellum about 160 times and 40 times faster than the K computer that had 11 petaFLOPS of theoretical performance when the same sizes of the models were set per one compute node.
These results suggest that exascale computing will be effective for a human-scale brain simulation and open new doors for studying the human brain in neuroscience.
[1] J. Igarashi, et al., Frontiers in Neuroinformatics, vol. 13, p. 71, 2019.
[2] H. Yamaura, et al., Frontiers in Neuroinformatics, vol. 14, p. 16, 2020.

Disclosures: **J. Igarashi:** A. Employment/Salary (full or part-time):; full. **T. Yamazaki:** A. Employment/Salary (full or part-time):; full. **H. Yamaura:** None. **K. Nomura:** None. **Z. Sun:** A. Employment/Salary (full or part-time):; full. **C. Gutierrez:** A. Employment/Salary (full or part-time):; full. **K. Doya:** A. Employment/Salary (full or part-time):; full.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.20

Topic: I.06. Computation, Modeling, and Simulation

Support: R01 NS104898-01

Title: Modeling traveling waves and propagating spatio-temporal patterns of cortical excitability in the primary motor cortex

Authors: *L. BACHSCHMID-ROMANO[1], N. G. HATSOPOULOS[2], N. BRUNEL[3];
[1]Duke Univ. Sch. of Med., Durham, NC; [2]Univ. of Chicago, Chicago, IL; [3]Neurobio., Duke Univ., Durham, NC

Abstract: LFP oscillations in the beta range are prominent in various areas of the motor system, and in the primary motor cortex of nonhuman primates they have been shown to propagate as

planar traveling waves along a rostro-caudal axis. Single-trial analysis showed that immediately prior to movement onset the amplitude of these beta oscillations attenuates, and the time of beta attenuation follows a spatial gradient on the cortical sheet along a similar rostro-caudal axis, suggesting that propagating patterns of cortical excitability are a signature of movement initiation. These beta attenuation patterns are distinct from the beta waves observed during movement preparation, and it is unclear whether these two phenomena are related. Here, we developed a network of leaky integrate and fire excitatory and inhibitory neurons with spatially-dependent connectivity as a model of the motor cortical sheet. We characterize the dynamical regimes that the model exhibits for different system parameters and we identify the parameters that allow the model to reproduce salient features of multielectrode recordings from the primary motor cortex of monkeys performing an instructed-delay, reaching task: the population rate dynamics, the typical LFP autocorrelation function, and the LFP cross-correlation profiles as a function of the distance between channels. For our choice of the synaptic couplings parameters, the LFPs oscillate with frequency in the beta range, individual cells fire irregularly, and traveling waves are observed. While a model with isotropic connectivity produces traveling waves that propagate either as planar or radial waves in any direction, after introducing anisotropy in the connectivity we observe predominantly planar waves traveling along a specific direction. If the strength of an homogeneous external input to the network is increased, the beta amplitude profile is reduced. Interestingly, as the strength of the external input increases, the model reproduces a spatial pattern of beta attenuation times consistent with experimental data. Overall, our model investigates mechanisms for the emergence and suppression of traveling waves in the beta range in the motor cortex and shows that traveling waves spontaneously emerge in a neuronal network with heterogeneous and spatially dependent synaptic connections when the network is in a globally oscillating regime, and that spatial patterns of beta attenuation at moment initiation can be a signature of a rapidly increasing and spatially homogeneous external input.

Disclosures: L. Bachschmid-Romano: None. **N.G. Hatsopoulos:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); N.G.H. serves as a consultant for BlackRock Microsystems, Inc., the company that sells the multi-electrode arrays and acquisition system used in this study. **N. Brunel:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.21

Topic: I.06. Computation, Modeling, and Simulation

Support: KAKENHI Grant 18H05213
KAKENHI Grant 19H04994

Title: Astrocytes facilitate self-organization and remodeling of cell assemblies under STP-coupled STDP

Authors: *R. KOSHKIN[1], T. FUKAI[2];
[1]Okinawa Inst. of Sci. and Technol., Okinawa Inst. of Sci. and Technol., Tancha, Japan;
[2]Okinawa Inst. of Sci. and Technol., Onna-son, Japan

Abstract: Cell assemblies - graph-like structures of neurons - are widely believed to represent the physical substrate of memory. Although it is well established that long-term plasticity (LTP) is crucial for cell assembly formation, little is known about how the emergence, reorganization and behavior of cell assemblies depends on other factors including astrocytes and short-term plasticity (STP). Here, we investigated the self-organization of cell assemblies in the absence of structured sensory input in a recurrent network model that mimics the area CA3 of the hippocampus. In our model, recurrent connections are modifiable by the symmetric STDP that was observed in CA3 (Mishra et al., 2016). Furthermore, our model incorporates two novel features into the synaptic learning rule. (1) Weight changes at excitatory synapses depend on the releasable amount of transmitters at the presynaptic terminals. In a computational model, the STP-dependent STDP was previously shown to facilitate goal-directed sequence learning through reverse replay (Haga & Fukai, 2018). (2) We implicitly modeled the effect of astrocytes on the network by introducing large variability into the neurotransmitter release probability. It was recently shown that the blockade of NMDA receptors on astrocytes significantly narrows the release probabilities of synapses on single neurons (Chipman et al. 2021). Our simulations reveal that both STP-dependent and STP-independent symmetric STDP supports the emergence of robust cell assemblies in the network that receives no structured input. Interestingly, however, the STP-dependent STDP, but not the STP-independent one, confers two computationally desirable properties on the spontaneous activity of the model. First, it develops a more stable cell assembly structure that better maintains self-similarity across time. Second, the model becomes more responsive to external stimulation, making it easier for new stable cell assemblies to emerge. Finally, we found that the presence of astrocytes facilitates both self-organization and stimulus-driven reorganization of the network. Our results suggest additional computational benefits of STP-dependent STDP and highlight a previously unknown regulatory function of astrocytes in memory formation.

Disclosures: R. Koshkin: None. **T. Fukai:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.22

Topic: I.06. Computation, Modeling, and Simulation

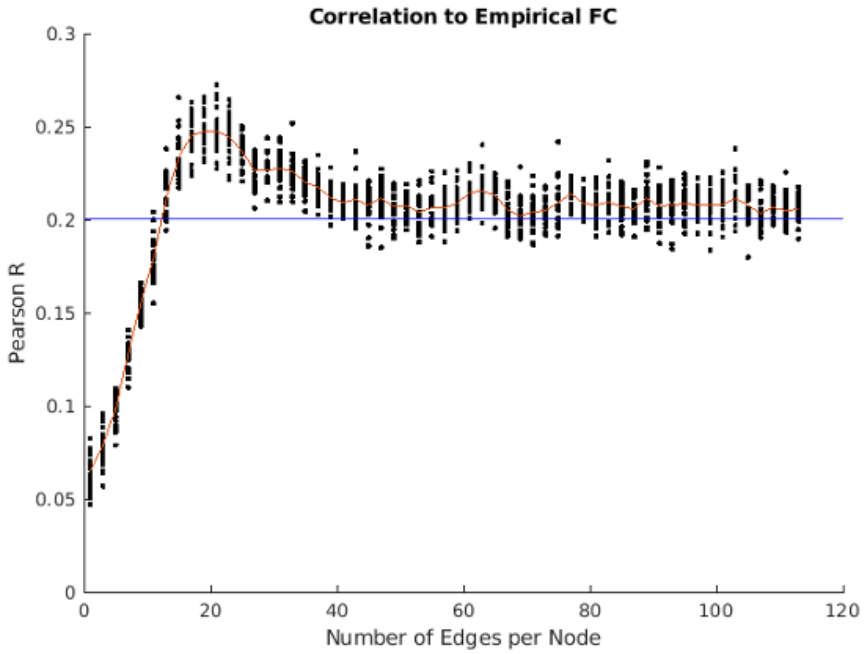
Support: NSF-NRT grant 1735095, Interdisciplinary Training in Complex Networks and Systems

Title: Implementing Communication Dynamics in a Large-Scale Computational Model of Human Cortex

Authors: *M. POPE, C. SEGUIN, O. SPORNS;
Indiana Univ., Bloomington, IN

Abstract: Both dynamic models and communication strategies have been proposed to provide a link between brain structure and function. The Kuramoto oscillator model is frequently used to model whole-brain neural dynamics as measured with BOLD fMRI. We argue that the Kuramoto model allows *diffusive* dynamics to unfold on a static network but is not designed to capture more efficient strategies for signal traffic described in communication models. We propose a modification to the traditional Kuramoto model. The proposed model limits causal influence to a set of 'active edges' by choosing the *m* most phase-synchronized neighbors of every node on each time step. Thus, diffusive dynamics are limited, and edges are turned on or off in accord with the local dynamics of the system. We simulate our model with a subject-averaged (n = 95) structural network from the Human Connectome Project across all possible *m* values. The simulated functional connectivity (FC) produces a better fit with subject-averaged empirical FC (mean r = 0.25, std=0.009, 24 simulations) than the standard Kuramoto model (mean r = 0.20, std=0.008) (Fig. 1) when m=20 edges per node, far less than the mean degree of the structural network (60 edges). Further analysis reveals that model performance varies under different edge-selection regimes and that edges within structural modules remain turned on while edges between structural modules are more dynamic. Findings replicate in an independent dataset. We discuss how, using node-level information, the model allows the network structure to respond to momentary fluctuations in system dynamics, and so represents a unification of perspectives from dynamic modeling and communication strategies.

Fig 1. 24 simulations of the proposed model were performed for each value of *m*, ranging from 1 to the max. degree of the structural matrix. Time-series were convolved with a haemodynamic response function and FC was correlated with subject-averaged empirical FC. Each point represents one run of the model. The blue line marks the correlation value for the standard Kuramoto model (avg. over 24 simulations).



Disclosures: **M. Pope:** None. **C. Seguin:** None. **O. Sporns:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.23

Topic: I.06. Computation, Modeling, and Simulation

Support: NSF GRFP

Title: Using graph theory to predict pattern completion neurons in a computational model of the visual cortex

Authors: ***C. BAKER**, Y. GONG;
Duke Univ., Duke Univ., Durham, NC

Abstract: Neural ensembles are found throughout the brain and are believed to underlie diverse cognitive functions including memory and perception. Methods to activate ensembles precisely, reliably, and quickly are needed to further study their role in cognitive processes. Previous work has found that ensembles in L2/3 of the visual cortex (V1) exhibit pattern completion properties in a stochastic manner: ensembles composed of tens of neurons can be activated by stimulation of just two neurons, but previous methods of identifying pattern completion neurons resulted in an ensemble recall rate of only 5%. In this study, we optimized selection of pattern completion neurons using a graph theory approach. We developed a computational model that replicated the structural and electrophysiological properties of different cell types in L2/3 in mouse V1. We identified ensembles of densely connected excitatory neurons in this model using k-means clustering. We then stimulated multiple pairs of neurons in the same ensemble while tracking the activity of the rest of the ensemble and the ensemble recall rate. To quantify a pair's power to activate an ensemble independent of the network's pre-stimulation state, we defined a novel metric called pattern completion capability (PCC), the mean pre-stimulation voltage needed for a pair of neurons to have a 5% ensemble recall rate. We found that PCC was directly correlated with multiple graph theory parameters and that LASSO regression could accurately predict PCC from these features. Of these parameters, degree and closeness centrality had the most predictive power. Additionally, sets of neurons with more post-synaptic neurons in common had better PCC than sets that shared only a few downstream neurons. To improve selection of pattern completion neurons *in vivo*, we computed from an ensemble's pattern of sequential activation a latency metric that was directly proportional to PCC and could potentially be estimated from modern physiological recordings. Neurons that, on average, fired earlier in an ensemble's activation pattern (shorter latency) were more likely to have better PCC. These findings can help researchers identify pattern completion neurons to stimulate *in vivo* during behavioral studies to control ensemble activation in a temporally specific manner.

Disclosures: **C. Baker:** None. **Y. Gong:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.24

Topic: I.06. Computation, Modeling, and Simulation

Support: NSF GRFP (DGE1746891)
NSF CAREER Award (1942963)
NIH BRAIN Initiative (RF1MH123233)
NSF NeuroNex Award (2014862)

Title: Generative network modeling reveals a first quantitative definition of bilateral symmetry exhibited by a whole insect brain connectome

Authors: *B. D. PEDIGO[1], M. POWELL[1], E. W. BRIDGEFORD[2], M. WINDING[4], C. E. PRIEBE[3], J. T. VOGELSTEIN[1];
[1]Biomed. Engin., [2]Biostatistics, [3]Applied Mathematics and Statistics, Johns Hopkins Univ., Baltimore, MD; [4]Zoology, Univ. of Cambridge, Cambridge, United Kingdom

Abstract: Comparing brain networks (connectomes) can help explain how neural connectivity is related to genetics, disease, development, or learning. However, the problem of making valid statistical inferences about the significance and nature of differences between networks is an open area of research, and such analysis has yet to be extensively applied to nanoscale connectomes. Here, we investigate this problem of comparing networks via a case study of the bilateral symmetry of a larval *Drosophila* brain connectome. We ask what it would mean for this brain to be bilaterally symmetric by translating this notion to generative models of the network structure of the left and right hemispheres, allowing us to test and refine our understanding of symmetry. Even when using the simplest model, which characterizes the level of hemisphere-wide connectivity, we detect a significant asymmetry. We then test whether this difference can be localized, finding asymmetry in specific groups of connections between cell types. When we adjust for the difference in hemisphere-wide connectivity level and omit a specific cell type, we no longer detect a difference between the hemispheres, providing a first quantitative definition of bilateral symmetry exhibited by an insect brain connectome. We also consider edge weight thresholds, finding that networks formed from only strong connections display no significant asymmetry under any model we considered. This work suggests aspects of connectome structure which are more likely to be preserved by the developmental program of this organism. More generally, it provides an illustrative example of how statistical inferences from networks can facilitate future comparisons of fine-scale neural structures. To enable others to use and extend these statistical tools, we make them available via our open-source Python package, graspologic, at https://github.com/microsoft/graspologic. We also document all analyses performed in this work at https://github.com/neurodata/bilateral-connectome.

Disclosures: **B.D. Pedigo:** None. **M. Powell:** None. **E.W. Bridgeford:** None. **M. Winding:** None. **C.E. Priebe:** None. **J.T. Vogelstein:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.25

Topic: I.06. Computation, Modeling, and Simulation

Support: NSF Grant 1845322

Title: Spatial embedding of edges in a synaptic generative model of C. elegans

Authors: Z. LABORDE, *E. J. IZQUIERDO;
Indiana Univ., Bloomington, IN

Abstract: Knowing how a nervous system develops is essential to understanding the neural basis of behavior. Despite a wealth of research into neuronal networks, their structural development is poorly understood. Although many factors play a role in synapse formation, including guidance molecules and the spatial location of cells and neurites, the relative role each plays in determining the final connectome remains unclear. With a relatively small and reconstructed connectome at the cellular level, *C. elegans* is an ideal subject to address this challenge. While experimental progress is being made, computational modeling of these processes can help validate existing assumptions or generate novel hypotheses, possibly leading to new theory-driven experiments. Previous research has tested how close a model could match the network properties of the *C. elegans* connectome by considering only the distance between neurons. This model generated networks within 1 standard deviation of the connectome in two key network properties: the average distance between connected nodes and the total number of bidirectional links. However, the networks differed from the connectome in other crucial metrics, including the clustering coefficient, being 2 standard deviations from the connectome. When replicating these results, we found these networks had degree distributions more akin to random networks than to that of *C. elegans*. This work extends the previous work by including the spatial location of neurites. We tested the hypothesis that spatial embedding of these processes would result in networks more like the *C. elegans* connectome than otherwise. We developed a model which formed connections at intersections of artificial neurites, passing through the densest region of the nervous system, the nerve ring. Because sensory/motor neurons often develop in a preset manner, testing was restricted to the frontal ganglia. The model's resulting networks were compared with those of the previous model, random networks, and two *C. elegans* connectomes. Using network metrics from previous studies, we found the new model was more accurate in reproducing the neural connectivity of *C. elegans*. The improvement in accuracy further supports the importance that spatial constraints of neural processes have in synapse formation. Although spatially embedding neurites improved the reconstruction of the *C. elegans* connectome, the specific connectivity remained unmatched in important ways. For example, connections between

neurons on opposite sides of the pharynx remain underrepresented. Future work will seek to consider the assumptions needed to explain the remaining differences.

Disclosures: Z. Laborde: None. **E.J. Izquierdo:** None.

Poster

578. Network Computation II

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 578.26

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NSF DBI 2015317

Title: Exploring Synaptic Strengths in the Canonical Motor Microcircuit for Control of a Rat Hindlimb

Authors: *C. JACKSON[1], W. NOURSE[2], Y. WANG[3], M. CHARDON[4], J. RUDI[7], M. C. TRESCH[5], C. HECKMAN[6], R. D. QUINN[1];
[1]Mechanical and Aerospace Engin., Case Western Reserve Univ., Lyndhurst, OH; [2]Electrical Engin., Case Western Reserve Univ., Cleveland, OH; [3]Electrical and Computer Engin., California State University, Los Angeles, Los Angeles, CA; [4]interdepartmental Neurosci., [5]Biomed. Eng, Physical Med. and Rehab, Physiol., Northwestern Univ., Chicago, IL; [6]Dept. of Physiol., Northwestern Univ., Oak Park, IL; [7]Mathematics, Virginia Tech., Blacksburg, VA

Abstract: This work explored synaptic strengths in a computational model of a controller for the hip joint of a rat consisting of Ia interneurons, Renshaw cells, and the associated motor neurons. This circuit has been referred to as the Canonical Motor Microcircuit (CMM). It is thought that the CMM acts to modulate motor neuron activity at the output stage. We first created a simplified biomechanical model of a rat hindlimb consisting of a pelvis, femurs, shins, feet, and flexor-extensor muscle pairs modeled with a linear-Hill muscle model. We then modeled the CMM using non-spiking leaky-integrator neural models connected with conductance-based synapses. Both the biomechanical and neural models were developed in Animatlab, a software environment for developing neuromechanical models. The CMM's internal parameters were then tuned such that the trajectory of the hip joint was similar to that of a rat during locomotion. We first specified parameters based on published results, combined with hand-tuning, demonstrating that this network is capable of reproducing joint kinematics. We additionally implemented an automated approach for parameter search using the Markov chain Monte Carlo (MCMC) method to solve a parameter estimation problem in a Bayesian inference framework. This approach is capable of exploring a larger parameter space than was feasible through hand-tuning and provided probability densities over the multidimensional space of parameters. We present marginals of these densities which allow us to determine if the solution space is uni- or multi-modal, as well as to determine the significance and sensitivity of each parameter. The hand-tuned results were compared to the results obtained with MCMC, showing that the MCMC approach

found parameters that produced similar results to those found while hand-tuning in terms of hip joint trajectory. We plan to use this approach to evaluate the function and significance of Ia feedback and of Renshaw cells and the interaction between these elements in a CMM and limb mechanics.

Disclosures: C. Jackson: None. **W. Nourse:** None. **Y. Wang:** None. **M. Chardon:** None. **J. Rudi:** None. **M.C. Tresch:** None. **C. Heckman:** None. **R.D. Quinn:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.01

Topic: I.07. Data Analysis and Statistics

Support: NIH RF1MH120034
NSF 2118583

Title: A universal language and format to describe behavioral tasks and data

Authors: *M. WULF[1], M. BOSC[2], R. LY[3], M. AVAYLON[3], S. STAROSTA[1], O. RUEBEL[3], A. KEPECS[1];
[1]Washington Univ. in St. Louis, St. Louis, MO; [2]The Francis Crick Insitute, London, United Kingdom; [3]Lawrence Berkeley Natl. Lab., Berkeley, CA

Abstract: Technological advances have revolutionized how we measure and manipulate brain activity, while behavioral technologies have lagged behind. Recent developments introduced methods for quantifying movements and poses but we lack general approaches to describe and communicate behavioral tasks, which are necessary to infer internal states not visible from movements alone. Different laboratories use different systems, hardware, and software to probe behavior, making it difficult to communicate task design, share data, or reproduce experiments. Furthermore, neural data archives require matching behavioral data archives for interpreting neural activity. Here we developed a universal framework for designing, implementing, communicating, and archiving behavioral tasks.
Our framework consists of two components, a description language and a data format along with associated software tools. BEADL, the BEhavioral tAsk Description Language, defines behavioral tasks as virtual finite state machines that can be described graphically as an easy-to-understand flow diagram. In each state, the sensed behavioral output of a subject is defined as events, causing transitions to other states. In addition, each state has a defined list of distinct actions, that the task controlling environment is performing (e.g., stimulus presentation). We use virtual inputs to generalize the descriptive power of this framework. BEADL's graphical representation of a behavioral task can be translated into a corresponding XML-based (eXtensible Markup Language) definition. XML provides a rigid but extensible basis and hides most of the hardware-related implementations of the behavioral control and acquisition system

from the graphical representation. A newly developed extension for the NWB-format (Neurodata Without Borders) allows for storing of behavioral data capturing both the BEADL task description together with the behavioral output of a subject. This universal task description language will help not only to further standardize data formats but also to have a consistent way of linking neural and behavioral data with the contingencies of the behavioral task.
To illustrate BEADL, we have created templates for numerous published tasks and a web-based graphical editor. We also present our workflow for automated code generation using BEADL's XML-description for one widely used control system (Bpod). In summary, we have developed and end-to-end concept for designing, communicating, executing, and archiving behavioral tasks that we hope will support our communities' efforts to understand brain function.

Disclosures: M. Wulf: None. **M. Bosc:** None. **R. Ly:** None. **M. Avaylon:** None. **S. Starosta:** None. **O. Ruebel:** None. **A. Kepecs:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.02

Title: WITHDRAWN

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.03

Topic: I.06. Computation, Modeling, and Simulation

Title: Simultaneous, unsupervised discovery of 'causally linked' neural and behavioral subspaces with external Dynamic Components Analysis

Authors: *J. YEUNG[1], J. BAK[2], K. BOUCHARD[2];
[1]Univ. of California, Berkeley, Univ. of California, Berkeley, Berkeley, CA; [2]Lawrence Berkeley Natl. Lab., Berkeley, CA

Abstract: Neural recordings are often high dimensional while the dynamics related to behavior and stimuli are thought to be low-dimensional. Similarly, many complex, ethological sensory stimuli and behaviors can be described by a large number of dynamic features. Popular dimensionality reduction techniques, such as Principal Components Analysis, Dynamics Components Analysis, or Preferential Subspace Identification, do not simultaneously identify the endogenous (neural) dynamics that are 'causal' for exogenous (behavior and stimuli) dynamics

when the relevant exogenous variables are unknown. Thus, identifying neural subspaces that 'cause' (e.g.,) an unknown subset of behavioral features is an outstanding gap.
To address this gap, we introduce external Dynamics Components Analysis (eDCA), a linear dimensionality reduction method that finds a subspace of the neural data that has maximal mutual information with a subspace of the exogenous data. Specifically, we determine the predictive information between past/future neural and exogenous dynamics. Thus, eDCA supports 'causality' in both exogenous directions - future neural 'caused' by past exogenous data such as sensory stimuli, and past neural 'causing' future exogenous data such as behavior.
First, to validate the method, we generate synthetic data using autoregressive processes with known ground truth endogenous (i.e., neural) dimensions relevant for exogenous (e.g., behavior) dimensions. We show that eDCA identifies 'causal' neural dimensions relative to an exogenous process: eDCA accurately weights the relative importance of the neural dimensions relevant for generating (a priori unknown) exogenous dynamics. We also demonstrate eDCA identifies the exogenous dimensions 'caused' by neural dynamics. Hence, eDCA simultaneously finds projections of neural and exogenous dynamics that are maximally 'causal'. Next, we apply eDCA to monkey arm reaching data with simultaneously recorded neurons in motor cortex collected by the Sabes lab. We show the neural subspace found by eDCA decodes behavioral data comparably to or better than other methods. When independent time series are included in the neural and behavioral data, we find that eDCA discards the superfluous appended dimensions.
Together, these results demonstrate that eDCA provides a framework to directly learn subspaces of neural dynamics that are causally relevant for subspaces of exogenous dynamics. eDCA can be used to identify relevant features in both neural and exogenous subspaces; thus, by compressing exogenous data, eDCA removes the burden of manual feature selection in complex, ethologically relevant stimuli and behavior.

