2018 IBANGS MEETING: THE 20TH ANNUAL GENES, BRAIN & BEHAVIOR MEETING

PROGRAM FOR THURSDAY, MAY 17TH

Days: next day all days

View: session overview talk overview

08:30-16:00 Session FV: Pre-IBANGS Satellite Meeting, Functional Validation for Neurogenetics

Location: Phillips Hall in the Siebens building room 1-11. The Siebens building is located at the Downtown Mayo Clinic Campus (not the Mayo Civic Center).

Description:The transformative nature of next generation sequencing has changed how neuroscientists approach genomic sequence variation. Highly multiplexed molecular testing is providing an expanded level of information from which to make informed phenotypic predictions. The importance of this is reflected in the unprecedented expansion of genomic testing to determine the basis of neurologic conditions. Genomic testing results in many instances provide a definitive basis of a neurologic condition. However, in almost a high proportion of cases, the genomic sequencing results are confounded by the ambiguity of variants with uncertain clinical significance. Herein lies the key with which institutions will lead in the area of genomic medicine. There exists a critical need to provide a mechanism by which uncertain findings can be functionally characterized and translated into clinically actionable results. It is within thisr ealm that academic societies such as IBANGS can have a substantial and informative role on the future of clinical research and practice. This symposium will introduce the challenges and opportunities that exist in the field of human clinical neurogenetics and follow this with presentations of active work in the field of functional genetic finding validation for neurogenetics with a look to the future of genomic neurogenetics.

CHAIRS: *Eric Klee* and *Lisa Schimmenti*
LOCATION: Phillips Hall (Siebens 1-11)

17:00-20:00 Session Reg

Meeting Registration and Information

CHAIR: *Stephanie Ferguson*
LOCATION: Mayo Civic Center Grand Lobby West

18:25-18:30 Session 1: Presidential Welcome

Welcome by Society President and Introduction to Plenary Speaker

CHAIR: *Marissa Ehringer*
LOCATION: Mayo Civic Center 102/103

18:30-19:30 Session 1: Presidential Plenary Lecture

CHAIR: *Marissa Ehringer*
LOCATION: Mayo Civic Center 102/103

19:30-20:30 Session : Welcome Reception

Welcome Reception

LOCATION: Mayo Civic Center Grand Lobby South & Riverfront Plaza

21:00-22:00 Session : Dinner on your own

Dinner on your own

2018 IBANGS MEETING: THE 20TH ANNUAL GENES, BRAIN & BEHAVIOR MEETING

PROGRAM FOR FRIDAY, MAY 18TH

Days: previous day next day all days

View: session overview talk overview

08:00-17:00 Session Reg: Registration

CHAIR: *Stephanie Ferguson*

LOCATION: Mayo Civic Center Grand Lobby West

08:30-10:30 Session 2: Symposium 1

Structure and function of circuits encoding sensory stimuli.

CHAIR: *Mark Wolman*

LOCATION: Mayo Civic Center 102/103

08:30 *Sreekanth Chalasani*

Flexible sensory neural circuits represent chemosensory stimuli in C. elegans

SPEAKER: *Sreekanth Chalasani*

ABSTRACT. Sreekanth H. Chalasani1, Sarah G. Leinwand1, and Laura A. Hale1

1 Molecular Neurobiology Laboratory, The Salk Institute for Biological Studies, 10010 N. Torrey Pines Road, La Jolla, CA 92037.

A central question in neuroscience is to understand how neural circuits extract relevant sensory information from their environment and use that information to drive appropriate responses. We use the small, well-defined nematode C. elegans to reveal how sensory neurons encode both stimulus identity and concentration.

We will present our data suggesting three principles of neural circuit coding. First, we find that odor coding is sparse in C. elegans, similar to other species. Using calcium imaging, we find that a small subset of neurons responds to varying concentrations of food-like odors. Second, we identify that chemosensory circuits are unexpectedly distributed and comprise of primary sensory neurons that directly detect stimuli and recruit secondary neurons to represent stimulus identify and concentration. Finally, we identify a novel form of plasticity in these sensory circuits. In response to particular sensory environments or animal age,

neuropeptides and neurotransmitters select one of several configurations for the active circuit. Consistent with these results, disruptions to the communication between sensory neurons cause significant deficits in odor-guided behaviors. This suggests that these sparse and flexible odor representations are essential for behavior. Furthermore, these flexible circuit configurations might represent alternate paths for information processing and are vital to an animal's ability to respond to the changing environment.

08:55 *Sanchez Jason*

Topography heterogeneity at a central auditory synapse

SPEAKER: *Sanchez Jason*

ABSTRACT. Hui Hong1, Xiaoyu Wang4,5, Ting Lu1, Diego A.R. Zorio4,5, Yuan Wang4,5, and Jason Tait Sanchez1,2,3,

Topography in the vertebrate brain represents an orderly organization of neuronal architecture responsible for encoding sensory information. Topography in the auditory system originates as a gradual change in frequency representation across the peripheral sensory epithelium and is defined by tonotopy; a process maintained in the central pathway. In the avian nucleus magnocellularis (NM) – an auditory brainstem structure analogous to the mammalian cochlear nucleus – tonotopy is maintained along a caudolateral-to-rostromedial axis. In this presentation, I describe novel organization and neuronal biophysics of the caudolateral NM (NMc), a region representing extreme low-frequency sound processing. Examination of neuronal and dendritic properties revealed that NMc neurons contain small somas and extensive dendritic architecture. This is in stark contrast to the adendritic, large neurons located rostromedially that encode higher-frequency sounds. Axonal tract tracing studies confirmed that NMc neurons receive afferent inputs from the auditory nerve, similar to adendritic NM. However, auditory axons synapse onto NMc neurons via small bouton-like terminals, unlike the large Endbulb of Held synapses on adendritic NM neurons. Whole-cell recordings revealed that NMc neurons are significantly more excitable than NM neurons. They generate multiple action potentials to sustained depolarization and rapidly burst fire to low-frequency sinusoidal inputs. Pharmacological and immunohistochemical experiments revealed that this functional phenotype is due to distinct ion channel properties, namely voltage-dependent potassium channels and resurgent sodium currents. Taken

together, NMc neurons are structurally, connectively, and physiologically unique from traditionally defined NM neurons, emphasizing specialized neuronal properties for processing sounds of varying frequencies.