Disclosures: **J. Yeung:** None. **J. Bak:** None. **K. Bouchard:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.04

Topic: I.07. Data Analysis and Statistics

Support: NIH Grant R01EB026936

Title: Learning Disentangled Behavior Embeddings

Authors: *C. SHI[1], S. SCHWARTZ[2], S. LEVY[2], S. ACHVAT[2], M. ABBOUD[2], A. GHANAYIM[2], J. SCHILLER[2], G. MISHNE[1];
[1]UCSD, La Jolla, CA; [2]Technion - Israel Inst. of Technol., Haifa, Israel

Abstract: To understand the relationship between behavior and neural activity, experiments in neuroscience often include an animal performing a repeated behavior such as a motor task. Recent progress in computer vision and deep learning has shown great potential in the automated analysis of behavior by leveraging large and high-quality video datasets. In this paper, we design Disentangled Behavior Embedding (DBE) to learn robust behavioral embeddings from unlabeled, multi-view, high-resolution behavioral videos across different animals and multiple sessions. We further combine DBE with a stochastic temporal model to propose Variational Disentangled Behavior Embedding (VDBE), an end-to-end approach that learns meaningful discrete behavior representations and generates interpretable behavioral videos. Our models learn consistent behavior representations by explicitly disentangling the dynamic behavioral factors (pose) from time-invariant, non-behavioral nuisance factors (context) in a deep autoencoder, and exploit the temporal structures of pose dynamics. Compared to competing approaches, DBE and VDBE enjoy superior performance on downstream tasks such as fine-grained behavioral motif generation and behavior decoding.

Disclosures: **C. Shi:** None. **S. Schwartz:** None. **S. Levy:** None. **S. Achvat:** None. **M. Abboud:** None. **A. Ghanayim:** None. **J. Schiller:** None. **G. Mishne:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.05

Topic: I.06. Computation, Modeling, and Simulation

Title: Guided pattern discovery for targeted, semi-supervised behavioral quanitification

Authors: *J. F. SCHWEIHOFF[1], A. HSU[2], M. K. SCHWARZ[4], E. A. YTTRI[3];
[1]Friedrich Wilhelms Univ. Bonn, Friedrich Wilhelms Univ. Bonn, Bonn, Germany; [3]Carnegie Mellon Univ., [2]Carnegie Mellon Univ., Pittsburgh, PA; [4]Med. Ctr., Friedrich Wilhems Univ. Bonn, Bonn, Germany

Abstract: The identification and quantification of animal behavior from video recordings have rapidly developed with the rise of easy-access, markerless pose estimation. In particular, unsupervised algorithms have enabled the unbiased discovery of movement motifs and simultaneously reduced effort costs. However, despite these considerable benefits, user refinement is often needed to align motifs with scientific nomenclature, particularly for specific social behaviors that coarse descriptions may only define.
Although some supervised methods exist that aim to reproduce human classification directly, their inflexibility and training costs are prohibitive and lack the benefits of unsupervised behavioral identification.
Our algorithm overcomes these challenges by incorporating an active-learning component into the behavioral discovery process, considerably improving accuracy and training cost. In addition, the developed pipeline allows the unsupervised discovery of latent structures (B-SOiD, Nat

Comm) within the dataset to identify subtypes within known groups or disentangle unlabeled data. These newly found groups can then be integrated into the active learning process to build a balanced, high-quality dataset for the robust classification of social behavior, combining user-defined actions with unsupervised pattern discovery in a single classifier.
We show its capabilities by investigating a sizeable human-annotated data set of social behavior in mice (CalMS21; Sun et al. 2021) and extending the range of detected behavioral expression. In our hands, the algorithm reached its highest performance (F1=87.2%), outperforming the current state-of-the-art solution (F1=86.4%; Sun et al. 2021) with only ~10% of the available training data. Thereby considerably increasing the data efficiency and reducing the effort cost. In addition, we were able to identify a known behavioral phenotype, anogenital approach, which is well described in male-female social behavior by exploring the 'investigation' class of the dataset. Notably, the dataset does not inherently discriminate between specific subtypes of investigative behavior.
Moreover, we show the real-time performance of our solution and its applicability within closed-loop experiments (DLStream, Comm Bio) and illustrate that the algorithm is agnostic to animal models in an independent single monkey dataset.
This new technique combines the benefits of supervised and unsupervised behavioral segmentation approaches to provide guided classification and discovery of conserved spatiotemporal movement patterns, all with greatly improved computational efficiency.

Disclosures: **J.F. Schweihoff:** None. **A. Hsu:** None. **M.K. Schwarz:** None. **E.A. Yttri:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.06

Topic: I.07. Data Analysis and Statistics

Title: Deep Animal Toolkit: an accessible toolbox for robust single animal tracking in complex environments

Authors: *G. KAUL, A. EBAN-ROTHSCHILD;
Univ. of Michigan, Ann Arbor, MI

Abstract: Automating the analysis of animal behavior is a major current challenge in neuroscience. In recent years, machine learning and computer vision techniques have become part of the neuroscience toolkit for the high-throughput study of animal behavior. While progress has been made in tracking single animals in simple environments, it is still a major challenge to do the same task in complex environmental conditions. This limitation has hampered the capacity to effectively study animals under ethologically-relevant conditions, such as when they are occluded inside their nests or during the dark phase/under poor lighting conditions. Moreover, as available tools often demand coding ability and specialized hardware, they are frequently inaccessible to scientists lacking the necessary computational background. Our goal

for this project was to develop a computer vision based open-source toolbox to track objects easily and automatically in complex environments. Utilizing a tracking via detection setup, we developed a robust single animal tracking method. We utilized the Faster R-CNN framework, which enables the identification of bounding boxes for objects. The bounding box predictions are then aggregated and refined using a filtering algorithm to infer the trajectory of an animal throughout the video. By using the Mask R-CNN framework (a simple model architecture extension to Faster R-CNN), our tool is able to collect additional predictions from the same videos, including keypoint/pose estimation (key points defining an animal posture) and segmentation masks (a binary mask indicating which pixels belong to the object) which can be used for further behavioral analysis. To validate our system, we curated a mini benchmark dataset that presents the relevant challenges that exist in complex environments (e.g., occlusions and low-lighting conditions). With our dataset we validated our single animal tracking system using tracking accuracy and further validated our detector's bounding box, keypoint and segmentation mask performance using COCO style evaluation (a computer vision benchmark which defines metrics for evaluating detection models). Lastly, we provide a google colab implementation of our toolbox, permitting scientists with limited computational resources or specialized hardware, to utilize it without a local setup.

Disclosures: G. Kaul: None. **A. Eban-Rothschild:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.07

Topic: I.06. Computation, Modeling, and Simulation

Support: NSF 1456560

Title: The OpenBehavior Project: A database and dissemination platform for open-source tools for behavioral neuroscience research

Authors: K. CHAVEZ LOPEZ[1], L. M. AMARANTE[2], C. B. DARDEN[1], J. A. FRIE[3], J. PALMER[1], C. L. ROOD[1], S. R. WHITE[1], A. E. BANDROWSKI[4], J. Y. KHOKHAR[3], A. V. KRAVITZ[5], *M. LAUBACH[1];
[1]Neurosci., American Univ., Washington, DC; [2]Johns Hopkins Univ., American Univ., Baltimore, MD; [3]Univ. of Guelph, Guelph, ON, Canada; [4]UCSD, La Jolla, CA; [5]Washington Univ. In St Louis, Saint Louis, MO

Abstract: The OpenBehavior Project promotes the use of open-source tools for behavioral neuroscience research. Since 2016, the project has disseminated information on more than 200 research tools on a weekly basis through blog posts to our website and through social media. Over the past 18 months, we have (i) created a database of all tools featured on openbehavior.com and issued Research Resource IDentifiers (RRIDs) that facilitate the citation

and tracking of the tools in research publications; (ii) created a repository of raw video recordings of animals performing behavioral tasks that are commonly used in neuroscience research, organized a series of community conversations on video analysis tools, and written a commentary on setting video methods in a lab and best practices for the use of video methods; and (iii) developed in-person and virtual training workshops on Arduino-based microcontrollers, which are heavily used in neuroscience research. The goal of this poster is to share these efforts with the community and also plan for three new initiatives over the coming year. First, we are launching a new repository of computer code from open-source control platforms (e.g. Arduino, Bonsai, PyControl) and electronic circuit designs used for commonly used behavioral tasks. In parallel, we will organize a community effort to better document the setup, use, and integration of behavioral control systems with other popular open-source tools for data acquisition (e.g. Open Ephys, Open Miniscope, DeepLabCut). Second, we will create a repository of open-source designs for 3D printed objects that are used in neuroscience research. Having access to design files will allow researchers to quickly implement new devices into existing experimental setups and also replace failed parts for devices as diverse as stereotaxic instruments, video cameras, and electronic and optical interfaces (e.g. commutators). These repositories will make research more reproducible and reduce barriers for the use of open-source methods and tools. Finally, we will organize training workshops on data analysis methods and modeling packages that are based on open-source computer languages (Python, R) and used for the study of behavior (e.g. Reinforcement Learning, Drift Diffusion Models). We hope that these efforts continue to stimulate development and innovation, as well as more widespread use of the powerful and cutting-edge methods that have emerged from the open-source neuroscience community.

Disclosures: **K. Chavez Lopez:** None. **L.M. Amarante:** None. **C.B. Darden:** None. **J.A. Frie:** None. **J. Palmer:** None. **C.L. Rood:** None. **S.R. White:** None. **A.E. Bandrowski:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); SciCrunch Inc.. **J.Y. Khokhar:** None. **A.V. Kravitz:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NSF 1456560. **M. Laubach:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NSF 1456560.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.08

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH DA036657

Title: Behavbox: a flexible, open-source system for behavioral tasks in neuroscience research

Authors: *T. QIU[1], Y. TIAN[3], L. L. SJULSON[2];
[2]Neuroscience, Psychiatry, [1]Albert Einstein Col. of Med., Bronx, NY; [3]Biomed. Engin., Univ. of Michigan, Ann Arbor, MI

Abstract: In systems neuroscience, both behavioral and physiological analyses are essential for research. An experimental setup that takes both of these into account requires hardware and software that can integrate various devices for different purposes in an experiment. We therefore developed a system called BehavBox, which utilizes a set of single-board Raspberry Pi computers running Python, a flexible and easy-to-learn programming language. On the hardware level, the system of Raspberry Pis is inexpensive, and the communication protocol between the Pis and other devices is easy to control, either using I2C, TTL pulses, or ethernet/WiFi. It also allows for time sensitive synchronization between the devices, hence allowing for high accuracy in tracking behavioral data and physiological recordings both in real time and after the experiment. The system uses a custom-written Python-based state machine architecture that allows beginner-level programmers to implement a wide range of behavioral paradigms. The objectives of developing the BehavBox system were to assist systems neuroscience and behavioral psychology researchers to be able to set up a wide range of behavioral and experimental protocols that allow efficient coordination within the system while constraining the setup to be easy to use in a research pipeline.

Disclosures: T. Qiu: None. **Y. Tian:** None. **L.L. Sjulson:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.09

Topic: I.06. Computation, Modeling, and Simulation

Support: Boehringer Ingelheim Ulm University Bio Center (BIU2)
Institute of Applied Physiology, Ulm University.

Title: Pymaze - an open-source integrated software and hardware for simplified deep-learning-based behavioural tracking and control of automated mazes

Authors: *S. T. KAPANAIAH[1], S. LAMMERICH[1], H. ROSENBROCK[2], B. HENGERER[2], D. KÄTZEL[1];
[1]Inst. für Angewandte Physiologie, Univ. Ulm, Ulm, Germany; [2]Boehringer Ingelheim Pharma GmbH & Co., Biberach an der Riss, Germany

Abstract: To assess cognitive and affective functioning in rodents, various arenas and mazes are widely used, since the advent of behavioural neuroscience. However, these are often run manually and use manual scoring or proprietary software for tracking. This entails several drawbacks: manual operation interferes with the animal's behaviour (e.g. to open doors), is labor-intense, and is mostly limited to one animal at a time. Standard tracking software, in turn,

usually comes with tracking errors, the limited possibility of pose-estimation (e.g., head-direction), and only costly integration with external hardware. To overcome these limitations while retaining the simple "plug-and-play" applicability that commercial tracking software offers, we developed and evaluated *pyMaze*. *pyMaze* includes a stand-alone Python-based graphical user interface (GUI) that allows setup experiments easily in mazes of arbitrary shape, defining zones, task stages, and outputs. This system is built on *DeepLabCut-live* inference and thereby attains practically error-free tracking and pose-estimation. pyMaze runs on personal computers with dedicated graphics cards; but to reduce costs, we used the NVIDIA Jetson development board, which can process images online up to 20 FPS from two setups simultaneously, or 30-35 FPS from a single setup (depending on camera hardware and image size). We trained a TensorFlow model (mobilenet_v2_1.0) using DeepLabCut™ and used the exported model for online tracking, which works in various mazes. Tracking results showed a higher correlation with manual scoring (r ~ 0.9-1.0) than commercial standard tracking software results. Beyond its function as high-performance tracking software, pyMaze is built for versatile bidirectional interaction with external devices (e.g., for time-stamping of physiological recordings) and microcontrollers. We used an STM32 high-performance Arm® Cortex®-M7 core microcontroller-based hardware, utilizing functions from the pyControl framework, to operate fully automated multi-arm mazes. We further present an optimized design for stepper-motor-driven rising doors with a very low audible noise that is controlled by pyMaze based on task sequences and tracking information about the animal's location. Their smoothly rising and automated operation minimizes interference with animal behaviour. Finally, we validated the integration of software and hardware of the pyMaze system, showing a seamless operation in complex multi-stage sequences and a fully automated maze. All designs and code will be open-source on GitHub (https://github.com/KaetzelLab).

Disclosures: **S.T. Kapanaiah:** None. **S. Lammerich:** None. **H. Rosenbrock:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co. **B. Hengerer:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co.. **D. Kätzel:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.10

Topic: I.07. Data Analysis and Statistics

Support: CIHR37790

Title: Pi Mouse Studio: Synchronize Capture and Identification of Mouse Posture Patterns in 3-Dimension

Authors: *T. L. FONG[1], H. HU[2], T. H. MURPHY[2];
[1]Univ. of British Columbia, Univ. of British Colombia, Vancouver, BC, Canada; [2]Psychiatry, Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Characterizing posture and quantifying movement patterns in animals are crucial for behavior phenotyping. Traditionally, this has been manually done by human observers, which is labor-intensive and often fails to detect differences in the rapid, fine-scale movements, ultimately resulting in the inability to characterize many behaviors of interest. Recent advances in computer vision, namely DeepLabCut, have popularized 2-D posture tracking due to its high accessibility and ability re-train underlying weights for posture detection in a wide variety of experimental settings. However, precise animal behavior characterization requires 3D measurements of whole-body movements using at least three distinct view angles to resolve posture ambiguities in cases such as visual occlusion. Currently, most synchronous multi-view video capture setups employ high-speed cameras with mirrors or expensive custom hardware, significantly reducing the field of view and requiring a high degree of expertise to set up, respectively. Therefore, we developed the Pi-Mouse Studio (PMS), an easy-to-setup and open-source assay for capturing mice movement in 3D. The setup joins four Raspberry Pi computers through ethernet connections and GPIO pins to achieve near synchronous recordings using light pulses. Three cameras were positioned horizontally to the animal at 120 degrees viewing angles to each other, and a fourth camera was positioned underneath the animal. Video Recordings were done in a cylinder with a high animal-to-background contrast environment under IR-light. During the offline analysis, postures were first detected for each camera in DeepLabCut and then triangulated to compose a 3D posture model using Anipose. Through UMAP dimension reduction and clustering, we were able to identify and quantify different behaviors, including rearing, turning, and reaching. We further plan to test mice models of Huntington's disease and stroke in PMS, to allow for precise classification of movement symptom progression/recovery.

Disclosures: **T.L. Fong:** None. **H. Hu:** None. **T.H. Murphy:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.11

Topic: I.06. Computation, Modeling, and Simulation

Support: Mong Cornell Neurotech Fellowship
NIH Grant R21AG066001

Title: A fast and immersive headset-based VR system for mouse neuroscience and behavior

Authors: M. ISAACSON[1], ***H. CHANG**[2], R. ZIRKEL[1], I. T. ELLWOOD[2], C. B. SCHAFFER[1];
[1]Biomed. Engin., [2]Neurobio. & Behavior, Cornell Univ., Ithaca, NY

Abstract: Virtual reality (VR) has been a useful tool for animal model neuroscience for decades, enabling the study of complex behaviors in head- or body-fixed animals and investigations of the neural circuitry underlying visual and spatial cognition, learning, and decision-making. Wider adoption of VR has been hindered by the high cost and/or technical complexity of panoramic-display systems, which due to their size can be cumbersome to integrate into existing experimental setups. Following the resource-efficient headset-based design of household VR systems for humans, we have developed a fully functional mouse-sized VR headset: a smaller, faster, simpler, and more immersive VR platform for mouse neuroscience. Using SPI-based circular LED displays and Fresnel lenses positioned for infinity focus, we created wide field-of-view eyepieces (120-140^{o} FOV per eye) which can fully enclose the mouse eye, blocking confounding visual stimuli (such as from microscope objectives, treadmills, or lick ports) as well as preventing light pollution from the displays from interfering with other experimental equipment (such as sensitive microscope PMTs). Using just a single eyepiece driven by a fast microcontroller (Teensy 4.0) and open-source graphics library, we created a compact (~1.5 in^{3}) display device for simple monocular visual stimulation - a convenient tool for vision neuroscience. Presenting drifting gratings on the monocular eyepiece to an anesthetized mouse during 2-photon calcium imaging of contralateral V1 layer 2/3, we successfully stimulated and identified orientation- and direction-selective cells. Using a two-display binocular headset driven by a Raspberry Pi 4 and custom SPI-display driver, we could create 3D environments using the cross-platform Godot video game engine and project the environments on the displays using a split-screen viewport and custom shaders. With this 3D platform, we can present interactive virtual environments such as linear-tracks and open fields to head-fixed mice walking on a spherical treadmill, simulating spatial navigation at high speed and low latency (<60 ms input-to-output delay at 60 fps). This headset-based VR system represents a significant advancement in VR technology for mouse neuroscience; it makes VR methodologies accessible to more labs due to its small size and low cost (~3.5 x 2.5 x 2.5 in, <$200 in parts), allows precise control over the visual stimuli presented to each eye independently, and improves our ability to study the neural circuitry underlying virtual navigation in head-fixed settings.

Disclosures: **M. Isaacson:** None. **H. Chang:** None. **R. Zirkel:** None. **I.T. Ellwood:** None. **C.B. Schaffer:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.12

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH Grant AG057622
NIH Grant R21AG066001

Title: Modular Mouse Maze: a powerful, easy solution for mouse behavior

Authors: M. ISAACSON[1], ***Y. PARK**[2], O. EMEROLE[2], L. E. BERKOWITZ[1], N. NISHIMURA[1], C. B. SCHAFFER[1];
[1]Meining Sch. of Biomed. Engin., [2]Neurobio. and Behavior, Cornell Univ., Ithaca, NY

Abstract: Mouse behavioral assays are used ubiquitously in neuroscience and translational research. The wide variety of behavioral apparatuses (chambers, mazes, open fields) and experimental paradigms (manual or automated; passive or operant) demonstrate how useful behavioral tools can be for studying navigation, anxiety, learning and memory, and many other aspects of cognition in healthy mice and disease models. However, despite the similar size and shape of many behavior apparatuses (e.g. Y-maze, T-maze) and that many are composed of nearly the same parts (e.g. similar height plastic walls; nose poke sensors; reward ports), many behavior setups are not interchangeable, requiring their own individual investment of lab space and set up time. To reduce the various costs and inefficiencies associated with specialized behavior setups, we created the Modular Mouse Maze: a simple, modular, and entirely open-source system for accommodating a wide variety of mouse behavioral experiments using a common set of plug-and-play parts. With 3D-printable wall and floor panels that can be slotted together in various configurations - designed for consistent quality even when using low-cost PLA filament 3D printers and with care taken to minimize any unintended visual landmarks of the panels - new maze configurations and chambers can be quickly assembled and rearranged. Additionally, taking advantage of the expanding ecosystem plug and play I2C devices (such as Adafruit's STEMMA QT and SparkFun's QWIIC systems), we created special wall panels with mounting slots that can hold LED displays, proximity sensors, distance sensors, and more, enabling easier data collection and experiment automation with zero soldering or custom PCBs required. These low-power, low- noise electronics can even be used with free-walking electrophysiology without interfering with the sensitive neural recordings, accommodating automated navigational assays with hippocampal recordings. Using a modular linear track outfitted with LED displays for visual cues, nose poke detectors, and liquid reward spouts, and controlling the I2C devices with the user-friendly CircuitPython programming language, we successfully trained mice to navigate to rewarded locations with an automated training protocol. Additionally, we could synchronize the automated data collection with an overhead-mounted machine vision camera for mouse body part tracking with DeepLabCut, to collect even richer behavioral datasets. A list of components necessary to build modular mazes (3D print design files, I2C part lists, and CircuitPython code) are organized and freely available online (ModularMouseMaze.org).

Disclosures: M. Isaacson: None. **Y. Park:** None. **O. Emerole:** None. **L.E. Berkowitz:** None. **N. Nishimura:** None. **C.B. Schaffer:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.13

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH Grant: R01AA029409
NIH Grant: P60-AA007611

Title: A Machine Learning Audio Detection Algorithm to Identify Bouts of Drinking in Rodent Voluntary Consumption Studies

Authors: *S. WEIR[1], C. ARDINGER[1], C. LAPISH[1,2];
[1]Indiana Univ. Purdue Univ. Indianapolis, Indiana University-Purdue Univ. Indianapolis, Indianapolis, IN; [2]Stark Neurosci. Inst., Indiana Univ. Sch. of Med., Indianapolis, IN

Abstract: Voluntary fluid consumption studies in rodents are a critical component for many fields of neuroscience research. Our lab combined awake-behaving electrophysiology recordings with standard home-cage ethanol drinking procedures to further our understanding of alcohol's impact on brain circuitry. We were unable to implement standard methods of identifying the timing of bouts of drinking, such as using either a circuit based lick'o'meter or volumetric drinking monitors, due to the electrical interference between these technologies and the electrophysiology equipment. To resolve this problem, we present an audio detection machine learning algorithm which is able to take sounds in the form of voltage recorded from a piezo microphone attached to a sipper and categorize them as Drinking or Non-Drinking. This algorithm was designed using the Tensorflow library and consisted of a 2D convolutional neural network which was trained on a total of 9.7 hours of data across 14 animals and resulted in a training accuracy of 99.7% and an out of sample testing accuracy of 93.3%.

Disclosures: S. Weir: None. **C. Ardinger:** None. **C. Lapish:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.14

Topic: I.06. Computation, Modeling, and Simulation

Support: Jazz Pharmaceuticals
NIH Grant NS107148

Title: A home-cage approach for quantification of harmaline-induced tremor in rats using piezoelectric sensors

Authors: *D. M. HUFFMAN[1], E. F. BRIGHAM[2], R. BERNAT[1], S. SUNDERAM[3], B. F. O'HARA[1];
[1]Signal Solutions, LLC, Lexington, KY; [2]Jazz Pharmaceuticals, Palo Alto, CA; [3]F. Joseph Halcomb, III M.D. Dept. of Biomed. Engin., Univ. of Kentucky, Lexington, KY

Abstract: Essential tremor (ET) is a common movement disorder characterized by uncontrollable shaking (tremor) of the hands, limbs, or other body parts. Administration of

harmaline in rodents induces acute tremor and is accepted as a valid model of clinical relevance to ET. Quantifying the induced tremor and its response to pharmacological intervention is a key step toward translation. While piezoelectric motion sensors are used for noninvasive tremor assessment, available systems rely on methods that limit the pace and scale of experimentation. Here, we assessed the feasibility of using a commercial piezo-based home-cage rodent monitoring system to detect tremor, which could enable continuous and high-throughput experimentation. Experiments were performed to determine whether: (1) this sensor configuration generates a signal that is representative of tremor; (2) a quantitative metric can be derived from this signal that reflects tremor onset and progression; and (3) this metric is sensitive to changes induced by a common tremor-suppressing agent used clinically to treat ET (propranolol) - serving as a positive control. Six adult Sprague-Dawley rats (2M, 4F) were continuously monitored in cages resting on piezoelectric sensors for several days. During the recording, animals received either subcutaneous injections of saline, harmaline (10 mg/kg), or saline/propranolol (20 mg/kg) followed by harmaline (10 mg/kg). Following harmaline administration, piezoelectric sensors produced signals representative of tremor behavior (peak frequency of 9-12 Hz), which appeared alongside observed tremor events. To quantify the tremor signal, data segments corresponding to 30 minute pre-, 20 minute inter- (when applicable), and 300 minute post-injection periods were processed to estimate the contribution of tremor band frequencies (9-12 Hz) to signal power (tremor band contribution; TBC) in 10-minute windows. Following harmaline injection, TBC quickly rose above baseline levels, and was sustained for approximately 3 hours before reverting to baseline levels as the effect of harmaline subsided. In contrast, TBC remained at baseline levels throughout the experiment in saline-treated rats. When pre-treated with propranolol, the peak TBC was delayed and reduced compared to saline-treated controls, a trend consistent with the literature. Overall, these results demonstrate the potential of this technology for non-invasive assessment of tremor.

Disclosures: **D.M. Huffman:** A. Employment/Salary (full or part-time):; Signal Solutions, LLC. **E.F. Brigham:** A. Employment/Salary (full or part-time):; Jazz Pharmaceuticals. **R. Bernat:** A. Employment/Salary (full or part-time):; Signal Solutions, LLC. **S. Sunderam:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; PI on a collaborative federal grant between Signal Solutions and the University of Kentucky. **B.F. O'Hara:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Signal Solutions, LLC.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.15

Topic: I.06. Computation, Modeling, and Simulation

Support: The Canadian Open Neuroscience Platform (CONP)

Title: Towards automatic 3D pose estimation using a Kinematic Evidence-based synthetic mouse body model

Authors: *H. HU, T. FONG, D. XIAO, L. A. BOLAÑOS, H. RHODIN, T. H. MURPHY; Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Robust and accurate measurement, representation, and analysis of 3D animal behaviors are of great interest in neuroscience. 3D behavioral data analysis has less bias and can be better studied across subjects, illumination, and background. Currently, the definition of the rodent 3D pose is skeleton-based and consists of a series of keypoints. Several attempts have been made to reconstruct skeleton-based 3D poses with specialized hardware (motion capture, depth camera, synchronized multi-view cameras) or data-driven methods (3D lifting using deep learning). However, the reconstruction of the skeleton-based 3D pose of rodents is challenging, due to the lack of 3D ground truth data, fewer consistent visual landmarks in video recording, and poor body representation of keypoints (obscured by fur). Recent advances in computer vision bring the volumetric representation from human to animal and have reached some success in 3D animal pose estimation by leveraging both the visual cue of keypoints and animal shape using silhouettes that are both present in video recording (models include dogs, horses, and birds). However, those models failed during applied to rodents' data due to the differences in body plan between species. Therefore, we developed the **P**ose **O**ptimization with a **K**inematic **E**vidence-based **mouse** model (Poke-mouse), which allows estimating 3D mice' pose and shape from images and existing 3D scans. The model is built based on computed tomography scans of Female C57BL/6 mice under anesthesia. It represents mouse shape using 12077 vertices from 97 parameters, and it was formed with the same format as the Skinned Multi-Animal Linear Model (SMAL). During 3D pose estimation, the keypoints are detected with DeepLabCut, a popular 2D posture estimation method with high accuracy. The shape information is extracted from backgrounds using U2Net. The parameters of Poke-mouse are estimated by minimizing the error in both keypoints and shapes using the EM algorithm. We further plan to validate the performance of Poke-mouse by comparing it with other 3D pose estimation algorithms like Anipose.

Disclosures: H. Hu: None. **T. Fong:** None. **D. Xiao:** None. **L.A. Bolaños:** None. **H. Rhodin:** None. **T.H. Murphy:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.16

Topic: I.06. Computation, Modeling, and Simulation

Support: The Michael Smith Foundation for Health Research trainee award to Dongsheng Xiao

Title: Constructing deformable dense body map for automated quantification of mouse social contacts using generative adversarial networks.

Authors: *D. XIAO, M. CHEN, T. FONG, E. YAN, T. H. MURPHY;
Dept. of Psychiatry, Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Subjective feelings, such as social contact, are associated with a unique set of bodily sensations. While body contact is a fundamental aspect of social behaviors and the body plays a crucial role in cognitive and emotional processes, there is no existing tool to quantify the detailed social contact on the body. To this end, we developed an approach to constructing a dense body model of mice and generating bodily maps to represent embodied space of social feelings. We leverage the power of pre-trained deep generative models that employ the latent space of a GAN (trained on the unaligned data) to automatically generate paired training data for a Spatial Transformer. First, we extracted frames from mouse open-field videos (social interaction of two mice recorded by a top-down camera) and cropped single mouse images (3.7 million mouse images) using YOLO bounding box. We then pre-trained a latent variable generative model, StyleGAN, on the unaligned mouse images. The pre-trained GAN model possesses some innate style-pose disentanglement that can be used to construct the per-image target. We used GANgealing algorithm that trains the Spatial Transformer to warp input images (unaligned mouse) to a common, jointly learned target mode (aligned mouse with dense correspondences). The body model of mice is then constructed based on the learned target mode. As the body shape of mice is changed during different social interactions and behavior states, we extend the method with a clustering algorithm to learn more than one target mode and more than one Spatial Transformer. We identify dense correspondences between real input images and the constructed mouse body model with the Spatial Transformer by propagating from congealed coordinate space. Finally, we propagate the dense mouse body model to all the frames of an input video and generate bodily maps of social contact (based on the pixel overlap) of two mice. Our approach can classify social behaviors based on the body shape in different interaction states and generate a detailed body map for quantifying the location, area, posture, and frequency of social contacts across time.

Disclosures: **D. Xiao:** None. **M. Chen:** None. **T. Fong:** None. **E. Yan:** None. **T.H. Murphy:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.17

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH Grant ES031050

Title: Social behavioral profiling by unsupervised deep learning reveals a stimulative effect of dopamine D3 agonists on zebrafish sociality

Authors: *Y. GENG[1], R. T. PETERSON[2];
[1]Univ. of Utah, SALT LAKE CITY, UT; [2]Univ. of Utah, Salt Lake City, UT

Abstract: It has been a major challenge to systematically evaluate and compare how pharmacological perturbations influence social behavioral outcomes. Although some pharmacological agents are known to alter social behavior, precise description and quantification of such effects have proven difficult. The complexity of brain functions regulating sociality makes it challenging to predict drug effects on social behavior without testing in live animals, and most existing behavioral assays are low-throughput and provide only unidimensional readouts of social function. To achieve richer characterization of drug effects on sociality, we developed a scalable social behavioral assay for zebrafish named ZeChat based on unsupervised deep learning. High-dimensional and dynamic social behavioral phenotypes are automatically classified using this method. By screening a neuroactive compound library, we found that different classes of chemicals evoke distinct patterns of social behavioral fingerprints. By examining these patterns, we discovered that dopamine D3 agonists possess a social stimulative effect on zebrafish. The D3 agonists pramipexole, piribedil, and 7-hydroxy-DPAT-HBr rescued social deficits in a valproic acid-induced zebrafish autism model. The ZeChat platform provides a promising approach for dissecting the pharmacology of social behavior and discovering novel social-modulatory compounds.

Disclosures: Y. Geng: None. **R.T. Peterson:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.18

Topic: I.06. Computation, Modeling, and Simulation

Support: NHMRC Investigator Award 1196855 to GJG

Title: Behavioral adaptations to changing energy constraints over development in larval zebrafish

Authors: *T. DARVENIZA[1], Z. PUJIC[2], M. H. MCCULLOUGH[2], S. I. ZHU[1], B. SUN[2], R. AGARWAL[1], G. J. GOODHILL[1,2];
[1]Washington Univ. in St. Louis, St Louis, MO; [2]Univ. of Queensland, Brisbane, Australia

Abstract: A significant constraint on animal behavior is energy consumption, however the energy required for movements changes differentially as the animal grows. This is particularly true for larval fish which progress from viscous to inertial fluid regimes during early life. Here we combine high-resolution video tracking of larval zebrafish with computational fluid dynamics

simulations to investigate how the energy required for different movements changes over development. We show that basic bout types are preserved over development, but at each age there is a monotonic, approximately power-law relationship between energy consumption and the frequency of different bout types during hunting behaviors. More energetically expensive movements are always used less often, even though the ordering of bouts according to energy changes over development. When fish were raised and tested in a viscous environment they also retained this monotonic relationship. Together this work suggests a strategy by which developing animals can preserve movement primitives yet still adapt to changing energy constraints as they grow.

Disclosures: **T. Darveniza:** None. **Z. Pujic:** None. **M.H. McCullough:** None. **S.I. Zhu:** None. **B. Sun:** None. **R. Agarwal:** None. **G.J. Goodhill:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.19

Topic: I.06. Computation, Modeling, and Simulation

Support: NSF DBI2015317
NSF GRFP DGE1745016

Title: Slugbot: a soft robotic gripper with a bio-inspired neural controller

Authors: ***M. J. BENNINGTON**[1], K. DAI[1], R. SUKHNANDAN[2], K. WHIRLEY[1], R. BAO[1], L. LI[3], J. P. GILL[5], H. J. CHIEL[5,6,7], V. A. WEBSTER-WOOD[1,2,4];
[1]Mechanical Engin., [2]Biomed. Engin., [3]Robotics Inst., [4]McGowan Inst. for Regenerative Med., Carnegie Mellon Univ., Pittsburgh, PA; [5]Biol., [6]Neurosciences, [7]Biomed. Engin., Case Western Reserve Univ., Cleveland, OH

Abstract: Bio-inspired robots have great potential as platforms for testing biomechanical and neuromechanical hypotheses [1,2]. The common housefly [3], cockroaches [4], and the praying mantis [5], among others [2], have all been the inspiration for robotic platforms to explore both biological control of animals and bio-inspired control of robotic platforms. To develop such platforms, it is critical to capture an adequate level of morphological detail *in roboto* and to have requisite knowledge of the biological neural networks that process information and control the periphery.
The neural circuitry of the marine mollusk *Aplysia californica's* feeding apparatus (buccal mass) has been well studied with the identification of many neurons and muscles that govern multifunctional behavior. These behaviors include biting (attempt to grasp food), swallowing (ingestion of food) and rejection (egestion of inedible food). These properties make *Aplysia* a model organism for applying bio-inspired control strategies to soft robotic platforms.
A Slug-Like Uniaxial Grasping roBOT, SLUGBOT, was developed as a step towards studying

Aplysia neuromechanics *in roboto.* SLUGBOT was designed to mimic key features of the *Aplysia* buccal mass musculature, and a previously developed *Aplysia*-inspired neural control model was modified to control the robot [6]. Additions to the controller include known motor neuron connections, regional innervation, and phenomenological connections based on previously reported behavioral and neural recordings. Regional activation was incorporated for the I3 muscle, which is primarily responsible for retraction. Independent control was incorporated for the I1 muscle and the radular opener/closer muscles. An additional motor neuron, B10, was added to include the hypothesized multifunctional role of I3 in protraction [7], which was required for cyclic behaviors in the robot. Despite morphological differences between the robot and animal model, the robot demonstrated cyclical translational and rotational kinematics that were qualitatively similar to biting and swallowing behaviors observed *in vivo* and in previously developed *in silico* biomechanical models. Further refinement and tuning of the mechanical system is needed for this robot to serve as a platform for testing novel neuromechanical hypotheses, but the presented work is an important step towards the use of robotic platforms in *Aplysia* research.
*Equal Contributions: Bennington, Dai, Sukhnandan
References: [1] Gravish, et al., 2018. [2] Nishikawa, et al. 2007. [3] Goldsmith, et al., 2019. [4] Choi, et al. 2005. [5] Szczenski, et al. 2017. [6] Webster-Wood, et al. 2020. [7] Sutton, et al. 2004.

Disclosures: **M.J. Bennington:** None. **K. Dai:** None. **R. Sukhnandan:** None. **K. Whirley:** None. **R. Bao:** None. **L. Li:** None. **J.P. Gill:** None. **H.J. Chiel:** None. **V.A. Webster-Wood:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.20

Topic: I.06. Computation, Modeling, and Simulation

Support: UKRI Grant Number (MR/T046619/1), part of the NSF/CIHR/DFG/FRQ/UKRI-MRC Next Generation Networks for Neuroscience Program
Royal Society (UF130507)
US Army Research Office (W911NF-15-038)

Title: A quasi-static approach to modeling soft-tissue structures

Authors: *B. KUNDU, S. ROGERS, G. SUTTON;
Sch. of Life Sci., Univ. of Lincoln, Lincoln, United Kingdom

Abstract: Behavior is produced through a combination of neural and muscle activity. Understanding and modeling how the nervous system controls geometrically complex muscular structures is extremely challenging. In addition, this complexity often leads to another challenge

for simulations: long computational times. We have studied the mechanical control of the geometrically complex muscular structures controlling feeding in the marine mollusk, *Aplysia californica*. The feeding apparatus (buccal mass) moves under the influence of several muscle forces activated by neural commands from the buccal ganglion. We have modeled this system with four components: 1) the odontophore, a spherical grasping structure; 2) the I2 muscle, a sheet of muscle posterior to the odontophore; 3) the I3 muscle, a large toroidal muscle anterior to the odontophore; and 4) a hinge which attaches the ventral odontophore to I3, using Newtonian mechanics similar to reference [1]. We reproduced and solved the primary model [1] (Second-order model, based on Newton's principle of motion) in *Mathematica*. The second-order model took 14 s to compute a simulation of 1 s of feeding behavior. However, this complex system moves slowly, and therefore the inertia of this system is negligible with respect to other forcing agents. This allows us to propose an alternate solution based on a quasi-static relationship between motion and neural activation, allowing us to replace the Newtonian formulation with a search for the equilibrium position of the buccal mass (i.e., equating the sum of all muscle forces to zero). This reformulation required just 0.12 s to simulate 1 s of feeding behavior, over 100 times faster than the Newtonian formulation. Both the quasi-static model and the second-order model produce results that are quantitatively near identical. Therefore, the quasi-static model reduces computational time without sacrificing accuracy. References: [1] Sutton, G.P., Mangan, E.V., Neustadter, D.M., Beer, R.D., Crago, P.E., Chiel, H.J.: Neural control exploits changing mechanical advantage and context dependence to generate different feeding responses in Aplysia. Biological cybernetics 91(5), 333-345 (2004)

Disclosures: B. Kundu: None. **S. Rogers:** None. **G. Sutton:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.21

Topic: I.06. Computation, Modeling, and Simulation

Support: NSF DBI2015317
NSF IIS1704436

Title: A Synthetic Nervous System Model that Reproduces Neuron Dynamics and Multifunctional Control of Aplysia Feeding

Authors: *Y. LI[1], V. A. WEBSTER-WOOD[3], J. P. GILL[2], G. P. SUTTON[4], H. J. CHIEL[2], R. D. QUINN[1];
[1]Dept. of Mechanical and Aerospace Engin., [2]Dept. of Biol., Case Western Reserve Univ., Cleveland, OH; [3]Mechanical Engin., Carnegie Mellon Univ., Pittsburgh, PA; [4]Dept. of Life Sci., Univ. of Lincoln, Lincoln, United Kingdom

Abstract: Building an accurate computational model can inform the study of feeding control in *Aplysia*. Feeding, a basic motor control task, has been extensively studied in animals. The marine mollusk *Aplysia californica* is a good candidate for studying this topic. Understanding its feeding may further provide insights on how animals can achieve multifunctional, adaptive, and robust behaviors. However, it is still too difficult to experimentally determine all relevant biophysical properties and connectivity in the nervous system of *Aplysia*. A computational model with predictive capabilities provides a controllable platform to test hypotheses. As a complement to experiments, it is a powerful tool to clarify the relationship between known neural circuitry and measurable behaviors. We developed a model of *Aplysia* feeding circuitry in the framework of Synthetic Nervous Systems (SNS). An SNS is a biologically plausible neural model in which neurons are treated as a single compartment with conductance-based inputs and firing-rated outputs. In the model, motor neurons and buccal ganglion interneurons are organized into 9 subnetworks according to their distinctive functions. CBI-2, CBI-3, and CBI-4, three cerebral-buccal interneurons coordinate the transition of feeding behaviors by selectively combining these subnetworks. The model emphasizes feedback integration by considering potential afferent pathways. These pathways provide proprioceptive and exteroceptive feedback to the model, allowing it to coordinate and control the feeding behaviors in response to different load conditions. Model parameters are tuned based on existing literature and experimental results. To test whether this SNS can accurately reflect the neural dynamics in *Aplysia*, we drove the model of representative neurons with specific inputs and quantitatively compared their responses with other models as well as animal data. Furthermore, we find that the model qualitatively reproduces biting, swallowing, and rejection behaviors when it is connected to a simplified biomechanical model of *Aplysia* [1]. The kinematic and dynamic responses of the model also share similar features with experimental data. The results suggest that this circuitry model has predictive capabilities and could be used for generating or testing hypotheses about *Aplysia* feeding control. It is also possible to generalize the model for robotic control considering its relatively low computational complexity. [1] Webster-Wood, et al. Biol Cybern 114, 557-588 (2020).

Disclosures: Y. Li: None. **V.A. Webster-Wood:** None. **J.P. Gill:** None. **G.P. Sutton:** None. **H.J. Chiel:** None. **R.D. Quinn:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.22

Topic: I.06. Computation, Modeling, and Simulation

Title: Expanding Caenorhabditis elegans electroshock seizure assay by introducing machine learning for behavioral analysis

Authors: *P. SCARPINATO[1], J. XIE[1], D. VASQUEZ[1], W. HAHN[2], K. DAWSON-SCULLY[1,3];

[1]Dept. of Biol. Sci., [2]Dept. of Mathematical Sci., Florida Atlantic Univ., Boynton Beach, FL; [3]Dept. of Psychology and Neuroscience, Col. of Psychology, Nova Southeastern Univ., Davie, FL

Abstract: Technology and tools used in biology are often the backbone to the experiments themselves. Our lab has developed a *Caenorhabditis elegans* electroshock assay, which provides high throughput screening of drugs for effectiveness on seizure-like behavior duration. These experiments are conducted and recorded for later analysis by hand. The analysis of the seizure-like behavior from these experiments are subjective to the researcher and time consuming. Currently, there are numerous programs that track the movement of *C. elegans* and provide simple analysis, but often have a steep cost and are not suitable for the behavioral analysis we require. This project is aimed to develop and evaluate an expandable program that is capable of worm tracking with thorough experimental analysis of our *C. elegans* electroshock assay. Additionally, the project will develop the supporting hardware to improve the quality and consistency of the collected experimental videos. The approach to this development will be based on supervised machine learning programming methods, which essentially teaches the computer what to do through examples that have been provided. Using this hardware, we have conducted experiments, which shows the antiepileptic effects of Isoguvacine ($p < 0.05$) compared to that of sodium valproate, an FDA approved anticonvulsant. This data will be used as a training set to develop a general machine learning behavioral analysis model for this assay. This development will improve the quality of data collection and accelerate the analyses of the experiments.

Disclosures: **P. Scarpinato:** A. Employment/Salary (full or part-time):; Florida Atlantic University. **J. Xie:** None. **D. Vasquez:** None. **W. Hahn:** None. **K. Dawson-Scully:** None.

Poster

579. Experimental Tools: Behavior Experiments

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 579.23

Topic: I.07. Data Analysis and Statistics

Support: NIH Grant R25 NS 80686

Title: Exploring the Use of Convolutional Neural Networks to Characterize Larval Behavior

Authors: *A. NWANKPA, Jr.[1], P. MCNULTY[2], R. WU[2], M. GERSHOW[2]; [1]BP Endure, [2]Physics, New York Univ., New York, NY

Abstract: We recently developed a tracking microscope that can record neural activity of freely behaving Drosophila larvae. However, relating activity and behavior has been complicated by the difficulty of analyzing video recordings of larvae crawling underneath the tracking microscope. Because of the geometry and nature of the microscope, these recordings have low contrast, uneven backgrounds, bright artifacts, and other features that complicate traditional

thresholding based approaches to determining larval posture. To overcome these challenges, we adapted a convolutional neural network developed to characterize the behavior of adult flies to determine the postures of larvae in the tracking microscope. We found that the network could find key points on the larva's body and that high accuracy could be achieved with small amounts of manual training for each larva. We used this system to detect body bending, a key element of the larva's navigational algorithm. Finally, we developed an automated analysis protocol to detect errors made by the network and flag these for manual labeling, decreasing the time it takes to train the network and increasing the overall accuracy of the final labeling.

Disclosures: **A. Nwankpa:** None. **P. McNulty:** None. **R. Wu:** None. **M. Gershow:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.01

Title: WITHDRAWN

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.02

Topic: I.07. Data Analysis and Statistics

Support: NIH grant K23MH050909
Minnesota MnDRIVE (Minnesota's Discovery, Research and Innovation Economy) Medical Discovery Team on Addiction

Title: Validation of Cluster-based Permutation Tests with GLMEs for the Analysis of Event-Related Potentials

Authors: *S. KOENIG[1], A. B. HERMAN[2], D. P. DARROW[3];
[1]Psychiatry and Neurosurg., Univ. of Minnesota, Minneapolis, MN; [2]Psychiatry, [3]Neurosurg., Univ. of Minesota, Minneapolis, MN

Abstract: Time series analysis is critical for understanding brain signals and their relationship to behavior and cognition. Event-Related Potentials (ERPs) are commonly used to analyze brain signals during behavioral tasks as they are relatively simple to compute and reproducible. In order to carry out such analyses a time window must be defined either a priori or empirically. One such method for empirically defining a time window is to use cluster-based permutation

tests (Maris & Oostenveld, 2007). Most often a t-statistic or comparable non-parametric statistic is used to define significant clusters of adjacent time points even when the experimental design is greater than a 1x2 factorial design. Here we propose a method for using Generalized Linear Mixed Effects Models (GLMEs) with cluster-based statistics. GLMEs allow for direct comparison of multiple variables in greater than 1x2 factorial experimental designs, can be easily expanded to include hierarchical components, allow for non-linearities, have less assumptions than normative statistics, are more robust to outliers, and allow for the inclusion of random effects. In Matlab simulations we show that GLMEs are at least as robust as other commonly used statistical measures including t-tests, ANOVA, and simple permutation tests especially in the presence of random effects. Additionally, the significance of the t-statistics provided by GLMEs in Matlab are 99.1% congruent with permuted t-statistics. Finally, we apply GLMEs with cluster-based permutations tests to a previously published data set analyzing face, house, and novelty encoding at ECoG recording sites in human epilepsy patients (Kai, et. al, 2016; Kai, 2019). We find that 17.2% (123/714, n = 14) of recording sites have ERPs selective for faces, houses, image novelty, or some combination thereof similar to previously reported numbers of 15.8% (Kai, et. al, 2016). In summary, our simulations show that GLMEs are at least as good as if not better than commonly used statistical models while also providing greater flexibility and robustness necessary for the analysis of neural data collected in a wide range of experimental designs. We also show that GLMEs with cluster-based permutations tests can be applied to real-world human ECoG data to identify recording sites with ERPs selective for different types of presented images in an experiment with a 2x2 factorial design.

Disclosures: S. Koenig: None. **A.B. Herman:** None. **D.P. Darrow:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.03

Topic: I.07. Data Analysis and Statistics

Support: NIAAA P50 AA026117
NIAAA T32 AA007565

Title: Alterations in functional connectivity within the Default Mode, Salience, and Motor Networks during alcohol cue viewing following alcohol abstinence

Authors: *H. PETERSON[1], R. G. LYDAY[2], C. TOMLINSON[4], W. J. REJESKI[5], S. L. SIMPSON[3], J. H. BURDETTE[6], P. J. LAURIENTI[6];
[1]Wake Forest Univ. Sch. Of Medicin Neurosci. Program, Winston-Salem, NC; [2]Lab. for Complex Brain Networks, Wake Forest Sch. of Med., Winson-Salem, NC; [3]Biostatistical Sci., Wake Forest Sch. of Med., Winston Salem, NC; [4]Biostatistics, Univ. of North Carolina, Chapel Hill, NC; [5]Hlth. and Exercise Sci., Wake Forest Univ., Winston-Salem, NC; [6]Wake Forest Univ. Sch. Med., Winston Salem, NC

Abstract: Alcohol consumption is a common social behavior worldwide, with approximately 70% of US adults reporting alcohol consumption within the past year and more than 5% of US adults meeting diagnostic criteria for an Alcohol Use Disorder (AUD). Periods of abstinence are not well tolerated, especially among those with AUD, although less data is available for moderate-to-heavy drinkers. For those with AUD, exposure to alcohol cues may serve as a stressor that increases risk of relapse, but in moderate-to-heavy drinkers, cue exposure alone may not serve as a relevant psychological stressor. However, exposure to alcohol related images during a period of imposed alcohol abstinence serves as a sufficient stressor in moderate-to-heavy drinkers to study the physiological underpinning of abstinence and what may lead an abstaining individual to begin consuming again, without the dangerous withdrawal symptoms associated with abstinence and AUD. Thirty-nine healthy, non-binging moderate-to-heavy drinkers (56% female) without history of AUD were recruited for this study. On average, participants were 41 years of age and had been consuming alcohol for approximately 25 years; males in this sample consumed an average of 23 drinks per week, while female participants consumed an average of 13 drinks per week. Functional MRI data was collected while participants were exposed to neutral and alcohol related images. Functional brain networks were constructed from fMRI data and community structure of the networks was assessed. Community structure of the Default Mode ($p = 0.0085$) and Salience Networks ($p = 0.0340$) was significantly more consistent across participants during alcohol image viewing compared to neutral image viewing; however, community structure of the Motor Network ($p = 0.0389$) was significantly less consistent. Also, community structure of the Default Mode Network during alcohol image viewing was significantly associated with participants' current ratings of desire for alcohol ($p = 0.0053$). This result was only found during a period of alcohol abstinence, with no significant results found under participants' typical consumption routine. Overall, the findings from this study suggest a reorganization of functional brain networks associated with alcohol image exposure only following short-term abstinence in healthy moderate-to-heavy drinkers. Increased ratings for craving, or desire for alcohol, are associated with AUD diagnosis and severity of drinking symptoms, and we suggest the observed changes in Default Mode community structure organization may be associated with vulnerability to AUD symptom development, although further study is needed.

Disclosures: H. Peterson: None. **R.G. Lyday:** None. **C. Tomlinson:** None. **W.J. Rejeski:** None. **S.L. Simpson:** None. **J.H. Burdette:** None. **P.J. Laurienti:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.04

Topic: I.07. Data Analysis and Statistics

Title: Machine learning approach to searching for effective subsets of the Bayley-III to assess child development

Authors: *W. KIM, G. KANG, J. LIM;
R&D Ctr., Lumanlab Inc., Seoul, Korea, Republic of

Abstract: Early detection of potential risk of neurodevelopmental delay is critical to the mental health and well-being of children. At these times, the brain grows fast, continues to develop, and change into adulthood. This leads to the importance of developing accurate child developmental instruments to measure the risk faster and easier to use than the current instruments. In this study, machine learning (ML) models are used to evaluate one of the most widely used instruments for measuring child development, the Bayley Scales of Infant and Toddler Development (Third Edition, Bayley-III) to evaluate whether only a subset of the Korean version of Bayley-III (K-Bayley-III) can help identify children with developmental delay. A total of 141 toddlers (13-38 months) was participated, excluding toddlers with known developmental delay and psychiatric diseases. A collection of developmental score sheets for all five modules of K-Bayley-III was assembled. Five unique ML regression models were then trained to identify the best ML model for prediction of the developmental scores using the full sets of features of K-Bayley-III. Consequently, the features were ranked using SHapley Additive exPlanations (SHAP). The optimal subsets consisting of the top-n ranked features were obtained from the best prediction model and used to compare the prediction performance to using that of the full set of features. Finally, the prediction results derived from the optimal subset were compared to those using the full sets of K-Bayley-III. By applying ML models tuned for sparse data, the subsets with approximately 57% reduction in the number of features were acquired with negligible loss of prediction. The average R-squared score (R^2) and root mean squared error (RMSE) for the ML models using the optimal subsets were 0.781, and 4.937, respectively. The results were similar with the values of 0.834, and 4.302 for the ML models using the full sets. In particular, the subset including only 7 questions resulted in similar performance of R^2, 0.928 and RMSE, 2.205 when compared to the full set involving 241 questions with the results of 0.936 and 2.067 corresponding to R^2 and RMSE. In this study, the results present that the fewer questions measured using ML models predict similar developmental scores to those obtained using the full sets of questions, suggesting that the subsets of K-Bayley-III as an effective tool for child developmental assessment. Further study including more participants should be considered to improve performance and stability of the ML models identifying the optimal subsets for child developmental assessment.

Disclosures: W. Kim: None. **G. Kang:** None. **J. Lim:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.05

Topic: I.07. Data Analysis and Statistics

Support: New York Obesity and Nutrition Research Center Pilot and Feasibility Grant

Title: Leveraging Epidemiologic Methods to Assess the Neural Correlates of Adiposity Among Underrepresented Youth - a Model for Improving Causal Inference in Neuroimaging Research

Authors: *J. W. COHEN[1,4], D. PAGLIACCIO[2,4], A. RUNDLE[5], A. E. MARGOLIS[3,4];
[1]The Dept. of Psychiatry, [2]The Dept. of Psychiatry,, [3]Div. of Child and Adolescent Psychiatry, Vagelos Col. of Physicians and Surgeons, Columbia Univ., New York, NY; [4]New York State Psychiatric Inst., New York, NY; [5]Dept. of Envrn. Hlth. Sciences,, Mailman Sch. of Publ. Health, Columbia Univ., New York, NY

Abstract: Demographic factors and predictors of interest often associate with participant self-selection into neuroimaging studies as well as attrition or loss to follow up in longitudinal studies. Epidemiologists have developed advanced analytical methods to address these issues and improve causal inference in population health studies, however these methods have only begun to be employed in neuroscience.
To address these shortcomings, we developed a standardized, generalizable and flexible pipeline in R for implementing sensitivity analyses, multiple imputation, and inverse probability weighting (IPW) in neuroimaging research. Specifically, we focus on vertex-wise analyses of cortical thickness, as structural MRI for morphometric analyses is ubiquitously acquired in neuroimaging work. As a proof of concept, we apply this pipeline in N = 286 Black and Latinx adolescents/young-adults (14-21 years old) to examine the effects of socioeconomic disparities on causal inference in the neuroscience of obesity. In a simplified model covarying for age and sex only (N=286), significant positive associations were found between visceral adipose tissue (VAT) and bilateral thickness of the cingulate and negative associations within the fusiform, superiorfrontal, and precentral gyri. Negative associations were observed between VAT and thickness in the cuneus, pars orbitalis, and supramarginal, pars triangularis, postcentral, and middle temporal gyri of the left hemisphere, and the right pars opercularis and lingual gyrus. To avoid sample size restriction when covarying for sociodemographic indicators with missing data (n=111), which eliminated significant effects in most regions, missing covariates were multiply imputed (N=25 iterations). Examining an imputed dataset, findings in the cingulate, pars orbitalis, cuneus, and supramarginal, fusiform, and precentral gyri remained significant whereas associations between VAT and CT in the superior frontal, postcentral, and middle temporal gyri were better accounted for by sociodemographic confounds. In summary, in-depth comparisons of neural correlates conducted with and without multiple imputation via sensitivity analyses demonstrate how failing to implement these strategies negatively impacts the validity of neuroimaging findings. We provide a robust and flexible, publicly available tool to increase more widespread use of epidemiological methods in neuroimaging research and in turn increase rigor and reproducibility of analyses across a wide range of topics.

Disclosures: J.W. Cohen: None. **D. Pagliaccio:** None. **A. Rundle:** None. **A.E. Margolis:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.06

Topic: I.07. Data Analysis and Statistics

Support: NIH Grant MH123192

Title: Cross-frequency coupling analysis by a deep learning network for the detection of absence seizures

Authors: *A. V. MEDVEDEV, B. LEHMANN;
Ctr. for Functional and Mol. Imaging, Georgetown Univ. Med. Ctr., Washington, DC

Abstract: High frequency oscillations are important biomarkers of epileptogenic tissue[1-2]. The interaction of oscillations across different time scales is revealed as cross-frequency coupling (CFC) representing a high-order structure in the functional organization of brain rhythms. New methods of artificial intelligence such as deep learning neural networks can provide powerful tools for automated analysis of EEG including the analysis of the CFC patterns which are likely to reflect different functional states of the brain. Here we present a Stacked Sparse Autoencoder (SSAE) trained to recognize absence seizure activity from the preceding preictal activity based on the CFC patterns within scalp EEG. We used EEG records from the Temple University Hospital database (the TUSZ corpus[3]) with seizures annotated by neurologists. Absence seizures (n = 94) from 12 patients were taken into analysis along with segments of preictal activity. Half of the records was selected randomly for network training and the second half was used for testing. Power-to-power coupling was calculated between all frequencies 2-120 Hz pairwise using the EEGLAB toolbox. The resulting CFC matrices were used as training or testing inputs to the autoencoder. The SSAE network created with MATLAB R2021b consisted of two encoder-decoder networks and the softmax layer with two outputs for binary classification 'seizure vs. preictal'. During training, the network achieved a squared error smaller than 10^{-2} with 400 iterations. The trained network was able to recognize preictal and ictal segments (not used in training) with sensitivity of 97.9%, specificity of 90.0% and the overall accuracy of 96.5%. Our post hoc analysis showed that the major difference between preictal and ictal activity was due to an increase in the power-to-power coupling within beta-gamma bands (13-120 Hz) during seizures. Our results provide evidence that the SSAE neural networks can be used for automated detection of seizures within scalp EEG. Importantly, the trained SSAE network showed generalizability detecting seizures with sensitivity and specificity at or higher than 90% in all patients tested. Deep learning networks can significantly accelerate the analysis of EEG data and increase their diagnostic value.

References

1. Jacobs J, Zijlmans M, Zelmann R, et al. (2010) High-frequency electroencephalographic oscillations correlate with outcome of epilepsy surgery. Ann Neurol 67:209-20
2. Staba RJ, Stead M, Worrel GA (2014) Electrophysiological biomarkers of epilepsy. Neurotherapeutics 11:334-46
3. Shah V, Von Weltin E, Lopez S, et al. (2018) The Temple University Hospital seizure detection corpus. Front Neuroinform 12:83

Disclosures: A.V. Medvedev: None. **B. Lehmann:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.07

Topic: I.07. Data Analysis and Statistics

Support: This study was funded by CHDI Foundation

Title: Predictive modeling of Huntington's disease unfolds thalamic and caudate atrophy dissociation: a multi-study validation

Authors: *E. CASTRO[1], P. POLOSECKI[1], D. PUSTINA[2], A. WOOD[2], C. SAMPAIO[2], G. A. CECCHI[1];
[1]Healthcare and Life Sci., IBM Res., Yorktown Heights, NY; [2]CHDI Management/CHDI Fndn., Princeton, NJ

Abstract: Huntington's disease (HD) is a neurodegenerative disorder characterized by cognitive, behavioral, and motor decline. Current treatment is only palliative and disease damage is irreversible, so potential therapeutics should aim to modify disease course and prevent the development of clinical disease. To this end, sensitive and reliable markers of disease progression are needed for the period before clinical motor diagnosis (before-CMD). One of the most reliable markers before-CMD is striatum atrophy, but volumetric reductions have also been reported in other subcortical structures. The study of the covariation of these structures could improve the detection of disease-related longitudinal progression before-CMD, provide mechanistic insights of the disease, and potentially be used to obtain accurate prospective estimates of atrophy in HD. We analyzed data from 337 before-CMD individuals, 236 healthy controls and 95 early after-CMD individuals from three studies (TRACK-HD, PREDICT-HD, IMAGE-HD) and used 9 subcortical regions volumes in two analyses. First, we discriminated before-CMD progression from healthy-control trajectories by integrating volume changes from these regions using logistic regression classification. In addition, we detected relevant subcortical interactions by evaluating classification weights and partial correlations between them. Second, we estimated prospective atrophy before-CMD and early after-CMD by considering the influence of a region's present volume over the future volume of another one (Granger causality) using vector autoregression. Before-CMD progression was robustly detected across studies. Indeed, detection of before-CMD progression improved when multiple structures were integrated, as opposed to analyzing the striatum alone, likely due to the reduced partial correlation between caudate and thalamic volume change before-CMD. Our multivariate atrophy prediction model found a thalamus-caudate association that is consistent with this pattern, which yields an improved caudate atrophy prediction in early after-CMD. This study is the first attempt to validate before-CMD multivariate subcortical change detection across studies and to do multivariate prospective atrophy prediction in HD. These models achieve improved performance by detecting a dissociation between caudate and thalamic atrophy trajectories, and provide a possible mechanistic understanding of the dynamics of HD.

Disclosures: **E. Castro:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; This study was funded by CHDI Foundation. **P. Polosecki:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; This study was funded by CHDI Foundation. **D. Pustina:** None. **A. Wood:** None. **C. Sampaio:** None. **G.A. Cecchi:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; This study was funded by CHDI Foundation.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.09

Topic: I.07. Data Analysis and Statistics

Support: Korea Republic of, Ministry of Science and ICT (22-BR-03-01)

Title: Machine Learning-based classification using electroencephalographic multi-paradigms between Drug-Naive patients with depression and healthy controls

Authors: *K.-I. JANG, C. LEE;
Korea Brain Res. Inst. (KBRI), Daegu, Korea, Republic of

Abstract: Objective: Electroencephalography (EEG) biomarker as a supplementary diagnostic tool has been used with lacks practical use in psychiatry. There is an issue with the lack of diagnostic ability of EEG for classification between patients with major depressive disorder (MDD) and healthy controls (HCs). The single EEG paradigm was mainly investigated in the previous studies because it represents a specific pathological attribute. However, MDD is a heterogeneous psychiatric disorder with complex pathologies. Using multiple EEG paradigms to classify patients with MDD and healthy individuals is essential in the practical implications of psychiatry. Furthermore, machine learning technique with EEG signals has raised in psychiatry. A better classification performance with the machine learning technique is still required in clinical practice. The present study tested the classification performance using the multiple EEG paradigms in drug-naïve patients with MDD and healthy controls (HCs). Materials and Methods: Thirty-one drug-naïve patients with MDD and 31 HCs were recruited. Resting-state EEG (REEG), loudness dependence of auditory evoked potentials (LDAEP), and P300 were recorded in all participants. Linear discriminant analysis (LDA) and support vector machine (SVM) classifier with t-test-based feature selection were used for classifying patients and HCs. Results: The highest accuracy was 94.52 % when 14 selected features including 12 P300 and 2 LDAEP features were layered. The final accuracy was 88.71% with SVM classifier for 38 selected

features were layered. All selected features included 14 P300, 19 LDAEP, and 5 REEG features. Furthermore, for each REEG, P300A, and LDAEP, the best accuracies were 71.57%, 87.12%, and 83.87%, respectively. Conclusions: The multiple EEG paradigms would be more beneficial for classifying drug-naïve patients with MDD and HCs in comparison to the single EEG paradigm.

Disclosures: K. Jang: None. **C. Lee:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.10

Topic: I.07. Data Analysis and Statistics

Support: U01AG068057

Title: Using a 3D Convolutional Neural Network to Compare Brain Age Prediction based on T1-weighted and Diffusion-Weighted MRIs

Authors: *S. THOMOPOULOS, T. CHATTOPADHYAY, T. M. NIR, H. ZHENG, E. NOUROLLAHIMOGHADAM, N. JAHANSHAD, P. M. THOMPSON;
USC, Los Angeles, CA

Abstract: Machine learning methods have been used for over a decade to estimate a person's age from their brain MRI scan: the difference between this predicted age (their "BrainAge") and their chronological age is linked with future clinical decline, dementia, and mortality, making it a promising biomarker of brain aging. BrainAge is typically predicted from T1-weighted brain MRI (T1w); here we tested whether we could predict chronological age with better accuracy using BrainAge predicted from diffusion weighted images (DWI), a variant of brain MRI sensitive to microstructural aging and degeneration. Specifically, we assessed the predictive capabilities of deep learning models using a 3D Convolutional Neural Network trained on 3D whole brain T1w and DWI-derived diffusion tensor imaging (DTI) mean and radial diffusivity (MD/RD) and fractional anisotropy (FA) maps from cognitively normal (CN) participants from the Alzheimer's Disease Neuroimaging Initiative (ADNI). We analyzed T1w and DWI images from 547 subjects (CN, mean age: 73.6+/-7.8 yrs., 325 F/ 222 M). T1w images were nonlinearly registered to the DWI, which were subsequently warped to a common template. The data was split into independent training, validation and testing sets in ratio 70:20:10. We used 3D CNN, with three convolution blocks of filter sizes 32, 64 and 128, instance normalization and max pooling. We trained it for 50, 75 and 100 epochs, with learning rate $1x10^{-4}$, and batch size 8. Adam optimizer and mean square error loss function were used during training. To deal with overfitting, dropout between layers and early stopping were used. Test performance was assessed using mean square error and mean absolute error to compare results for different modalities. For chronological age prediction, T1w derived BrainAge had the poorest performance (MSE: 48.85,

MAE: 5.64) on test dataset at 50 epochs. FA maps gave the best MSE of 19.53 and best MAE of 3.36 for 100 epochs. For BrainAge Prediction, diffusion-weighted images may offer better results for deep learning algorithms involving 3D CNNs, in comparison to T1w. Future work in additional cohorts, with additional diffusion metrics, will be valuable.

Disclosures: S. Thomopoulos: None. **T. Chattopadhyay:** None. **T.M. Nir:** None. **H. Zheng:** None. **E. Nourollahimoghadam:** None. **N. Jahanshad:** None. **P.M. Thompson:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Research grant from Biogen, Inc. for research unrelated to this abstract.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.11

Topic: I.07. Data Analysis and Statistics

Support: NIMH intramural research program 1ZIAMH002949

Title: A sex-stratified analysis of autosomal influences on neuroanatomical variation in the UK Biobank

Authors: *R. SHAFEE, S. LIU, D. MORACZEWSKI, A. G. THOMAS, A. RAZNAHAN; Natl. Inst. of Mental Hlth. (NIMH)/NIH, Bethesda, MD

Abstract: Humans show reproducible sex differences in brain anatomy and risk for psychopathology. Interindividual variation in neuroanatomy and behavior are both heritable and can show partially overlapping genetic effects. Understanding potential sex-differences in the genetic architecture of brain anatomy may shed light on biological factors underlying sex-differences in psychopathology. Moreover, the hypothesis that males and females may show differences in the genetic architecture of brain anatomy is motivated by existence of known sex-biases in biological factors that influence brain anatomy such as sex-steroid signaling and sex chromosome complement. In prior work, we have compared X-chromosome influences on human brain anatomy in males and females through the lens of common genetic variation. Here we systematically screen for potential sex-biases in the influence of common autosomal genetic variants on brain anatomy using T1-weighted magnetic resonance imaging (MRI) derived traits in the UK Biobank sample (15183 males, 16767 females of white British ancestry, age = 64.3 +/- 7.5 yrs). Individual level genotype data were used to perform sex-stratified genome-wide association analyses on cortical thickness, area and volume measures of 74 regions on each hemisphere (Destrieux atlas, FreeSurfer). Brain traits were adjusted for head size, age, age^2 and scanner and location confounders and the first 10 ancestry principal components were included in the association model. Genetic relatedness matrices were constructed using GCTA in each sex

as well as for the entire sample. After correcting for multiple-testing across SNPs (single nucleotide polymorphisms), brain regions and morphometric phenotypes we failed to find any evidence of: sex-differences in regional SNP-based heritability; deviation of genetic correlation between males and females from 1; individual common SNPs with a sex-biased effect on anatomical variation. Taken together, these results suggest a lack of any substantial sex-differences in the architecture of autosomal genetic influences on human brain anatomy as measured by MRI.

Disclosures: R. Shafee: None. **S. Liu:** None. **D. Moraczewski:** None. **A.G. Thomas:** None. **A. Raznahan:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.12

Topic: I.07. Data Analysis and Statistics

Support: Gilead Sciences, Inc.

Title: Computer-aided prediction of severe COVID mortality utilizing neurologic symptoms

Authors: *B. RAJWA[1], M. DUNDAR[2], C.-H. FANG[1], M. A. NAVED[1], A. GRAMA[1], B. A. KHAN[3,4], J.-C. ROCHET[1];
[1]Purdue Univ., West Lafayette, IN; [2]Indiana University-Purdue Univ. Indianapolis, Indianapolis, IN; [3]Indiana Univ. Sch. of Med., Indianapolis, IN; [4]Regenstrief Inst., Indianapolis, IN

Abstract: As the world emerges from the pandemic caused by SARS-CoV-2, there is a need to understand factors that determine the long-term effects of COVID-19, as well as the diagnostic features that may be used to predict the occurrence of severe cases. Approximately 20% of SARS-CoV-2 infections lead to acute respiratory distress syndrome (ARDS) caused by the harmful actions of inflammatory mediators. Patients with severe COVID-19 are often afflicted with neurologic symptoms, and individuals with pre-existing neurodegenerative disease (ND) have an increased risk of severe COVID-19. Although collectively, these observations point to links between severe COVID-19 and ND (and vice versa), little is known about the mechanisms. We hypothesize that a *bidirectional relationship exists between COVID-19 and ND:* individuals with pre-existing ND are likely to have an increased risk of severe COVID-19 involving ARDS and neurologic complications, and COVID-19 survivors (particularly individuals having shown CNS symptoms) are predicted to have an elevated risk of ND. To test this hypothesis, we conducted a study to determine (i) the relationship between the lethality of COVID-19 and CNS symptoms, (ii) the connection between the COVID severity and pre-existing dementia, and (iii) the degree of cognitive decline in individuals who have recovered from severe COVID-19. This presentation describes the preliminary findings from the first part of the study, in which the electronic health records of ~400 patients with severe COVID were analyzed to identify the

clinical characteristics most predictive of COVID-19 fatalities. The analysis was conducted by training a regularized logistic regression classifier that serves as a machine learning model with an embedded feature selection capability. The classifier's training revealed that a small ensemble of clinical features, including clinical characteristics associated with acute neuroinflammation, could predict death with an accuracy greater than 0.80 (expressed as the area under the ROC curve of the classifier). Following our preliminary analysis, we constructed a partial correlation network that will serve as a canvas for causal inference linking neurologic symptoms with disease mortality. Aligned with observations made by other researchers who demonstrated links between dysregulation of the immune response and cognitive COVID-19 symptoms, our data suggest that neuroinflammation is a marker of severe disease with a likely fatal outcome. When completed, the study results will provide a foundation to justify developing treatments targeting neuroinflammatory effectors found to mediate COVID-19 susceptibility.

Disclosures: B. Rajwa: None. **M. Dundar:** None. **C. Fang:** None. **M.A. Naved:** None. **A. Grama:** None. **B.A. Khan:** None. **J. Rochet:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.13

Topic: I.07. Data Analysis and Statistics

Support: NIH/NIA AG073356

Title: Use of Canonical Correlation Analysis for the study of adaptation capacity

Authors: *L. PERALTA MALVAEZ[1], A. TURNBULL[2,1], M. ANTHONY[2,1], E. ADELI[1], F. V. LIN[1];
[1]Psychiatry and Behavioral Sci., Stanford Univ., Stanford, CA; [2]Brain and Cognitive Sci., Univ. of Rochester, Rochester, NY

Abstract: Locus of control (LOC) describes a personality trait based on whether an individual thinks that they themselves (internal LOC) or external factors (external LOC) have more influence on their lives. Adaptation capacity reflects the ability of the autonomic nervous system to respond to both physical and cognitive stressors, and it has been proposed that individuals with a more internal LOC are better able to respond to stressors via a greater adaptation capacity. Older adults at risk for dementia have been shown to shift their reliance of LOC from internal to external, potentially leading to reduced resilience. To better understand whether a shared neural and physiological marker of adaptation capacity was related to internal LOC, we used Canonical Correlation Analysis (CCA), which maximizes the linear correspondence between two sets of variables, to obtain correlated features from heart rate variability (HRV) and resting state functional connectivity (FC). We collected resting and task-based HRV measures, as well as resting state MRI, in 65 older adults (*mean age = 74.71; SD = 7.30*), and measured LOC using

the Personality in Intellectual Aging Contexts (PIC) Inventory Control Scales-short. Following segmentation of HRV data into sliding windows of 30 seconds and calculation of within- and between-network FC, CCA identified a shared component (CC1) characterized by a correlation of 86.9% between features from HRV and FC matrices that was highly similar when constraining the CCA using HRV or FC. HRV features in CC1 were predominantly provided by segments 6, 12, and 14 (corresponding to minutes 2.5, 5.5, and 6.5 of 8 minutes in total). Connections between visual, somatomotor, limbic, and frontoparietal regions contributed most strongly to the FC features of CC1. To understand if this shared component was associated with LOC, we performed bivariate correlations. Internal PIC was significantly related to CC1 constrained by both HRV and FC (RMSSD: $r = 0.266$, $p = 0.031$; FC: $r = 0.289$, $p = 0.019$); there was no relationship to external LOC (RMSSD: $r = 0.095$, $p = 0.447$; FC: $r = 0.038$, $p = 0.758$). CC2 and CC3 were not correlated with internal or external PIC, suggesting the relationship was specific to the most strongly correlated shared component. These outcomes suggest that CC1, a shared neural and physiological marker of adaptation capacity, may act as a biomarker for intact internal LOC. Future work should attempt to better understand the neurophysiological basis of this component in relation to brain aging and resilience.

Disclosures: L. Peralta Malvaez: None. **A. Turnbull:** None. **M. Anthony:** None. **E. Adeli:** None. **F.V. Lin:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.14

Topic: I.07. Data Analysis and Statistics

Support: The Clarke's Family Foundation

Title: Heritability of global and local graph properties of the intrinsic functional connectivity network

Authors: H. POURMOTABBED[1], D. CLARKE[1], C. CHANG[2], ***A. BABAJANI-FEREMI**[1]; [1]Neurol., The Univ. of Texas at Austin, Austin, TX; [2]Electrical and Computer Engin., Vanderbilt Univ., Nashville, TN

Abstract: Graph analysis of the intrinsic functional connectivity network can provide information about the organization of the brain. Brain network properties are associated with healthy behavior and cognition (Farahani et al., 2019). Neurological disorders are characterized by alterations in network architecture (Hojjati et al., 2019). Previous studies have examined resting-state functional MRI (rfMRI) to show that global network properties are influenced by genetic factors (Sinclair et al., 2015). The purpose of this study was to investigate heritability of rfMRI global and local network properties. This study included ICA-FIX cleaned rfMRI data of 1003 healthy subjects (28.7 ± 3.7 yr, 469 male) of the HCP1200 release. The subjects consist of

120 monozygotic twin pairs, 65 dizygotic twin pairs, and 152 non-twin families. The CONN toolbox was used to calculate the correlation between the regions of the Brainnetome atlas. Global (global efficiency [GE], characteristic path length [CPL], transitivity [T], synchronizability) and local (strength, clustering coefficient, eigenvector centrality, nodal efficiency) graph measures were computed. The SOLAR-Eclipse and APACE toolboxes were used to evaluate heritability (h^2) of the global and local measures, respectively, after adjusting for in-scanner motion, age, and sex. GE, CPL, and T for positive correlations and all global measures for negative correlations were significantly heritable ($p < 0.05$, false discovery rate [FDR]-corrected) (**Fig. 1)**. All local measures for positive correlations were significantly heritable ($p < 0.05$, FDR-corrected) in specific brain regions (**Fig. 1)**. The most consistently significant regions included the thalamus, basal ganglia, left amygdala, frontal lobe, superior temporal gyrus, entorhinal cortex, postcentral gyrus, cingulate gyrus, right insula, and right occipital lobe. Our results indicate that global and local network characteristics are influenced by genetic factors and have the potential to serve as biomarkers in human recognition and genetic profiling of neurological disorders.

Heritability h^2 of Global Graph Measures

Positive Correlations	
GE	**0.628 (0.046)**
CPL	**0.661 (0.043)**
T	**0.533 (0.053)**
S	0.060 (0.061)
Negative Correlations	
GE	**0.462 (0.055)**
CPL	**0.469 (0.054)**
T	**0.331 (0.058)**
S	**0.289 (0.062)**

Bold entries denote graph measures that are significantly heritable ($p < 0.05$, FDR-corrected).

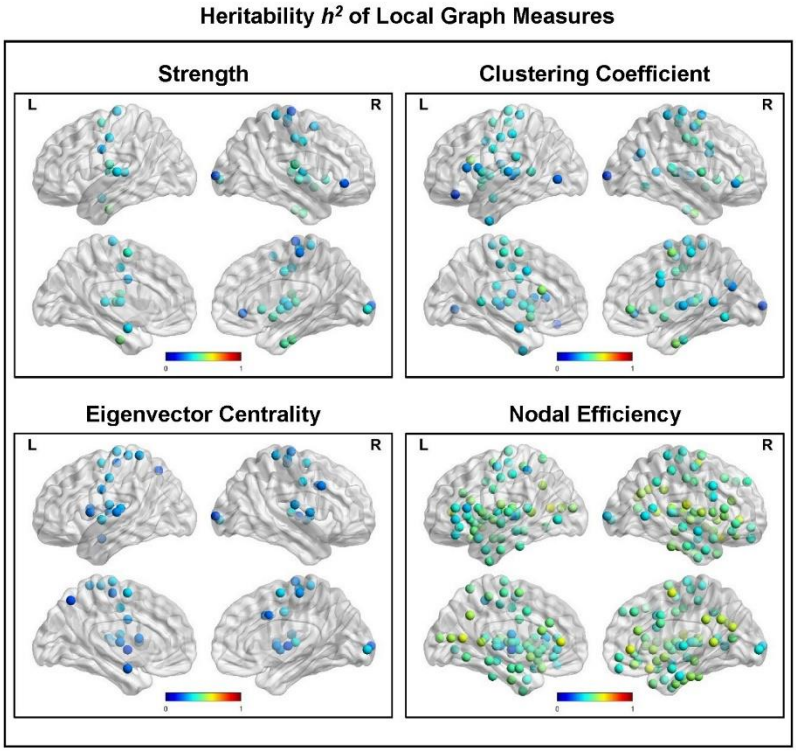


Fig. 1. [Left] Heritability, h^2 estimate (standard error), of four global graph measures (global efficiency [GE], characteristic path length [CPL], transitivity [T], synchronizability [S]). [Right] Brain regions whose local graph measures for positive correlations were significantly heritable ($p < 0.05$, false discovery rate [FDR]-corrected). The h^2 value ranges from 0 to 1 and corresponds to the proportion of phenotypic variance explained by genetic factors.

Disclosures: H. Pourmotabbed: None. **D. Clarke:** None. **C. Chang:** None. **A. Babajani-Feremi:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.15

Topic: I.07. Data Analysis and Statistics

Support: NIH Grant R00-MH11748

Title: Examining age-related differences in frequency content of resting-state fMRI signals

Authors: *S. M. BAILES, L. D. LEWIS;
Boston Univ., Allston, MA

Abstract: fMRI is widely used to investigate changes in brain function across the lifespan. However, interpreting fMRI data collected from aging populations poses a particular challenge because both the neural and vascular dynamics that drive the blood-oxygenation-level-dependent (BOLD) signal change in healthy and pathological aging. Distinguishing the respective effects of these neural and vascular dynamics on the BOLD signal is critical for proper comparison of older and younger populations using BOLD fMRI. Recent evidence suggests that the frequency spectra of resting-state fMRI signals contain information about local differences in neurovascular coupling. Using a set of quantitative measures reflecting the relative contributions of low- and high-frequency power, it is possible to characterize and predict local differences in the temporal dynamics of the hemodynamic response function (HRF). We aim to test whether these resting-state spectral features can reveal age-related differences in neurovascular coupling. We used a subset of the Human Connectome Project Aging dataset to test whether the spectral properties of resting-state fMRI signals differ in older individuals. We first extracted summary features representing the relative amplitude of low- vs. high-frequency BOLD signals in the resting state. Then, we split the subjects into two age groups (young: ≤50 years old, N = 50; older: N = 100) and compared the relative amplitude of these features both across the whole brain and on a regional level. We found that the average magnitude of these features across the whole brain and in the majority of brain regions were higher in the younger group, with multiple features and regions demonstrating significant differences (Wilcoxon rank sum test, $p<0.05$). The standard deviations for each feature were consistently higher in the older population, suggesting greater individual variability with age. Taken together, these results suggest age-related declines in hemodynamic spectral properties that are more pronounced in a subset of individuals. Our observed age-related decrease in low-frequency power aligns with prior work that has reported a shift towards higher frequency resting-state fMRI fluctuations in older adults that is not mediated by EEG power or frequency. Conversely, there is evidence of overall slowing of EEG activity with age. These conflicting findings suggest that the observed changes in spectral content are due to a mixed effect of changing vascular and neuronal factors with age, and that it is critical to consider age-related changes in neurovascular coupling when interpreting differences across younger and older populations using BOLD fMRI.

Disclosures: S.M. Bailes: None. **L.D. Lewis:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.16

Topic: I.07. Data Analysis and Statistics

Support: R01 MH122389
R01 MH115046

Title: Multi-echo fMRI for precision functional brain imaging in newborn human infants

Authors: *J. MOSER[1], T. MADISON[1], S. KOIRALA[1], L. A. MOORE[1], E. FECZKO[1], S. SUNG[1], J. T. ELISON[1], C. M. SYLVESTER[2], D. A. FAIR[1];
[1]Univ. of Minnesota, Minneapolis, MN; [2]Washington Univ. in St. Louis, St. Louis, MO

Abstract: Efforts in precision functional mapping of adult brain networks have opened up a new avenue for precision medicine. Similar efforts in infants remain sparse because infant functional data collection is generally difficult, complicated by the extended data collection time needed for generating reliable infant functional connectivity networks. The use of multi-echo (ME) functional magnetic resonance imaging (fMRI) has shown reliable signals in shorter recording times in adults. In this pilot investigation, we examined two methods to improve feasibility of precision functional mapping in infants: ME fMRI and NORDIC (NOise reduction with DIstribution Corrected PCA) thermal noise reduction. T2 decay curves, which form the basis of fMRI, are slower in newborns than infants and adults, implying that optimal echo times (T2*) vary with age. ME fMRI offers the possibility to account for differences in T2* between participants and tissue types as data of all echos are optimally combined using a weighting scheme based on the T2* values. We compared data from ME fMRI recordings with five echos (14ms, 38ms, 63ms, 88ms, 113ms) in two newborns, one two-year-old and one adult. In one newborn we examined connectivity reliability by comparing connectivity matrices generated from one half of the data to matrices from various minutes of randomly chosen data (100 permutations) from the other half of the data (using 84min of low motion data). We see longer T2* times in infants with a broader spread of values across different brain regions, particularly in newborns. Cortical grey matter shows a median of 79.9ms (MAD=32, Q1=48.9ms, Q3=104.2ms) and 68.2ms (MAD=28.59, Q1=41.2ms, Q3=93.9ms) in the newborns, 67ms (MAD=11.66, Q1=57ms, Q3=74.9ms) in the two-year-old and 47.6ms (MAD=11.84, Q1=39.8ms, Q3=54.4ms) in the adult. Visual inspection of dense connectivity matrices generated with optimally combined ME and single echo (echo time 37ms) data from the same newborn shows an advantage of ME data acquisition particularly in areas with very short or very long T2*. Reliability of connectivity matrices reaches a mean value of r=0.8 with 40min of data and r=0.84 with additional NORDIC denoising. Values reach a plateau using ~15min of data (mean r=0.77, min=0.75, max=0.79) which was further elevated with NORDIC (mean r=0.82, min=0.8, max=0.84). This pilot investigation shows benefits of ME data acquisition and NORDIC denoising for attempting precision functional imaging in newborn infants. We plan to

substantiate our results by collecting data in further infants and additionally comparing reliability values between precision imaging data collected with single echo and ME fMRI sequences.

Disclosures: **J. Moser:** None. **T. Madison:** None. **S. Koirala:** None. **L.A. Moore:** None. **E. Feczko:** None. **S. Sung:** None. **J.T. Elison:** None. **C.M. Sylvester:** None. **D.A. Fair:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.17

Topic: I.07. Data Analysis and Statistics

Title: To Identify neural differences in threat responsiveness amongst Aggressive vs non-Aggressive participants using a machine learning approach

Authors: *A. MATHUR[1], M. DOBBERTIN[2], K. BLAIR[3], J. BASHFORD-LARGO[4], J. ELOWSKY[4], A. DOMINGUEZ[4], S. BAJAJ[4], R. R. BLAIR[5];
[1]Multimodal Clin. Neuroimaging Lab. (MCNL), [2]Child and Adolescent Psychiatric Inpatient Ctr., [3]1Multimodal Clin. Neuroimaging Lab. (MCNL), Ctr. for Neurobehavioral Res., [4]Multimodal Clin. Neuroimaging Lab. (MCNL), Ctr. for Neurobehavioral Res., Boys Town Natl. Res. Hosp., Boys Town, NE; [5]Child and Adolescent Mental Hlth. Centre, Mental Hlth. Services, Copenhagen, Denmark

Abstract: Prior univariate analysis-based research has associated high levels of aggression in adolescents with atypical neural responsiveness within the bilateral inferior frontal gyrus (IFG), right inferior parietal lobe (IPL), right superior/middle temporal gyrus (STG/MTG), and right uncus. In this study, we examined this association via advanced machine learning (ML) approaches. Ninety-eight adolescents (68 males) participated in a looming threat task; 49 aggressive (mean age=16.14 (*s.d.*=1.77), 32 males) and 49 non-aggressive (mean age=16.06 (*s.d*=1.33), 36 males). We employed two distinct ML data analysis approaches. (1) Region of interest (ROI)-based: 41 ROIs showing threat responsiveness were selected from an independent sample of healthy subjects. The Z-scores obtained for each subject from trial-wise data for 41 ROIs were used as inputs for the least absolute shrinkage and selection operator for feature selection. Thereafter, a support vector machine algorithm was used to classify the two groups using a nested cross-validation framework. (2) Searchlight analysis: AFNI trial-by-trial beta maps for each subject were subjected to searchlight analysis. Classification accuracy maps were generated for each subject using a linear discriminant analysis classifier and a leave-one-out cross-validation method with an n-fold partitioning scheme. A two-sample t-test was run across individual accuracy maps to identify regions where threat responsiveness differed significantly between groups. Both analysis approaches showed convergent results with classifiers identifying regions including bilateral IFG/mid frontal gyrus, subcallosal gyrus, right anterior/mid/posterior cingulate cortex, bilateral postcentral gyrus/precuneus/IPL, bilateral STG/MTG/parahippocampal gyrus/uncus, and bilateral insula (accuracy 73.37%, sensitivity 76.92% and specificity 71.68%).

In conclusion, the current data and the proposed ML approach reveal brain regions associated with increased adolescent aggression.

Disclosures: **A. Mathur:** None. **M. Dobbertin:** None. **K. Blair:** None. **J. Bashford-Largo:** None. **J. Elowsky:** None. **A. Dominguez:** None. **S. Bajaj:** None. **R. R. Blair:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.18

Topic: I.07. Data Analysis and Statistics

Title: An insight into language development in children during the COVID-19 pandemic

Authors: *G. KANG, J. LIM;
R&D Ctr., Lumanlab Inc., Seoul, Korea, Republic of

Abstract: It is emerging that the COVID-19 pandemic has disproportionately affected neurodevelopment of children. The implementation of preventive strategies such as wearing mask, restricting social distance, and closing daycare centers may unintentionally influenced child development, particularly in communication and language skills. This study aims to investigate association between the COVID-19 pandemic and delay in language developmental functioning of infants and toddlers. From January to April in 2022, children aged between 13 and 38 months were assessed for the developmental level of language skills by clinical staff using the most widely used instruments such as (Korean version of) Bayley Scales of Infant and Toddler Development, Third Edition (K-Bayley-III) and vineland. Children with known neurodevelopment diseases or history of the treatment for developmental delay were excluded for the study before the assessments. As a result, a total of 141 children remained for the study. At the end of the data collection, scale scores of K-Bayley-III and vineland were analyzed to determine if there was delay in development of language skills. The results showed that there were 30% of children, whose language development scores were below the average obtained from normative data of K-Bayley-III. None of these children had any medical history that would affect them in language development. In details, 76% of the children, who had low scores for the language skills showed lower scale scores than the average in the receptive communication, one of subscale of the language skill. Similarly, the scale scores of the other subscale, expressive communication, were also shown to be lower in 69% of the children below the average in language development. Interestingly, it was observed that 67% of the children with lower scale scores in language development were male. Finally, there was association between the language and the subtype of adaptive behavior, communication, in which 62% of children with lower language skills than the average exhibited scale scores in adaptive behavior-communication below the average. Language development normally occurs during early childhood in an achievement of receptive and expressive skills gradually. The study presented here indicates the potential risk of the COVID-19 pandemic on the language development in children. Although

more studies are required to characterize the long-term effects of the pandemic on the child development, public health strategies associated with parents and clinicians for this matter should be considered.

Disclosures: G. Kang: None. **J. Lim:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.19

Topic: I.07. Data Analysis and Statistics

Support: NIH UH3 NS095495

Title: Cycle detection in sparse, irregularly sampled, long-term behavioral timeseries: Basis pursuit denoising of ecological momentary assessments in people with epilepsy

Authors: *I. BALZEKAS[1], J. TRZASKO[1], G. YU[1], P. E. CROARKIN[2], G. A. WORRELL[1]; [2]Psychiatry, [1]Mayo Clin., Rochester, MN

Abstract: Rationale: Multiday periodicities exist in numerous biological processes with neuro-behavioral implications, including cycles of seizure risk. Ecological momentary assessments (EMAs) are behavioral measures wherein patients self-report symptoms in the present moment. The identification of multiday rhythms in EMAs is complicated by their sparse, irregular sampling, which invalidates many classic statistical methods that assume continuity or consistent repeated measures. We borrowed a model from digital compression methodologies, basis pursuit denoising (BPDN), to identify oscillations in sparsely and irregularly collected EMA data from patients with temporal lobe epilepsy (TLE). Methods: Three patients with TLE participated in this study as part of a larger investigational device trial of deep brain stimulation for drug resistant TLE. Participants were prompted to complete a 12-item Likert scale inventory of depression and anxiety symptom severity, one to four times per week, on a random day and time. We designed a BPDN model that yielded spectral coefficients representing component oscillations in the EMA timeseries, given the EMA scores, temporal spacing between samples, and a discrete cosine transform (DCT) basis as inputs. Using simulated and real data, parameter sweeps and model-based quality metrics accommodating both measurement and model error were employed to select optimal hyperparameters for each participant. Oscillations were deemed significant if their associated DCT coefficients exceeded the 99th percentile of the output distribution calculated by randomly shuffling the timeseries and recalculating the DCT coefficients 1000 times. Results: Strong participant engagement yielded 460 cumulative entries representing 5 years of data in ambulatory subjects living in their natural environment. In simulation and with real data, BPDN successfully recovered component oscillations. BPDN yielded significant, participant-specific cycles around 1 day, 2-4 weeks, and up to 70 days. Conclusions: Assuming adequate data and scientific rationale to anticipate an underlying

behavioral periodicity, BPDN-based models are a viable method to identify cycles in sparsely and irregularly sampled behavioral timeseries. This may be a powerful tool to evaluate cycles of behavior in long-term studies with ambulatory patients.

Disclosures: **I. Balzekas:** Other; Cadence Neuroscience Inc. **J. Trzasko:** Other; Mayo Clinic. **G. Yu:** Other; McGill University. **P.E. Croarkin:** Other; P. E. Croarkin has received research grant support from Neuronetics, Inc., NeoSync, Inc., and Pfizer, Inc. He has received grant in-kind (equipment, supply, and genotyping support for research studies. **G.A. Worrell:** Other; GW has licensed intellectual property developed at Mayo Clinic to NeuroOne, Inc. GW is an investigator for the Medtronic Deep Brain Stimulation Therapy for Epilepsy Post-Approval Study (EPAS) and for.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.20

Topic: I.07. Data Analysis and Statistics

Support: CBIR15MIG004

Title: Fractal analysis for estimating Hausdorff dimension from MRI white matter that effectively differentiates TBI from normal brain

Authors: *E. SUVISESHAMUTHU[1], C. SERPA[2], S. H. SALEH[1], V. SHENOY HANDIRU[1], G. H. YUE[1];
[1]Kessler Fndn., West Orange, NJ; [2]Inst. Superior de Engenharia de Lisboa and CMAFcIO - Ctr. de Matemática, Aplicações Fundamentais e Investigação Operacional, Lisbon, Portugal

Abstract: A recently proposed fractal analysis approach (Serpa, 2022) is applied to estimate the fractal dimension (FD) of segmented brain white matter (WM) MRI image in healthy and traumatic brain injury (TBI) subjects to investigate the differences among the two groups. The WM volume is extracted from the structural MRI data with an image processing algorithm. The number of voxels pertaining to WM are then summed up separately along the three axes of the Cartesian coordinate system. The row and column of each matrix with the largest sum of array elements are considered as the candidate sequences for FD estimation. Each one of these sequences deduced from the WM-MRI data is fit to a mathematical function, which has a fractal structure and is the solution of a system of equations. Such a fractal function is known to characterize the self-similarity at the fractal levels. The fractal function that approximates the real data is estimated with a regression procedure in two steps: first, the sum of square residuals (SSR) between a formally defined function and the real values will be computed; next, the critical points will be sought, where the partial derivatives of SSR vanish, meaning that the error is minimized. The coefficients of the fractal function are therefore regarded as the parameters to be estimated by the fractal regression method. As there is no explicit analytical solution for the

regression system, the fitting goal is achieved by a numerical approach implemented through a software, namely Fractal Real Finder (Serpa, 2022). The theoretical properties of the underlying fractal function allow us to obtain a more reliable estimate of Hausdorff (fractal) dimension compared to traditional FD estimation approaches and render a better characterization of the fractal structure of the data (Serpa & Buescu, 2017, Buescu & Serpa, 2019). The preliminary results with five healthy and five TBI subjects show that some specific MRI-derived sequences corresponding to both groups have markedly different FD estimates. Future research is to include more MRI data from either category and to perform statistical tests to confirm our findings and to compare the results with those obtained with the box-counting FD estimation approach.
References Serpa, C. (2022). Affine fractal least squares regression model. Fractals. Accepted. Serpa, C., & Buescu, J. (2017). Constructive solutions for systems of iterative functional equations. Constructive Approximation, 45(2), 273-299. Buescu, J., & Serpa, C. (2019). Fractal and Hausdorff dimensions for systems of iterative functional equations. Journal of Mathematical Analysis and Applications, 480(2), 123429.

Disclosures: E. Suviseshamuthu: None. **C. Serpa:** None. **S.H. Saleh:** None. **V. Shenoy Handiru:** None. **G.H. Yue:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.21

Topic: I.07. Data Analysis and Statistics

Title: Bibliometric study of the relationship between calcium channels and epilepsy in Latin America

Authors: *N. GALLARDO LÓPEZ[1], C. RUBIO[1], V. MENA[1], E. URIBE[1], D. FLORES[1], A. LEE[3], M. RUBIO[2];
[1]Neurofisiologia, instituto nacional de neurologia y neurocirugia, ciudad de mexico, Mexico; [2]Neuroquimica, instituto nacional de neurologia y neurocirugia, Ciudad de México, Mexico; [3]Hosp. Angeles Pedregal, Ciudad de México, Mexico

Abstract: Bibliometrics is a field of research that uses quantitative analysis to understand better the impact of the results of the scientific literature. On the other hand, epilepsy is considered chronic neuropathology that affects around 50 million people worldwide, which makes it one of the most common neurological diseases in Latin America. The etiology of this pathology is broad; considering the genetic causes have found a relationship between calcium channels and the generation of epileptic seizures. A search of the scientific production on this topic was carried out considering the database provided by Scopus, from the beginning of the publications on this topic, until March 2022. The variables studied were type of publication, number of citations, area (clinical/experimental), chronological profile, and publication journals. In the period from 1976 to 2022 in Latin America, a total of 224 publications were found. The country

that leads epilepsy and calcium channel research in Brazil, with a total of 96 articles. While Mexico ranks second, with 63 documents related to the theme. The main collaborations between Latin American countries have been with the United States United and United Kingdom. The trending topics in each area are pharmacology (clinical and experimental). This bibliometric study allows us to infer that there are still some areas that require attention. in the study of epilepsy. Due to its frequency and incidence in developing countries, more research should be carried out on the mechanisms involved in its appearance.

Disclosures: **N. Gallardo López:** None. **C. rubio:** None. **V. mena:** None. **E. uribe:** None. **D. Flores:** None. **A. Lee:** None. **M. Rubio:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.22

Topic: I.07. Data Analysis and Statistics

Support: Suomen Akatemia 321235, 332017
Sigrid Juséliuksen Säätiö
Lastentautien Tutkimussäätiö
Suomen Aivosäätiö

Title: Eeg-based measures of cortical connectivity link to clinical outcomes in newborn infants

Authors: *A. TOKARIEV[1,2], M. AIRAKSINEN[1,3,2], S. VANHATALO[1,2];
[1]Dept. of Physiol., Univ. of Helsinki, Helsinki, Finland; [2]Dept. of Clin. Neurophysiology/BABA Ctr., Helsinki Univ. Hosp., Helsinki, Finland; [3]Dept. of Signal Processing and Acoustics, Aalto Univ., Espoo, Finland

Abstract: Early postnatal period of a human brain is characterized by a rapid activity-dependent development of brain connections (Molnar et al., 2020), the foundation of lifetime cognitive functions. This process is sensitive to a range of medical adversities and environmental effects. An assessment of infant's neuronal network function holds significant promise for early prediction of neurodevelopmental disorders and guidance of intervention strategies. Here, we propose an analytical framework that uses connectivity-based features computed from infant sleep EEG in a combination with machine learning to predict clinical outcomes. We collected multi-channel EEG data from N = 107 newborn infants during daytime sleep at conceptual age of 42.3 ± 0.9 weeks (mean ± std). Three-minute-long artifact-free EEG epochs of active (AS) and quiet sleep (QS) were selected for further analysis. Next, these data were band-pass filtered into four frequency bands of interest: delta (0.4-1.5 Hz), theta (4-8 Hz), alpha (8-13 Hz), beta (13-22 Hz) - and source reconstructed into 58 cortical parcels using a realistic infant head model and dynamic statistical parametric mapping (Tokariev et al., 2019). Functional connectivity by phase-phase correlations was estimated between pairs of all cortical parcels using weighted

phase lag index. We computed connectivity features as the mean connectivity levels within and between anatomical regions (frontal, central, occipital, and temporal) at each frequency. A Support Vector Regression classifier (with ridge regularization and 10-fold cross-validation) was trained to predict neurological performance of these infants, as assessed with Hammersmith Neonatal Neurological Examination. Our trained classifier provided significant prediction of infant's neurological performance with cross-validated correlation to real scores $R = 0.33$ ($p < 0.001$; Spearman test). The connectivity measures during AS showed stronger prediction ($R > 0.3$) compared to connectivity during QS ($R = 0.1$). Comparison of frequency-specific feature combinations (using mutual information criteria) showed that the most predictive power is concentrated within the delta frequency band. Our results suggest that cortical brain networks of neuronal function carry latent information that correlates with clinical performance. Combination of multiple features may have potential for clinical use as an early functional biomarker in the newborn infant brain.

Disclosures: **A. Tokariev:** None. **M. Airaksinen:** None. **S. Vanhatalo:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.23

Topic: I.07. Data Analysis and Statistics

Title: Topological Data Analysis reveals alterations in the brain functional connectome in autism spectrum disorder.

Authors: *J. DIAZ-PATINO[1], S. ALCAUTER[1], N. LÓPEZ GUERRERO[2];
[1]Inst. de Neurobiología, Univ. Autónoma de México, Queretaro, Mexico; [2]Inst. de Neurobiología, Univ. Autónoma de México, Querétaro, Mexico

Abstract: Topological Data Analysis (TDA) is a recent mathematics research area that has been used to study brain networks; previously, it was used to differentiate study groups with ADHD with good results (Gracia-Tabuenca et al., 2020, https://doi.org/10.1523/ENEURO.0543-19.2020). The approach of TDA may be used to extract different topological features as a function of the connectivity threshold, exploring all possible connectivity values instead of exploring a single threshold. Previous studies show differences of brain morphology in individuals with Autism Spectrum Disorder (ASD) (Zheng W., et al 2021, https://doi.org/10.1002/hbm.25251, Kazeminejad A. et al., 2019, https://doi.org/10.3389/fnins.2018.01018). This work focused on the Betti_n functions for dimensions n=0,1,2 to differentiate patients with ASD over typically developing (TD) subjects. We used the preprocessed data of the ABIDE I database and three different atlases: AAL, CC200, and CC400 (http://preprocessed-connectomes-project.org/). We omitted any subject with void time-series and kept all the subjects with complete full-scale IQ test data. Our final sample has 854 subjects (ages between 6 and 64 years old, 410 ASD patients, 444 TD). We computed

the Betti functions for each atlas and then computed the area under the curve (AUC) of each of these functions. The AUC of dimension 0 gives an idea of how the initially disconnected nodes connect when decreasing the connectivity threshold so that a connectome with lower AUC is associated with edges with greater connectivity that bind the nodes at higher connectivity thresholds. The AUC for dimensions 1 and 2 indicates the number of topological holes through the connectivity values. A hole occurs when there is a less dense set of connections between the nodes, i.e., a hole is a set of nodes that form a component, but they miss enough connections to form the hole. We applied an ANOVA for the AUC of all Betti functions. For the AAL, significant differences were identified for the AUC for dimension 0 in the Parietal lobe ($p=0.02$) and for dimension 1 in the Temporal lobe ($p=0.01$), where the area of the TD subjects is smaller than the area of the ASD subjects, but no differences were found when exploring the whole brain as a network. For the atlases CC200 and CC400, we found differences between groups in dimensions 1 and 2 ($p=0.014$ and $p=0.017$ for dimension 1; $p=0.047$ and $p=0.045$ for dimension 2, respectively) with the same pattern seen in dimension 0. These results suggest that TD subjects have some higher connections that bind the brain network at higher connectivity thresholds, and ASD patients have less density of connections, favoring the presence of topological holes.

Disclosures: **J. Diaz-Patino:** None. **S. Alcauter:** None. **N. López Guerrero:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.24

Topic: I.07. Data Analysis and Statistics

Support: NIA/NIH U24 AG072122
NIA/NIH UO1AG016976

Title: Aggregate Dietary Supplement Use with Cognitive Status From 2015-2019 in NACC participants Aged 55 and Older

Authors: *K. E. SANDOVAL;
Southern Illinois Univ. Edwardsville, Southern Illinois Univ. Edwardsville, Edwardsville, IL

Abstract: Research and Objective: Within the US, dietary supplements are commonly used. Supplement use and/or the types of supplements used may be influenced by cognitive status. The goal of this analysis was to investigate the association between aggregated supplement use and cognitive status (normal cognition (NC), mild cognitive impairment (MCI), dementia, or cognitive impairment not MCI (CI-nMCI)) from 2015-2019 in National Alzheimer's Coordinating Center (NACC) participants aged 55 and older. Methods: Analyses were conducted using data from the NACC. NACC participants aged 55 years and older with a visit from 1/1/2015 to 12/31/2019 with a complete medications form were included. Co-participants filled

out a medication form for the participant having used a supplement/medication in the past 14 days. The first available visit for participants meeting inclusion criteria was taken to maintain statistical independence. Supplement use was defined as reported use of any vitamin/mineral (VM) or nonvitamin/nonmineral (NVNM) supplement. The number participants using supplements was aggregated across 2015-2019. Results: When restricting the dataset to 2015-2019 with an age of 55 or greater, 19,932 participants were obtained. 19,696 (98.8%) of these participants had at least one complete medications form, resulting in a total of 19,696 participants. Of these participants, 9,357 (47.51%) were classified with normal cognition, 3,934 (19.97%) with MCI, 5,503 (27.94%) with dementia, and 902 (4.58%) with CI-nMCI. Use of any supplement was reported by 73% of participants in the analysis. A significant association was found between use of any supplement and cognitive status ($P<0.0001$, chi-square). Use of any supplement was 75.1% (95% CI: 74.2, 75.9%) in normal cognition, 74.6% (95% CI: 71.8-77.5%) in CI-nMCI, 73.1% (95% CI: 71.7-74.5%) in MCI, and 67.9% (95% CI: 66.7-69.1%) in dementia. Supplement use decreased by 7.2% (95% CI: -8.7, -5.7%) in dementia and decreased by 1.9% (95% CI: -3.6, -0.3) in MCI when compared to normal cognition. An association was found between cognitive status and use of: any vitamin ($P<0.0001$, chi-square), any vitamin including multivitamins (MVM) ($P<0.0001$), any vitamin excluding MVM ($P<0.0001$), any mineral ($P<0.0001$), and any NVNM supplement ($P<0.0001$). Significant decreases were found within each of these supplement categories for MCI and dementia when compared to normal cognition, with the percent decrease in use being greatest for dementia. Conclusion: A significant association was found between many categories of dietary supplement use and cognitive status, with use decreasing as cognitive status worsened.

Disclosures: K.E. Sandoval: None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.25

Topic: E.04. Voluntary Movements

Support: NSF-M3X-1825942
NIH-R37-HD087089
NAF-CRCNS-1723998

Title: How to average movement data? Decoupling spatial and temporal variability to extract salient features in time series

Authors: *A. KROTOV[1], R. SHARIF RAZAVIAN[2], M. SADEGHI[2], D. STERNAD[3]; [1]Bioengineering, [2]Biol., [3]Departments of Biology, Electrical & Computer Engineering, and Physics, Northeastern Univ., Boston, MA

Abstract: Motor neuroscience research examines a wide spectrum of actions, ranging from finger tapping to cracking a whip. Even the simplest of such actions show variability arising from various sources in the sensorimotor loop. To extract interpretable features from the measured spatiotemporal profiles, the multiple repetitions of movement are often summarized by resampling and binning the time series, and calculating the mean and variance in the spatial domain. The resulting mean profile aims to highlight the essential characteristics of the ensemble to provide a basis for insights into control priorities. The band of standard deviations around the mean may indicate exploration, performance refinement, and learning. However, such extraction of summary statistics is far from straightforward, as every single movement is affected by potentially different variabilities in the spatial and temporal domains that cannot easily be dissociated. Due to such coupling, simple averaging across repeated time series can lose information in both spatial and temporal dimensions of the movement. The issue has been recognized in the motor control literature but has rarely been accounted for in a systematic way. This work explored and discussed several techniques to decouple variability into spatial and temporal components and to extract salient characteristics separately for these two domains. In particular, we present a powerful analysis tool, called elastic functional analysis, which has been successfully applied in statistics, animation, and shape analysis, but only very rarely in motor neuroscience. This method uses optimization procedures to rescale the temporal evolution in a nonlinear fashion to align salient features, such as peaks, valleys, and slopes, of an ensemble of time series. This technique was compared with conventional time-normalization and time-padding, using synthesized signals with controlled levels of induced variability, as well as real hand kinematics of a three-dimensional unconstrained reaching task. Systematic analysis of these data demonstrates that the method of elastic functional analysis can be successfully applied to movement signals. It teases apart temporal and spatial variability and can substantially mitigate the potential biases in movement features when using conventional methods. Other application examples are presented and implications of this method in motor neuroscience are discussed and may inform the research when extracting subtle and otherwise covert features of movement kinematics.

Disclosures: **A. Krotov:** None. **R. Sharif Razavian:** None. **M. Sadeghi:** None. **D. Sternad:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.26

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: CONACYT CF-2019-6390

Title: Intelligence quotient (IQ) is not correlated in Mexican twins

Authors: *T. V. ROMAN-LOPEZ[1], X. J. LÓPEZ-CAMAÑO[1], V. MURILLO-LECHUGA[1], D. RAMÍREZ-GONZÁLEZ[1], R. CASA-MADRID[1], B. GARCÍA-VILCHIS[2], D. ZENTENO-MORALES[2], Z. ESPINOSA-VALDÉS[2], A. TAPIA-ATILANO[2], S. PRADEL-JIMÉNEZ[2], O. ALDANA-ASSAD[3], A. MEDINA-RIVERA[3], M. E. RENTERÍA[4], A. E. RUIZ-CONTRERAS[2], S. ALCAUTER[1];
[1]Inst. de Neurobiologia, Univ. Nacional Autonoma de Mexico, Queretaro, Mexico; [2]Lab. Neurogenomica Cognitiva, Fac. Psicologia, UNAM, Univ. Nacional Autonoma de Mexico, D.F., Mexico; [3]Lab. Internacional de Investigación sobre el Genoma Humano, Univ. Nacional Autonoma de Mexico, Queretaro, Mexico; [4]Queensland Inst. of Med. Res., Brisbane, Australia

Abstract: Twins studies are based on the premise that Monozygotic twins (MZ) share about 100% of their DNA and dizygotic (DZ), about 50%, whereas the environment remains the same for both types of twins. Then, if a trait has a higher resemblance or covariance in MZ than DZ, this can be interpreted as a higher genetic contribution. Historically, intelligence has been one of the most fascinating cognitive traits to explore in twin studies. Previous twin meta-analysis has shown that the Intelligence quotient (IQ) has a genetic contribution (or heritability) of around 80% in adulthood, another study showed a heritability of 72% in the American population; however, this is unknown in the Mexican population. This study aimed to compare the IQ of MZ and DZ twins to evaluate a potential genetic contribution in the Mexican population. IQ was evaluated using the A version of the Shipley-2 (standardized by age) scale; this scale measures both crystallized and fluid cognitive ability. For MZ and DZ comparison, three different methods were performed to estimate a potential genetic contribution: IQ values correlation between twins, concordance rates of the IQ range, and ACE analysis. A sample of 146 adult participants (18-61 years old), corresponding to 73 twin pairs (n=48 MZ and n=25 DZ) part of the Mexican Twin Registry, TwinsMX (https://twinsmxofficial.unam.mx/) answered the Shipley-2. As result, no IQ correlation was observed between MZ twins (r=0.14, p=0.35) nor DZ (r=-0.04, p=0.83). Additionally, the IQ range of interpretation (i.e., well below average, below average, average, above average, and well above average) was compared between twins to the concordance rate analysis, when both twins' score corresponds to the same range of interpretation (e.g., twin 1: average IQ, twin 2: average IQ) was classified as concordant twins, if they do not match then were classified as discordant. The probandwise concordance rate for the IQ range in MZ was 0.73 and 0.79 for DZ; the likelihood-ratio test showed no significant differences between groups (p=0.43). Finally, ACE modeling showed for genetics (A) a contribution of 13%; meanwhile, environmental (E) had an 86% contribution. These preliminary results through the three methods suggest a higher environmental contribution to IQ level; however, it is necessary to increase the sample size and to include relevant covariables (e.g., age or sex) to improve the precision of the estimations. This will allow us to understand the genetic or environmental basis of cognitive ability in the Mexican population.

Disclosures: T.V. Roman-Lopez: None. **X.J. López-Camaño:** None. **V. Murillo-Lechuga:** None. **D. Ramírez-González:** None. **R. Casa-Madrid:** None. **B. García-Vilchis:** None. **D. Zenteno-Morales:** None. **Z. Espinosa-Valdés:** None. **A. Tapia-Atilano:** None. **S. Pradel-Jiménez:** None. **O. Aldana-Assad:** None. **A. Medina-Rivera:** None. **M.E. Rentería:** None. **A.E. Ruiz-Contreras:** None. **S. Alcauter:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.27

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: CONACYT CF-2019-6390

Title: Mental Health Traits in Mexican Twins

Authors: *I. M. ESPINOSA-MÉNDEZ[1,2], A. PIÑA-HERNÁNDEZ[1,2], T. V. ROMAN-LOPEZ[1], A. B. GARCÍA-VILCHIS[3], X. J. LÓPEZ-CAMAÑO[1], V. MURILLO-LECHUGA[1], D. RAMÍREZ-GONZÁLEZ[1], R. CASA-MADRID[1], D. ZENTENO-MORALES[3], Z. X. ESPINOSA-VALDÉS[3], A. TAPIA-ATILANO[3], S. A. PRADEL-JIMÉNEZ[3], E. CHIU-HAN[1], O. ALDANA-ASSAD[4], A. MEDINA-RIVERA[4], M. E. RENTERIA[5], A. E. RUIZ-CONTRERAS[3], S. ALCAUTER[1];
[1]Inst. de Neurobiología, Univ. Nacional Autónoma de México, Querétaro, Mexico; [2]Escuela Nacional de Estudios Superiores Juriquilla, Univ. Nacional Autónoma de México, Querétaro, Mexico; [3]Lab. de Neurogenómica Cognitiva, Coordinación de Psicobiología y Neurociencias Facultad de Psicología, Univ. Nacional Autónoma de México, Ciudad de México, Mexico; [4]Lab. Internacional de Investigación sobre el Genoma Humano, Univ. Nacional Autónoma de México, Querétaro, Mexico; [5]QIMR Berghofer Med. Res. Inst., Brisbane, Australia

Abstract: Twins studies allow the assessment of the weight of genetic and environmental contribution to different traits and diseases, being extremely helpful in physical and mental health research. The variability of a phenotype is compared between Monozygotic (MZ) or identical twins who share about 100% of their DNA, and dizygotic (DZ) or fraternal twins who share about 50% of their DNA. Furthermore, MZ and DZ twins share their environment for a large portion of their lives. If a trait is more frequent or similar among MZ twins, it is likely to have a higher genetic load. Twin studies in the Latin American population have been very limited, and it is of great need to obtain results that can be translated to the genetically admixed Mexican population. This study aims to compare the score of the Symptom Checklist-90-Revised (SCL-90-R) used to assess mental health traits in nine symptomatic dimensions in Mexican MZ and DZ twins. A sample of 174 pairs of twins (n=108 MZ and n=66 DZ) answered the SCL-90-R through the official website of the Mexican Twin Registry, TwinsMX (https://twinsmxofficial.unam.mx). A significant correlation for MZ, but not for DZ twins was found in the traits of depression (MZ rs= 0.468, $p<0.01$; DZ p= 0.086), hostility (MZ rs= 0.319, $p<0.01$; DZ p= 0.132), phobic anxiety (MZ rs= 0.323, $p<0.01$; DZ p= 0.426), and somatization (MZ rs= 0.306, $p<0.01$; DZ p= 0.202) suggesting a greater genetic contribution to these traits. Whereas significant correlations for both MZ and DZ were found in the traits of obsessive-compulsive disorder (MZ rs= 0.420, $p<0.01$; DZ rs=0.279 $p<0.05$), interpersonal sensitivity (MZ rs= 0.466, $p<0.01$; DZ rs=0.288 $p<0.05$), anxiety (MZ rs= 0.413, $p<0.01$; DZ rs=0.255 $p<0.05$), paranoid ideation (MZ rs= 0.450, $p<0.01$; DZ rs=0.380 $p<0.01$), and psychoticism (MZ rs= 0.409, $p<0.01$; DZ rs=0.268 $p<0.05$) which suggest that these are probably mediated by environmental influences. These results are the first step to evaluate the genetic and

environmental contributions in mental health traits; however, further analyses are required to estimate their heritability of these traits in the Mexican population.

Disclosures: **I.M. Espinosa-Méndez:** None. **A. Piña-Hernández:** None. **T.V. Roman-Lopez:** None. **A.B. García-Vilchis:** None. **X.J. López-Camaño:** None. **V. Murillo-Lechuga:** None. **D. Ramírez-González:** None. **R. Casa-Madrid:** None. **D. Zenteno-Morales:** None. **Z.X. Espinosa-Valdés:** None. **A. Tapia-Atilano:** None. **S.A. Pradel-Jiménez:** None. **E. Chiu-Han:** None. **O. Aldana-Assad:** None. **A. Medina-Rivera:** None. **M.E. Renteria:** None. **A.E. Ruiz-Contreras:** None. **S. Alcauter:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.28

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: CONACYT CF-2019-6390

Title: Sex and Zygosity Differences in State-Trait Anxiety in Mexican Twins

Authors: *Z. X. ESPINOSA-VALDES[1], T. V. ROMÁN-LÓPEZ[3], D. ZENTENO[1], S. A. PRADEL-JIMÉNEZ[1], A. TAPIA-ATILANO[1], A. GARCÍA-VILCHIS[1], V. MURILLO-LECHUGA[3], A. V. ZANABRIA-PÉREZ[1], E. CHIU-HAN[1], D. RAMÍREZ-GONZÁLEZ[3], O. ALDANA-ASSAD[2], A. MEDINA-RIVERA[2], M. E. RENTERÍA[4], A. E. RUIZ-CONTRERAS[1], S. ALCAUTER[3];
[1]Facultad de Psicología, Univ. Nacional Autónoma de México, Ciudad de México, Mexico; [2]Lab. Internacional de Investigación sobre el Genoma Humano, Univ. Nacional Autónoma de México, Querétaro, Mexico; [3]Inst. de Neurobiología, UNAM, Queretaro, Mexico; [4]Queensland Inst. of Med. Res., Herston, Australia

Abstract: There are two concepts that most people are unaware of and that can help us to understand how a person experiences anxiety. These are “state anxiety” and “trait anxiety”. The first one is the response to certain situations, at a certain time, and symptoms associated with anxiety can be identified; while trait anxiety is the way of responding frequently to life situations and helps to identify tendencies when acting anxiously. The 'heritability' (i.e., the proportion of a trait in a population that can be attributed to genetic factors) of anxiety in Latinos in the United States has been found to be approximately 7.2%, while in populations of European ancestry, genetic factors could be contributing to around 31% of the variance in anxiety symptoms. The purpose of this study was to correlate symptomatology of anxiety in Monozygotic (MZ) and Dizygotic (DZ) twins separated by sex. A sample of 195 twin pairs (n=120 MZ pairs in which n=100 women and n=20 men; and n=75 DZ pairs in which n=57 women and n=18 men) from the Mexican Twin Registry, TwinsMX, answered the State-Trait Anxiety Inventory (STAI). lt was observed a significant correlation for Anxiety-Trait in MZ twins (r_s=0.49; p<6.21e-09), and

in DZ (r_s=0.35;p<0.0018). On the other hand, for Anxiety-State there were significant results for MZ (r_s=0.37;p<3.002e-05) but not in DZ (r_s=0.20;p=0.71). Meanwhile, there was a significant correlation for both MZ females (r_s= 0.51; p<3.791e-08) and for DZ females (r_s=0.43;p<6.319e-04); however, there was no significant correlation for MZ males (r_s= 0.42; p=0.06) neither DZ males (r_s= 0.15; p=0.53). On the other hand, for Anxiety-State there were significant results for MZ females (r_s=0.40;p<2.709e-05) but not in DZ females (r_s=0.35;p=0.059). In addition, there were no significant results in both MZ males (r_s=0.10;p=0.64) and DZ males (r_s=0.15;p=0.54). Anxiety-State significant results in MZ twins, but not in DZ, suggest that it is attributed to genetic factors. Furthermore, the results segmented by sex, show a greater difference between MZ females and DZ females in Anxiety-State rather than Anxiety-Trait. Nethertheless, results of Anxiety-State and Anxiety-Trait in males, for both MZ and DZ are still preliminar because of the small number of men, therefore a larger sample is necessary to refine the statistical analysis. On the other hand, these studies will be of great value for the advancement in the field of mental health in the Mexican population. We expect that the recently created TwinsMX, in the future, may contribute to develop strategies for prevention, diagnosis and more suitable treatments.

Disclosures: Z.X. Espinosa-Valdes: None. **T.V. Román-López:** None. **D. Zenteno:** None. **S.A. Pradel-Jiménez:** None. **A. Tapia-Atilano:** None. **A. García-Vilchis:** None. **V. Murillo-Lechuga:** None. **A.V. Zanabria-Pérez:** None. **E. Chiu-Han:** None. **D. Ramírez-González:** None. **O. Aldana-Assad:** None. **A. Medina-Rivera:** None. **M.E. Rentería:** None. **A.E. Ruiz-Contreras:** None. **S. Alcauter:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.29

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: CONACYT CF, 2019. No.6390

Title: Quality sleep in Mexican Twins

Authors: *D. ZENTENO-MORALES[1], T. V. ROMÁN-LÓPEZ[2], Z. ESPINOSA-VALDES[1], S. PRADEL-JIMÉNEZ[1], A. TAPIA-ATILANO[1], I. E. ORTEGA-MORA[1], U. CABALLERO-SÁNCHEZ[1], A. B. GARCÍA-VILCHIS[1], V. MURILLO-LECHUGA[2], D. RAMÍREZ-GONZÁLEZ[2], E. CHUI-HAN[2], O. ALDANA-ASSAD[3], M. E. RENTERÍA[4], A. MEDINA-RIVERA[3], S. ALCAUTER[2], A. E. RUIZ-CONTRERAS[1];
[1]Lab. de Neurogenómica Cognitiva, Facultad de Psicología, Univ. Nacional Autónoma de México, Coyoacán, Mexico; [2]Inst. de Neurobiología, [3]Lab. Internacional de Investigación sobre el Genoma Humano, Univ. Nacional Autónoma de México, Querétaro, Mexico; [4]QIMR Berghofer Med. Res. Inst., Brisbane, QLD, Australia

Abstract: The twin studies allow us to detect the relative genetic and environmental contribution to a trait; the principal assumption is that the genetic resemblance in monozygotic (MZ) twins is close to 100%, whereas in the dizygotic (DZ) twins is ~50%, and both of them share the same environment. Regarding this fact, when MZ twins have more resemblance in a trait, rather than DZ twins, it suggests that there is a higher genetic contribution for that trait. Sleep quality is a very broad concept that includes several measures, such as duration, latency, efficiency, sleep disturbances, use of drugs for sleep, and how well people perform during the day. Sleep quality has been measured in several studies using the Pittsburgh Sleep Quality Index (PSQI). The genetic contribution of sleep quality has been studied in various populations. For example, in the Spanish adolescent population, a genetic contribution of 42% has been estimated, while in the American, English, and Finnish populations a contribution of 34%, 43% and 44%, respectively, has been found. This suggests that environmental and genetic contributions to sleep quality may differ between populations. The aim of this study was to assess the PSQI total scores in Mexican monozygotic (MZ) and dizygotic (DZ) twins. A sample of 143 pairs (n=93 MZ pairs and n=50 DZ pairs) answered through the website of the Mexican Twin Registry, TwinsMX (https://twinsmxofficial.unam.mx/) the PSQI, which analyzes the quality of sleep in the last month. This instrument considers different categories to measure sleep quality (included: habitual sleep efficiency, sleep disturbances, subjective sleep quality, among others). As a result of this study, a significant correlation was observed for the PSQI total score in MZ twins (r_s=0.41; p=0.003), but not in DZ (p=0.41). The PSQI total score results suggest that sleep quality is probably associated with a greater genetic contribution. However, more data collection is needed to perform ACE analyses to calculate more accurately the heritability of sleep quality in our population. Overall, this study shows the need for twin data in the Mexican population to strengthen the knowledge of mental health traits such as sleep quality.

Disclosures: D. Zenteno-Morales: None. **T.V. Román-López:** None. **Z. Espinosa-Valdes:** None. **S. Pradel-Jiménez:** None. **A. Tapia-Atilano:** None. **I.E. Ortega-Mora:** None. **U. Caballero-Sánchez:** None. **A.B. García-Vilchis:** None. **V. Murillo-Lechuga:** None. **D. Ramírez-González:** None. **E. Chui-Han:** None. **O. Aldana-Assad:** None. **M.E. Rentería:** None. **A. Medina-Rivera:** None. **S. Alcauter:** None. **A.E. Ruiz-Contreras:** None.

Poster

580. Imaging and Analysis of Specific Human Populations and Traits

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 580.30

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: CONACYT CF, 2019. No.6390

Title: Personality traits in a cohort of Mexican twins

Authors: B. GARCÍA-VILCHIS[1], T. V. ROMAN-LOPEZ[3], E. ORTEGA-MORA[2], U. CABALLERO-SÁNCHEZ[2], E. CHIU-HAN[4], D. RAMÍREZ-GONZÁLEZ[2], O. ALDANA-

ASSAD[5], A. TAPIA-ATILANO[2], S. PRADEL-JIMÉNEZ[2], D. ZENTENO-MORALES[2], Z. X. ESPINOSA-VALDES[2], X. J. LÓPEZ-CAMAÑO[4], M. RENTERÍA[6], A. MEDINA-RIVERA[6], S. ALCAUTER[4], A. E. RUIZ-CONTRERAS[2];
[1]Univ. Nacional Autónoma de México, Coyoacan, Mexico; [2]Univ. Nacional Autónoma de México, Ciudad de México, Mexico; [3]Inst. de Neurobiologia, UNAM, Queretaro, Mexico; [4]Inst. de Neurobiología, Univ. Nacional Autónoma de México, Queretaro, Mexico; [5]Lab. Internacional de Investigación sobre el Genoma Humano, Univ. Nacional Autónoma de México, Queretaro, Moldova, Republic of; [6]Lab. Internacional de Investigación sobre el Genoma Humano, Univ. Nacional Autónoma de México, Queretaro, Mexico

Abstract: The Big Five Instrument is widely used in psychology to assess people's personality. Through a self-assessment, this instrument can report the degree of extraversion, agreeableness, conscientiousness, neuroticism, and openness of a person. The present work aims to evaluate the hereditary and environmental components of each of the traits evaluated with the Big Five Instrument in the Mexican population, which presents a genetic admixture, different from that observed in other regions of the world. To achieve this objective, we developed a classic study of twins with the data collected by the Mexican Twin Registry, which has around 4100 registries (May 2022). In a twin study, it is possible to compare the similarity of a trait in monozygotic twins (who have a resemblance of 100% of their DNA and about 100% of environmental factors) and dizygotic twins (who have a resemblance of about 50% of their DNA and about 100% of environmental factors). With the help of structural equation models and the ACE model, the relative contribution of genetic (heritability; a2) and environmental factors to a specific trait can be calculated. Of the total records, 270 pairs of twins have completed the Big Five Instrument, of which 173 are monozygotic and 78 are dizygotic. Participated a higher proportion of female (71%) and young people between 20-30 years old (40%). According to the data we currently have, the following explained variance percentages by factors have been obtained (V: explained variance by A: additive genetic; C: common environment; E: unique environment): Extraversion: a2=VA=48.9%, VC=0%, VE=51.1%; Agreeableness: a2=VA=26.0%, VC=7.6%, VE=66.4%; Awareness: a2=VA=40.9%, VC=0%, VE=59.1%; Neuroticism: a2=VA=39.3%, VC=0%, VE=60.7%; Openness: a2=VA=45.7%, VC=0%, VE=54.3%.As preliminary conclusions, in the Mexican population, extraversion is the trait with the highest genetic variance, while agreeableness is the trait with the lowest genetic variance. This last trait is the only one to date that presents a significant common environment component.This preliminary study is one of several others from the Mexican Twin Registry, which is a recently started effort and is expected to be an invaluable resource for the characterization of different traits in the Mexican population.

Disclosures: B. García-Vilchis: None. **T.V. Roman-Lopez:** None. **E. Ortega-Mora:** None. **U. Caballero-Sánchez:** None. **E. Chiu-Han:** None. **D. Ramírez-González:** None. **O. Aldana-Assad:** None. **A. Tapia-Atilano:** None. **S. Pradel-Jiménez:** None. **D. Zenteno-Morales:** None. **Z.X. Espinosa-Valdes:** None. **X.J. López-Camaño:** None. **M. Rentería:** None. **A. Medina-Rivera:** None. **S. Alcauter:** None. **A.E. Ruiz-Contreras:** None.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.01

Topic: I.08. Methods to Modulate Neural Activity

Support: Defense Advanced Research Projects Agency (DARPA) Contract No. N65236-19-C-8013
Naval Information Warfare Center

Title: Behavioral and Neural Changes induced by Focused Ultrasonic Stimulation in Primate Frontal Eye Fields

Authors: *M. R. RILEY[1], B. M. ROEDER[1], J. P. CAFARO[2], M. P. WEISEND[2], G. F. PINTON[3,4], A. O. BILIROGULU[3], F. Y. YAMANER[3], O. ORALKAN[3], R. E. HAMPSON[1], P. M. CONNOLLY[2];
[1]Wake Forest Univ. Sch. of Med., Winston Salem, NC; [2]Teledyne Scientific Co., Research Triangle Park, NC; [3]NC State Univ., Raleigh, NC; [4]Univ. of North Carolina at Chapel Hill, Chapel Hill, NC

Abstract: Activation of neural tissue while documenting behavioral change is the gold standard for studying brain function. For decades electrical currents, and more recently, optogenetics produce similar tissue activation and behavioral results when applied to the same brain region. However, both techniques require invasive surgeries to implant stimulating electrodes, virus vectors and/or optic sources. State of the art noninvasive stimulation techniques such as transcranial magnetic stimulation (TMS) have poor spatial resolution. In contrast, low intensity transcranial focused ultrasound (TFUS) is noninvasive and capable of high spatial resolution (~1mm^3). We report here, the use of TFUS to elicit changes in electrophysiology and behavior at unprecedented spatial resolution.
As part of the DARPA Next-generation Noninvasive Neurotechnology program, we delivered TFUS to the right frontal eye field (FEF) region of a macaque performing a task requiring fixation followed by saccades to targets. In brief, we delivered 2.8 MHz pulsed TFUS to the FEF for 200 ms from a phased array of 4096 transducer elements. TFUS was targeted with phase delays calculated to correct for inhomogeneities between transducer and target location. Multiple locations in the FEF were targeted without physically moving the device by steering the beam using unique phase delays. Local field potentials were recorded with multi-channel electrodes in FEF.
TFUS elicited theta and alpha (5-12 Hz) activity during the stimulation epoch that was not seen during non-stimulated “sham” trials. Changes in FEF activation altered saccade behavior and disrupted behavioral performance. TFUS caused saccades to 5 locations distinct from the visual target positions, even effecting sham trial performance. In addition, the TFUS targets in FEF appeared to be volumetrically confined enough to elicit different & specific effects. Targeted stimulation at one point caused generalized disruption of performance while a target 1 mm away induced saccades to a very specific, non-target location. Our results demonstrate that TFUS affects small, specific regions of brain tissue and that steering the beam to different locations in FEF produces differentiable behavioral outcomes.
This work was supported by the Naval Information Warfare Center (NIWC) and the Defense Advanced Research Projects Agency (DARPA) under Contract No. N65236-19-C-8013. The

views, opinions, and/or findings contained in this abstract are those of the author and should not be interpreted as representing the official views or policies, either expressed or implied, of the NIWC Atlantic, DARPA, or the Department of Defense.

Disclosures: M.R. Riley: None. **B.M. Roeder:** None. **J.P. Cafaro:** None. **M.P. Weisend:** None. **G.F. Pinton:** None. **A.O. Bilirogulu:** None. **F.Y. Yamaner:** None. **O. Oralkan:** None. **R.E. Hampson:** None. **P.M. Connolly:** None.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.02

Topic: I.08. Methods to Modulate Neural Activity

Support: Defense Advanced Research Projects Agency (DARPA) Contract No. N65236-19-C-8013
Naval Information Warfare Center

Title: Effects of Focused Ultrasound Delivery Angles on Local Field Potentials and Saccadic Behavior

Authors: *R. E. HAMPSON[1], M. R. RILEY[1], B. M. ROEDER[1], J. P. CAFARO[2], M. P. WEISEND[2], G. F. PINTON[3,4], A. O. BILIROGLU[3], F. Y. YAMANER[3], O. ORALKAN[3], P. M. CONNOLLY[2];
[1]Wake Forest Univ. Sch. of Med., Winston Salem, NC; [2]Teledyne Scientific Co., Research Triangle Park, NC; [3]NC State Univ., Raleigh, NC; [4]Univ. of North Carolina at Chapel Hill, Chapel Hill, NC

Abstract: Low-intensity transcranial focused ultrasound (TFUS) is a promising method for non-invasive stimulation of brain tissue. TFUS has been used for neural stimulation in rodent, non-human primate, and human subjects. However, the parameters for optimized TFUS effects remain a topic of investigation.
As part of the DARPA Next-generation Noninvasive Neurotechnology program, we delivered TFUS to the right frontal eye field (FEF) region of a macaque performing a task requiring central fixation followed by saccades to visual targets. In brief, we delivered 2.8 MHz pulsed TFUS to the FEF for 200 ms from a phased array. TFUS was targeted with phase delays calculated to correct for inhomogeneities between transducer and target. The TFUS transducer arrays were placed in two locations. The “tilted” transducer position targeted FEF through a window of thin compact bone and positioned the transducer plane tangential to the dorsal lateral skull surface. The “tilted” position resulted in two transducer array segments close to scalp and two more distant requiring connection to scalp via thick ultrasound gel. The second, “flat”, location targeted FEF by placing the transducer array where all array segments were tangent to and approximately equidistant from the scalp. Local field potentials were recorded with multi-

channel electrodes in FEF.
We observed that the “flat” position was superior to the “tilted” position in producing temporally and spectrally specific neural responses and behavioral outcomes. In the “flat” position the activation of neural tissue was temporally focused during periods of stimulation and specific to the theta and alpha bands (5-12 Hz). Conversely, the “tilted” position produced broadband differences across frequencies from 2-40 Hz that lasted more than 500 ms. In concert with the neural responses TFUS in the “tilted” position elicited small variable changes in saccade performance after fixation. While TFUS in the “flat” position induced larger saccade amplitudes as well as saccades to multiple, replicable, non-target saccade locations. In conclusion, the position of the transducer relative to scalp and skull tissues significantly influenced the outcome of TFUS in this study and should be carefully considered when developing paradigms.
This work was supported by the Naval Information Warfare Center (NIWC) and the Defense Advanced Research Projects Agency (DARPA) under Contract No. N65236-19-C-8013. The views, opinions, and/or findings contained in this abstract are those of the author and should not be interpreted as representing the official views or policies, either expressed or implied, of the NIWC Atlantic, DARPA, or the Department of Defense.

Disclosures: **R.E. Hampson:** None. **M.R. Riley:** None. **B.M. Roeder:** None. **J.P. Cafaro:** None. **M.P. Weisend:** None. **G.F. Pinton:** None. **A.O. Biliroglu:** None. **F.Y. Yamaner:** None. **O. Oralkan:** None. **P.M. Connolly:** None.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.03

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant R01 EB028319-01A1

Title: Personalized editing of sensory representation using scanning low intensity focused ultrasound: a translational model.

Authors: *J. HEHIR, Y. SHEN, J. A. FISHER;
Physiol., New York Med. Col., Valhalla, NY

Abstract: Low intensity focused ultrasound (FUS) is an emerging technology for noninvasively modulating brain activity with millimeter-scale spatial resolution. Studies in humans and nonhuman primates have provided evidence that the technology’s high spatial resolution can be leveraged to induce measurable changes in fine motor and sensory function, for example in digit-specific cortical modulation. Small animal models present an attractive, higher-throughput alternative for optimizing stimulus and targeting parameters partly because there is a wide array of genetically-driven toolboxes for imaging that are available. However, the majority of such preclinical research has heretofore focused on sensory modalities that are behaviorally of limited

interest in humans, e.g. gross stimulation of forelimb/hindlimb. Here, we use a steerable, phased-array FUS transducer to target regions of primary auditory cortex (A1) in mice. The tonotopically-organized auditory cortex provides a convenient testbed for assessing high-resolution editing of a cortical sensory target that requires millimeter precision in humans as well. In transgenic mice expressing the Ca^{2+}-sensitive reporter GCaMP6s under the Thy1 promoter, we performed simultaneous *in vivo* wide-field Ca^{2+} imaging in A1 and targeted FUS through chronically implanted cranial windows. Rather than directly observing FUS-evoked activity, we measured resulting modulation of tone-evoked responses, thus avoiding direct auditory sensory artifacts from ultrasound stimulation that have been well documented. We observed spatially complex, tonotopic maps of cortical activity in response to acoustic frequencies ranging from 5kHz - 20 kHz and explored the impact of targeting FUS at individual frequency centroids. We observed post-sonication changes in cortical responses including altered spatial profile. While in most cases the effects appeared to resolve on a timescale of minutes, other changes such as alterations in response intensity, persisted throughout the remainder of imaging sessions (~30 min). We additionally performed behavioral tests and imaged blood flow in the cortical microvasculature using optical coherence tomography angiography to ensure that FUS induced no gross changes. Beyond immediate applications to animal models of auditory dysfunction such as tinnitus, this platform can more broadly be used to elucidate the mechanisms of FUS neuromodulation *in vivo*, as well as to ultimately inform parameters and limitations for personalized therapy for a multitude of other conditions that require high-resolution intervention.

Disclosures: **J. Hehir:** None. **Y. Shen:** None. **J.A. Fisher:** None.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.04

Topic: I.08. Methods to Modulate Neural Activity

Title: Sonogenetic Therapy For Visual Restoration

Authors: *I. ALCALA[1], S. CADONI[1], C. DEMENÉ[2], J.-A. SAHEL[1], M. TANTER[2], S. PICAUD[1];
[1]Inst. de la Vision, Paris, France; [2]Physics for Med., Paris, France

Abstract: In a recent study, we showed that sonogenetic therapy can be applied to stimulate the visual cortex at a high spatiotemporal resolution with ultrasound following the expression of the MscL mechanosensitive ionic channel in cortical neurons[1].
Here we investigated if the sonogenetic activation of the visual cortex can generate visual perception using an associative learning test.
Methods: The mechanosensitive ion channel (MscL) was expressed by AAV injection in the mouse visual cortex. Mice under water restriction and head-fixed were first taught to associate water delivery to a flash of light. After for days fo the associative learning task , ultrasounds

were then applied using an ultrasound transducer on a cranial window above the visual cortex in head-fixed mice.
Results: In the associative learning task, all the animals increased their licking behavior in the time window between the flash and the water delivery from a mean of 30.9 ± 17.9% the first day up to 86.2 ± 14.1% the fourth day for injected mice and 30.5 ± 28.2% up to 91.7 ± 10.3% for non-injected mice. After this association training, the flash stimulus was replaced by an ultrasound stimulation. For only the injected mice expressing MscL (n=14), the efficacy of the ultrasound stimulations induced response (69.3 ± 25.4%) was not statistically different to the efficacy of their visually induced response at the fourth day of associative learning (86.2 ± 14.1%). On the other hand, it was not the same case for the non-injected mice (n=9) which efficacy of the ultrasound stimulations induced response (38.1 ± 18.5%) was far from the one that was induced by the flash (91.7 ± 10.3%). The US-elicited response was highly dependent upon the ultrasound intensities in AAV-injected animals.
Conclusion: We here demonstrated that mice expressing the MscL ionic channel in their cortical neurons perceived an ultrasonic stimulation of the visual cortex as a natural visual stimulation. These results are encouraging for the application of sonogenetics as a novel strategy to restore vision at a cortical level in blind patients with optic nerve atrophy.
1)S. Cadoni et al. (In Press). “Ectopic expression of a mechanosensitive channel confers spatiotemporal resolution to ultrasound stimulations of neuronal circuits”.

Disclosures: I. Alcala: None. **S. Cadoni:** None. **C. Demené:** None. **J. Sahel:** None. **M. Tanter:** None. **S. Picaud:** None.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.05

Topic: I.08. Methods to Modulate Neural Activity

Support: 2018YFA0701400
2021YFF0702200
LR21C090001
LY20C090006

Title: Exploration of improving social deficit in ASD by FUS-aided OT administration

Authors: T. HE[1,2], X. LI[3], Y. LUO[3], Y. GU[4], H.-Y. LAI[1,2], L. WANG[3];
[1]Col. of Biomed. Engin. and Instrument Science, Zhejiang Univ., Hangzhou, China; [2]Dept. of Neurol. of the Second Affiliated Hospital, Interdisciplinary Inst. of Neurosci. and Technology, Zhejiang Univ. Sch. of Med., Hangzhou, China; [3]Dept. of Neurol. of the First Affiliated Hospital, Interdisciplinary Inst. of Neurosci. and Technology, Zhejiang Univ. Sch. of Med., Hangzhou, China; [4]Zhejiang Univ., Ctr. of Stem Cell and Regenerative Medicine, and Dept. of Neurol. of the Second Affiliated Hospital, Zhejiang Univ. Sch. of Med., Hangzhou, China

Abstract: Autism spectrum disorder (ASD) refers to a group of neurodevelopmental disorders characterized by deficits in social communication, restrictive interests and repetitive behaviors. To date, there is no effective treatment for ASD yet. Recently, emerging evidence suggests oxytocin (OT), a nine amino-acid peptide, might be used for ASD treatment in clinic. However, due to its molecular weight, OT cannot efficiently pass the blood brain barrier (BBB), which may significantly limit its effect. Focused ultrasound (FUS) combined with microbubbles is a newly developed technique which can temporarily disrupt the BBB, showing the great potential to precisely deliver drugs into local brain regions. However, whether this novel technique can be applied for ASD treatment has not been explored. In the present study, using valproic acid (VPA)-exposed rat as ASD animal model, we explored the effect on social novelty deficits in ASD by selectively delivering OT into the medial prefrontal cortex (mPFC) by FUS-induced BBB opening. We first verified that 0.55-mechanical index FUS could open the BBB of 5-week rats without cellular and circuit damage. We then found precise OT delivery in the mPFC by FUS partially recovered social novelty deficit in VPA-exposed rats. Moreover, compared with the effect of intravenous injection of OT, FUS did improve the effect of OT at low concentration on treating social novelty deficits in ASD. Together, our results suggest that FUS-aided OT administration in the mPFC could be a valuable strategy to be applied in the treatment of ASD in clinic.

Disclosures: **T. He:** None. **X. Li:** None. **Y. Luo:** None. **Y. Gu:** None. **H. Lai:** None. **L. Wang:** None.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.06

Topic: I.08. Methods to Modulate Neural Activity

Support: NSERC RGPIN-2020-04176

Title: Investigating the effects of cerebellar transcranial ultrasound stimulation on motor cortex excitability

Authors: *J.-F. NANKOO[1], N. RAIES[2], A. FOMENKO[3], J. BAARBÉ[4], X. XIA[1], Y. WANG[1], Y. SHAMLI OGHLI[2], C. SARICA[1], G. DARMANI[5], A. M. LOZANO[6], R. CHEN[1];
[2]Neurol., [3]Neurosurg., [1]Univ. Hlth. Network, Toronto, ON, Canada; [4]Univ. Hlth. Network, Toronto, AB, Canada; [5]Krembil Res. Inst., [6]Dept Neurosurg., Univ. Hlth. Network, Toronto, ON, Canada

Abstract: Transcranial low intensity focused ultrasound (TUS) is a novel non-invasive brain stimulation (NIBS) technique that uses acoustic waves to alter neuronal excitability. TUS has a greater spatial focality and can reach deeper brain structures compared to other NIBS techniques such as transcranial magnetic stimulation (TMS). This makes TUS particularly well-suited to

target functional subregions of the cerebellum. We investigated the effects of cerebellar TUS (cbTUS) on motor cortex (M1) excitability. We hypothesized that 1) longer cbTUS duration will induce greater inhibition of M1 excitability; 2) TUS will transiently lower M1 excitability post-sonication; 3) cbTUS of cerebellar lobule VIII will have greater inhibition and longer lasting effects compared to scalp-based coordinates (i.e., Crus I/II). Real cbTUS stimulation consisted of sonication at 20 W whereas sham consisted of 0 W. A 12 kHz tone was used to mask the mechanical sound of the transducer. In Experiment 1 (n = 16), cbTUS was applied to Crus I/II and motor evoked potentials (MEPs) from TMS of contralateral M1 were measured at pre-TUS (i.e., baseline), after 150ms, 300ms, and 450ms of TUS, and 150ms following 500 ms of TUS. We observed a trend that longer sonication duration induced greater inhibition, with peak inhibition observed following 450 ms of cbTUS and 150 ms post-sonication. Comparison of 450 ms of real and sham cbTUS showed that MEPs were significantly lower after real stimulation. In Experiment 2 (n = 15) we assessed longer latencies post-sonication and compared Crus I/II stimulation to lobule VIII stimulation. We again found a significant inhibition following 450 ms of cbTUS and at 150 ms post 500 ms sonication. However, MEPs 150 ms post-real TUS did not differ from sham. No effect of stimulation location was found. In Experiment 3 (n = 7), we used similar timepoints as Experiment 2 but tested primarily 0W sham and included two active sham conditions: 450ms of 20 W TUS targeting V1, and 450 ms of 20 W stimulation with the active face of the transducer facing away from the scalp. A real stimulation condition consisting of 450 ms cbTUS was also included. Preliminary results show that inhibition occurred at multiple times points post-sonication and following 450ms of sham stimulation, likely due to an inhibitory effect of the sound mask. MEPs immediately following 450ms of real cbTUS was lower than 450 ms of sham stimulation. The results provide evidence of an inhibitory effect of cbTUS in addition to the inhibitory effect of the sound mask. Our study shows that cbTUS can effectively modulate motor cortical excitability and is the first demonstration of an effect of cerebellar TUS in humans.

Disclosures: **J. Nankoo:** None. **N. Raies:** None. **A. Fomenko:** None. **J. Baarbé:** None. **X. Xia:** None. **Y. Wang:** None. **Y. Shamli Oghli:** None. **C. Sarica:** None. **G. Darmani:** None. **A.M. Lozano:** None. **R. Chen:** None.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.07

Topic: I.08. Methods to Modulate Neural Activity

Support: Swiss National Science Foundation

Title: Ex vivo characterization of ultrasound neuromodulation in peripheral fibers using a microfabricated nerve-on-a-chip-platform

Authors: *E. VICARI[1], T. LEMAIRE[2,1], V. PAGGI[1], E. NEUFELD[3], S. LACOUR[1], S. MICERA[1,4];
[1]École polytechnique fédérale de Lausanne, Lausanne, Switzerland; [2]New York Univ. Sch. of Med., New York, NY; [3]Computat. Life Sci., IT'IS Fndn., Zurich, Switzerland; [4]Scuola Superiore Sant'Anna, Pisa, Italy

Abstract: Focused ultrasound (FUS) is emerging as a promising non-invasive neuromodulation modality. Although it is widely employed to modulate brain circuits, its application to peripheral nerves is still limited and debated. In this study we aim to (1) characterize FUS-evoked responses in isolated peripheral neural fascicles, and (2) elucidate the FUS recruitment properties of different fiber types by analyzing the propagation properties of the evoked responses, in order to confirm the potential of FUS for peripheral neuromodulation and guide the design of effective sonication protocols. We used an ex vivo experimental setup to apply electrical and ultrasonic exposures to neural fascicles and record the evoked neural activity. Nerve fascicles were isolated from explanted rat spinal roots and inserted in a microfabricated nerve-on-a-chip platform (Gribi et al., 2018), consisting of two aligned microchannels electrodes: one for electrical stimulation and one for recording (the last one with multiple active sites placed along the channel length). Acoustic stimulation was delivered through a FUS transducer (500 kHz central frequency) placed above the fascicle. The stimulation protocol consisted of single FUS pulses varying in peak pressure amplitude and duration. Electrical stimulation was applied to assess the viability of the fascicle. We recorded propagating compound action potentials (CAPs) generated by both electrical and ultrasonic stimuli along the fascicle. We checked that the applied US stimulations did not damage the neural fibers. Our data shows that FUS evokes neural responses with very high success rates (above 80%) with an optimized combination of acoustic parameters. These responses have shapes and amplitudes similar to the ones obtained with electrical stimulation. Furthermore, clear strength-duration excitability patterns emerged when exploring the 2D stimulation parameters space. Thanks to a velocity selective recording algorithm, we differentiated the responses from different fiber types and evaluated their distinct FUS recruitment properties. This ex vivo characterization of FUS-evoked responses in isolated neural fascicles confirms that FUS stimulation can directly excite peripheral fibers and reveals effective stimulation parameters. Comparing these experimental results with modeling predictions could give insights on the mechanism of interaction between FUS and the neural membrane. Moreover, leveraging distinct FUS recruitment properties of different fiber types could open new possibilities for selective peripheral neural stimulation strategies.

Disclosures: E. Vicari: None. **T. Lemaire:** None. **V. Paggi:** None. **E. Neufeld:** None. **S. Lacour:** None. **S. Micera:** None.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.08

Topic: I.08. Methods to Modulate Neural Activity

Title: Mfsd2a mediated mechanism of BBB opening using low intensity focused ultrasound

Authors: *K. CHANG[1], Y. SEO[1], J. LEE[2], C. KONG[1], J. SHIN[1], J. CHANG[1], Y. NA[3], W. CHANG[1];
[1]Yonsei Univ., Seoul, Korea, Republic of; [2]Ewha Womans Univ., Seoul, Korea, Republic of; [3]Catholic Kwandong Univ. Col. Of Medicine, Intl. St. Mary's Hosp., Incheon, Korea, Republic of

Abstract: Low-intensity focused ultrasound is an effective method for inducing blood-brain barrier (BBB) opening, but its underlying mechanisms remain unknown. Here, we investigated the molecular mechanisms of BBB opening induced by low-intensity focused ultrasound. Rats were sacrificed at different timepoints (1, 4, 24, and 48 h) after receiving focused ultrasound sonication (FUS). Immunohistochemistry and western blot were performed to assess levels of tight junction proteins (occludin and ZO-1) and transcytosis proteins (major facilitator superfamily domain-containing 2a [MFSD2a] and caveolin-1). Levels of ZO-1 and occludin were most prominently decreased at 1 h after FUS at the timepoint when the transient BBB opening was most prominent based on Evans blue extravasation. Caveolae formed predominantly at 4 h post-sonication. MFSD2a levels were lower after FUS. At 4 h post-sonication, MFSD2a levels showed the greatest decrease whereas caveolin-1 levels showed the greatest increase. In conclusion, our results highlight a temporal window between transcytosis and tight junction mechanisms. Therefore, the timepoint of injections should be taken into consideration depending on specific characteristics of the drug when delivering a drug following ultrasound-induced BBB opening.

Disclosures: **K. Chang:** None. **Y. Seo:** None. **J. Lee:** None. **C. Kong:** None. **J. Shin:** None. **J. Chang:** None. **Y. Na:** None. **W. Chang:** None.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.09

Topic: I.08. Methods to Modulate Neural Activity

Support: CIHR Grant FDN 154292
CIHR Grant ENG 173742
NSERCC RGPIN-2020-04176
CIHR Grant MFE-171317

Title: Effects of different sonication parameters of theta burst transcranial ultrasound stimulation of human motor cortex

Authors: *K. ZENG[1], Y. DING[1,2], Z. WANG[1,3], X. XIA[1], G. DARMANI[1], T. GRIPPE[1], A. LOZANO[1,4], R. CHEN[1,5];
[1]Krembil Res. Institute, Univ. Hlth. Network, Toronto, ON, Canada; [2]Inst. of Med. Science,

Univ. of Toronto, Toronto, ON, Canada; [3]Sch. of Psychology, Shanghai Univ. of Sport, Shanghai, China; [4]Div. of Neurosurgery, Dept. of Surgery, Univ. of Toronto, Toronto, ON, Canada; [5]Div. of Neurology, Dept. of Medicine, Univ. of Toronto, Toronto, ON, Canada

Abstract: Background: Transcranial ultrasound stimulation (TUS) is a promising non-invasive brain stimulation technique with advantages of high spatial precision and ability to target deep brain regions. Our pervious study found that theta burst TUS (tbTUS) can produce long-term potentiation (LTP)-like plasticity in human motor cortex. However, the effects of different sonication parameters of tbTUS on the induction of brain plasticity have not been investigated. The objective of this study is to examine a range of sonication parameters, including pulse repetition frequency (PRF), duty cycle (DC), sonication duration (SD) and acoustic intensity (AI) to maximize the efficacy of tbTUS while ensuring safety.
Method: We studied 12 right-handed healthy subjects who attended 8 study visits on separate days. One visit applied the standard tbTUS (PRF=5Hz, DC=10%, SD=80s, AI = 20W). In the other 7 visits, one sonication parameter was changed (PRF=2Hz or 10Hz; DC= 5% or 15%; SD=40s or 120s; AI=10W). The effects of tbTUS on motor cortical excitability were tested before, and at 0, 30, 60 and 90 minutes after TUS. The transcranial magnetic stimulation (TMS) measures of motor cortical excitability were motor-evoked potentials (MEP) amplitudes, short interval intracortical facilitation (SICF), short-interval intracortical inhibition (SICI) and facilitation (ICF).
Results: The results showed that standard tbTUS increased motor cortical excitability for at least 60 mins. In contrast, tbTUS with PRF of 2Hz and 10Hz produced significant changes for only 5 mins and 30 mins. Motor cortical excitability increased less than 60 mins with 5% DC and more than 90 mins with 15% DC. 40s train of tbTUS increased motor cortical excitability more than 30 mins and 120s train of tbTUS prolonged the effect to more than 90 mins. When the AI was set as 10W, tbTUS had no effect on motor cortical excitability. In addition, tbTUS reduced SICI and enhanced SICF, but had no effect on ICF.
Conclusion: This work demonstrated 5Hz is the best PRF in tbTUS to induction human plasticity and AI should be ~20W to effectively target the human cortex. Furthermore, higher duty cycle and longer sonication time are associated with longer duration of increased cortical excitability induced by tbTUS. These findings have important implications for the potential use of tbTUS as treatment of neurological and psychiatric disorders.

Disclosures: K. Zeng: None. **Y. Ding:** None. **Z. Wang:** None. **X. Xia:** None. **G. Darmani:** None. **T. Grippe:** None. **A. Lozano:** None. **R. Chen:** None.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.10

Topic: I.08. Methods to Modulate Neural Activity

Support: Internal Lim Lab Funds
SecondWave Systems
DARPA Grant 140D0419C0092

Title: Guiding noninvasive peripheral neuromodulation ultrasound therapy using bone echo detection and spleen stimulation targeting on a wearable platform device.

Authors: *A. B. TUMA[1], A. J. ORGAN[1], C. R. W. KAISER[2], M. ZEBARJADI[2], M. NEWHOFF[1], H. H. LIM[2];
[1]Otolaryngology, [2]Biomed. Engin., Univ. of Minnesota, Minneapolis, MN

Abstract: Electrical vagus nerve stimulation has been shown to reduce systemic inflammation by activating the cholinergic anti-inflammatory pathway (CAP) through the spleen. Our group hopes to accomplish similar effects in humans using low- intensity pulsed ultrasound (US) stimulation delivered directly to the spleen. CAP is controlled by efferent vagus and splenic nerve activity which causes norepinephrine release in the spleen and initiates a signaling cascade involving T-cells and macrophages to inhibit pro-inflammatory cytokine release. We have shown that focused US directed at the spleen decreases inflammation in a mouse model of inflammatory arthritis. We are now developing a wearable US device for human use to stimulate the spleen. In humans, delivery of splenic US is complicated by the rib cage shielding the spleen. A solution is to determine rib location using US echoes received by the therapy device. A small human study was conducted to classify US rib reflection features to indicate the presence of ribs and locate intercostal regions. In a cohort of 20 healthy volunteers (8 male, 12 female) with a mean age of 39 years (range 21-74), the position of the ribs and spleen were first identified using a commercial US imaging system. Next, using the therapy device, US echoes were recorded at five positions over and between two ribs above the spleen. Reflection signals fell into three categories: intercostal, partially over a rib, and fully over a rib. Using a threshold algorithm, we correctly detected a rib 76% of the time and determined that there is no rib 83% of the time. Separating the data set into two groups by body mass index (BMI), using a cutoff of BMI=30, increased classification accuracy to 92% and 96% respectively for low BMI participants and 85% and 88% for high BMI. This is evidence that body type is a significant factor in determining spleen and rib location. To provide automated onboard classification and further improve performance, we are also applying machine learning methods that leverage anatomical data such as BMI, torso landmarks, intercostal space width, and rib and spleen depth as input features to different classifiers to more reliably predict the echo signals. We will compare the accuracy of these classifiers with each other and the thresholding method with the goal of showing near perfect detection of ribs. We will also utilize deep learning algorithms to track respiration-induced splenic motion to provide targeted therapy delivery. These technologies will enable the SecondWave noninvasive US platform to be an effective, cost-efficient and portable peripheral stimulation therapy for the potential treatment of acute and chronic inflammation.

Disclosures: **A.B. Tuma:** None. **A.J. Organ:** None. **C.R.W. Kaiser:** A. Employment/Salary (full or part-time):; Part-time Employee with SecondWave Systems. **M. Zebarjadi:** None. **M. Newhoff:** A. Employment/Salary (full or part-time):; Part-time Employee with SecondWave Systems. **H.H. Lim:** A. Employment/Salary (full or part-time):; Part-time Employee with SecondWave Systems. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Co-founder of SecondWave Systems.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.11

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Project 1RF1MH117080

Title: Auditory Mondrians mask the airborne-auditory confound of ultrasound stimulation in humans

Authors: W. LIANG[1], H. GUO[2], D. MITTELSTEIN[2,3], M. G. SHAPIRO[2], S. SHIMOJO[1], ***M. SHEHATA**[1,4];
[1]Biol. and Biol. Engin., [2]Chem. Engin., Caltech, Pasadena, CA; [3]Keck Sch. of Med. of USC, Los Angeles, CA; [4]Toyohashi Univ. of Technol., Toyohashi, Japan

Abstract: Transcranial ultrasound stimulation (TUS) shows great promise as a powerful, non-invasive focal brain stimulation (Rabut et al., 2020). However, recent studies discovered an auditory confound using common human TUS parameters, which was clearly audible in humans. There is a discrepancy in the literature on whether auditory masking can be used as mitigation (Braun et al., 2020 & Johnstone et al., 2021). Here, we ran three experiments focusing on the airborne-auditory confound. In experiment 1, we traced the possible sources of the auditory confound and tested whether it is audible using a wide range of the parameters used in human TUS. We recorded sound clips of the ultrasound stimulation using a specialized microphone that covers the full-human hearing range — we presented the audio clips online to 50 participants who performed a detection task followed by confidence ratings. We recorded three waveforms: continuous, pulsed, and ramped ultrasound bursts. The results, calculated as D prime, showed that participants were reliably able to hear the auditory artifact with high confidence for most of the presented ultrasound bursts.In experiments 2 (n=80) and 3 (n=98), we texted the effectiveness of auditory tones to mask the TUS auditory confound. Participants performed a two-interval forced-choice (2IFC) task in which they presented online with a mask-only audio clip or a mask with an ultrasound burst one, followed by a confidence rating. Their task was to choose which stimulus contained the ultrasound burst. We used two ultrasound waveforms: continuous and pulsed (in both cases, we used increasing pressure ranging from 0.4 ~ 1.2 MPa). We tested a square wave monotone mask (as in previous reports), a pulsed sin wave monotone mask (to match the auditory artifact), and a random multi-tone mask or “Auditory Mondrians” (inspired by the Mondrians used in the continuous flash suppression to mask visual targets; Tsuchiya & Koch, 2005). Results showed that the correct response dropped to a chance level with 50% confidence using monotone masks for all the continuous ultrasound bursts and low pressure (0.4 MPa) pulsed waveform ultrasound bursts. However, participants achieved around 80 - 90 % correct response with high confidence for the high-pressure pulsed ultrasound bursts (0.8 ~ 1.2 MPa). For Auditory Mondrians, the correct response dropped to a chance level with 50% confidence for all the tested ultrasound bursts, including high-pressure pulsed waveform.

These results show that Auditory Mondrians were more effective than monotone masks at masking the auditory confound of the ultrasound bursts at all the parameters tested.

Disclosures: W. Liang: None. **H. Guo:** None. **D. Mittelstein:** None. **M.G. Shapiro:** None. **S. Shimojo:** None. **M. Shehata:** None.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.12

Topic: I.08. Methods to Modulate Neural Activity

Title: Shockwaves and focused ultrasound are physically different: Accordingly, their interaction with tissue is also different

Authors: *P. NOVAK;
Business Develop., Storz Med. AG, Taegerwilen, Switzerland

Abstract: Low-intensity shockwaves are used in orthopedic, cardiology, urology, dermatology and aesthetics since 1990. Shockwaves transmitted through the skull were first applied to the brain in 2005. Next to shockwaves, there is another focal, mechanical stimulation tool in medicine, the focused ultrasound (FUS). It is used for tissue ablation and drug delivery since the last decade of the 20th century. There are emerging neurological applications with low-intensity tFUS (transcranial Focused Ultrasound) for neuromodulation. Transcranial Pulse Stimulation (TPS) uses focused shockwave pulses. Shockwaves are single pulses with high positive amplitude, very steep leading edge and short duration. Due to the asymmetrical pulse shape broad frequency spectrum results. Typical focal zone of an electromagnetic generator is an ellipsoid with 5mm diameter and 20-30mm length. TPS shockwave pulses have Energy Flux Density (EFD) of up to 0.25mJ/mm^2. Due to the application through the skull, pressure amplitude is reduced to 35% and correspondingly, 85% of energy are absorbed. Compared to shockwaves, ultrasound is a continuous train of sinusoidal pressure oscillations with typical frequency of 0.5 to 5MHz. The frequency spectrum contains only the main frequency with its harmonics. Focused ultrasound can have focal area similar to the focal size of an electromagnetically generated shockwaves. The intensity is expressed as power density (W/cm^2). The skull attenuates transcranial FUS in a similar way like shockwaves. For minimizing the tissue heating capacity of the FUS, the continuous ultrasound train is applied intermittently with repeated pulse bursts. The TPS is used for experimental treatment of Alzheimer's and Parkinson's disease since 2010 with positive results. The average power density is less than 0.1W/cm^2 due to the low repetition pulse rate. There is no heating. TPS working principle is mechanotransduction, which describes the mechanical stimulation of biological processes. tFUS is meanwhile an established method for tissue ablation in the brain. Its working principle is the tissue heating. The neuromodulation applications are still experimental. Here is the average power density <25W/cm^2. The high frequency of the ultrasound pulses compared to the single

shockwave pulses (1’000’000 times) results in tissue temperature increase. The formation of bursts is a compromise between biological effect (neuromodulation) and tissue heating. On the contrary, with TPS there is no tissue heating, although the pressure amplitude is higher.

Disclosures: **P. Novak:** A. Employment/Salary (full or part-time):; part-time employee, Storz Medical AG.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.13

Topic: I.08. Methods to Modulate Neural Activity

Support: CIHR

Title: Metaplasticity associated with transcranial focused ultrasound induced plasticity in humans

Authors: *Y. DING[1], K. ZENG[1], Y. KIM[2], R. CHEN[1];
[1]Univ. Hlth. Network, Toronto, ON, Canada; [2]Univ. of Western Ontario, London, ON, Canada

Abstract: Low intensity transcranial ultrasound stimulation (TUS) is a novel technique for non-invasive brain stimulation (NIBS) that can deliver more focal and deeper stimulation compared to currently used forms of NIBS. Neuroplasticity refers to the ability of the brain to modify its connections in response to internal and external stimuli. It is primarily mediated by long-term potentiation (LTP) and depression (LTD), a persistent strengthening or weakening of synapses. Plasticity induction is further modulated by metaplasticity, which can be homeostatic or non-homeostatic. Homeostatic metaplasticity acts as a negative-feedback mechanism, with a history of LTD enhancing future LTP and inhibiting future LTD. This process is critical for maintaining synaptic volume within the physiological range. LTP is also subject to depotentiation, the abolishment of LTP induction by subsequent stimulation with no plasticity-inducing effects alone.
TUS delivered in a theta burst pattern (tbTUS) induces LTP-like effects in the human primary motor cortex (M1) for 30-60 minutes after 80s of sonication. While homeostatic metaplasticity and depotentiation have been shown to regulate plasticity induction by other forms of NIBS, their effects on TUS are unknown. This study characterized the effects of metaplasticity on TUS by investigating interactions between tbTUS and continuous theta burst stimulation with 150 pulses (cTBS150), a sub-threshold LTD-like repetitive transcranial magnetic stimulation (rTMS) protocol. Based on results in other NIBS, we expected enhanced plasticity induction when cTBS150 is delivered immediately before tbTUS and reversal of plasticity induction when cTBS150 is delivered immediately after.
Four interventions: 1) sham cTBS150 → real tbTUS, 2) real cTBS150 → sham tbTUS, 3) real cTBS150 (priming) → real tbTUS, and 4) real tbTUS → real cTBS150 (depotentiation) were

conducted in randomized order on separate days. To characterize the effects of interventions, TMS measures of average motor-evoked potential amplitude and M1 intracortical circuits (IC) including short-interval intracortical inhibition, long-interval intracortical inhibition, intracortical facilitation, and short-interval intracortical facilitation were measured before and 5, 30, 60, and 90 minutes after each intervention.
The study showed that priming with real but not sham cTBS150 increased the duration of plasticity induction by tbTUS to at least 90 minutes and changes in some ICs may underlie plasticity induction. Depotentiation was observed when cTBS150 was delivered after tbTUS. The results suggest that TUS is modulated in manners consistent with metaplasticity.

Disclosures: **Y. Ding:** None. **K. Zeng:** None. **Y. Kim:** None. **R. Chen:** None.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.14

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH R01NS109794
NIH 1R01NS115797
NSF 1848029
NIH 5T32GM008541-23
NSF DGE-1840990

Title: Activity state-dependent transcranial ultrasound modulation of neural activity in the awake mammalian brain

Authors: *J. SHERMAN[1], E. BORTZ[2], H.-A. TSENG[1], X. HAN[3];
[1]Boston Univ., Boston, MA; [2]Biomed. Engin., Boston Univ., Brookline, MA; [3]Biomed. Engin., Boston Univ., Boston, MA

Abstract: Abstract
Title: Activity state-dependent transcranial ultrasound modulation of neural activity in the awake mammalian brain
Author: Jack. P. Sherman, Emma P. Bortz, Hua-an Tseng, Xue Han
Transcranial ultrasonic neuromodulation offers a promising new technology for the treatment of neurological and psychiatric disorders. Ultrasound has superior spatial resolution when compared to other non-invasive brain stimulation technologies, such as transcranial magnetic stimulation or transcranial direct current stimulation. Thus far, a few studies have demonstrated that ultrasound stimulation can activate neurons in the brain at single cell level, analyzed with calcium imaging or extracellular electrophysiology. However, most studies have used pulsed ultrasound delivered at a high pulse repetition frequency (PRF) of kilohertz rate. To understand whether pulsing ultrasound at slower PRF within the physiological range will produce distinct

neuronal effects, we performed single cell GCaMP calcium imaging in awake head-fixed mice, while delivering transcranial ultrasound stimulation at 350Khz.
We compared ultrasound delivered at a PRF of 10Hz and 140Hz for 1 second. We found that ultrasound stimulation significantly increased calcium activity across all the parameters tested, without significant difference between the parameters. Interestingly, the ultrasound-induced effect in individual neurons strongly correlated with their basal excitability, highlighting the weak modulatory nature of pulsed ultrasound. Together, these results demonstrate that ultrasound stimulation produces weak modulatory effects of neurons, which increases the probability of evoked suprathreshold calcium activities. This observation provides a cellular mechanism for the heterogeneous effect of ultrasound stimulation across individual neurons and highlights the importance of neuronal states in mediating ultrasound stimulation effects.
Acknowledgement X.H. acknowledges funding from NIH (1R01NS109794 and 1R01NS115797), and NSF grant # 1848029. J.S. acknowledges funding from NIH 5T32GM008541-23, and E.B. acknowledges NSF grant # DGE-1840990.

Disclosures: **J. Sherman:** None. **E. Bortz:** None. **H. Tseng:** None. **X. Han:** None.

Poster

581. Ultrasound Techniques for Neuromodulation

Location: SDCC Halls B-H

Time: Tuesday, November 15, 2022, 1:00 PM - 5:00 PM

Program #/Poster #: 581.15

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant 1R01NS109794
NIH Grant 1R01NS115797
NSF Grant 1848029
NSF Grant DGE-1840990
NIH Grant 5T32GM008541-23

Title: Noninvasive transcranial ultrasound stimulation alters neuronal membrane voltage in the brain

Authors: *E. P. BORTZ[1], J. P. SHERMAN[2], X. HAN[1];
[1]Biomed. Engin., Boston Univ., Brookline, MA; [2]Pharmacol. & Exptl. Therapeut., Boston Univ., Boston, MA

Abstract: Transcranial ultrasound is an emerging technique for non-invasive neuromodulation. Ultrasound has good penetrating depth, capable of targeting deep brain structures with millimeter-scale spatial precision, making it appealing for clinical translation. Previous studies demonstrate that transcranial ultrasound can induce various behavioral and neural responses such as muscle twitch, changes in blood oxygenation, and population-level neuronal calcium fluctuations. While these results are promising, ultrasound has been largely inconsistent and unreliable in evoking single-neuron spiking responses. To examine how noninvasive ultrasound

stimulation influences neuronal membrane voltage, we performed optical voltage imaging in mice using the recently developed voltage indicator SomArchon. Specifically, we examined how superficial layer neurons in the motor cortex responded to pulsed ultrasound delivered transcranially at a spatial-peak pulse average intensity of 286 W/cm^2 with a fundamental frequency of 350 kHz. We detected prominent membrane voltage depolarization of individual cortical neurons by transcranial ultrasound, leading to a significant increase in spike rate during ultrasound stimulation compared to the baseline period immediately prior to ultrasound stimulation. These findings provide first direct experimental evidence that noninvasive ultrasound can directly depolarize cortical neuron membrane potential, leading to increased spiking activity.

Disclosures: **E.P. Bortz:** None. **J.P. Sherman:** None. **X. Han:** None.