1Roxelyn and Richard Pepper Department of Communication Sciences and Disorders, 2Department of Neurobiology, 3The Hugh Knowles Hearing Research Center, Northwestern University, Evanston, IL, USA, 4Department of Biomedical Sciences, 5Program in Neuroscience Florida State University College of Medicine, Florida State University, Tallahassee

09:20 *Teresa Nicolson*

Insights into the sensory deficits caused by mutation of the deafness genes tomt and tmie

SPEAKER: *Teresa Nicolson*

ABSTRACT. Teresa Nicolson Oregon Hearing Research Center and the Vollum Institute Oregon Health and Science University Portland, Oregon Recent genetic studies have identified essential components of the mechanotransduction complex in sensory hair cells. Although controversial, the Transmembrane channel-like proteins 1 and 2 (Tmc1/2) are promising candidates for the pore forming subunits of the transduction channel in auditory/vestibular hair cells. How the Tmcs are assembled with other members of the mechanotransduction complex and transported to the site of mechanotransduction within the stereocilia of hair bundles is not known. Our studies show that mutations in zebrafish transmembrane o-methyltransferase (tomt), the orthologue of which is required for hearing in humans, abolish localization of Tmc1/2 to hair bundles. We find that tomt is exclusively expressed in hair cells and is enriched in the Golgi compartment. Mutations in tomt cause a complete loss of mechanotransduction in zebrafish hair cells. The phenotype is not due to a developmental defect because acute expression of Tomt expression in mature mutant hair cells is sufficient to rescue the transduction defect. Furthermore, expression of Tomt can restore the localization of Tmc1/2 to stereocilia in mutant hair cells. The loss of mechanotransduction along with normal morphology of hair bundles in tomt mutants is unusual among mechanotransduction mutants. These observations prompted us to take a closer look at another mutant, tmieru1000, with a similar phenotype. Mutations in transmembrane inner ear protein (tmie) are also associated with human deafness and result in functional but not morphological defects in hair cells. We found that

localization of Tmie to the hair bundle is a conserved feature among vertebrates. Like tomt mutants, we discovered that the Tmc1/2 proteins are absent from the hair bundles in mutant tmie hair cells. However, in contrast to Tomt, overexpression of Tmie greatly enhances localization of the Tmcs to the hair bundles, suggesting an additional role of stabilizing protein levels of the Tmc subunits. We propose that Tomt directly or indirectly modifies Tmc1/2 proteins in the Golgi compartment, and Tmie acts in part as a chaperone, enabling the transport of the Tmcs to the site of mechanotransduction in hair cells.

09:45 *Pavel Mašek*

The fat tastes different - the taste perception and discrimination of fatty acids in Drosophila

SPEAKER: *Pavel Mašek*

ABSTRACT. Pavel Mašek1, John M. Tauber2, Elizabeth B. Brown2, Yuanyuan Li1, Maria E. Yurgel2, and Alex C. Keene2 1. Department of Biological Sciences, Binghamton University, Binghamton, NY, 13902, USA. 2. Department of Biological Sciences, Florida Atlantic University, Jupiter, FL 33458, USA. Fat represents a calorically potent food source for many animals. The taste of fat is represented by response to free fatty acids (FAs), one of the building blocks of fat. Previously, we showed that a broad population of sugar-sensing taste neurons expressing Gustatory Receptor 64f (Gr64f), is required for reflexive feeding responses to both FAs and sugars but functional phospholipase C in these neurons is necessary only for FAs perception. Here, we describe specific populations of taste neurons mediating FA response that are identified by expression of Ionotropic Receptor 56d (IR56d). IR56d forms, together with IR76b and IR25a, a receptor for medium-chain FAs. Functional imaging reveals that IR56d-expressing neurons are responsive to short- and medium-chain FAs. Silencing IR56d neurons selectively abolishes response to medium-chain FAs, and their activation is sufficient for feeding responses. Analysis of co-expression with Gr64f identifies two subpopulations of IR56d-expressing neurons. While physiological imaging reveals that both populations are responsive to FAs, IR56d/Gr64f neurons are activated by short- and medium-chain FAs and are sufficient for reflexive feeding responses. Behaviorally, we show that flies display different response to FAs and sugars relative to their intensity. They also discriminate between sugars and FAs in an aversive taste memory assay, further supporting the notion that

FA taste is a unique modality. In addition, we test the discrimination ability between multiple classes of FAs and sweet stimuli that allows us to categorize appetitive taste stimuli into functional groups and help us to understand the principles of taste perception and coding.

10:10 *Marc Wolman*

Pregnancy associated plasma protein-aa (Pappaa) regulates photoreceptor synaptic development to mediate visually guided behavior

SPEAKER: *Marc Wolman*

ABSTRACT. To guide behavior, sensory systems detect the onset and offset of stimuli and process these distinct inputs via parallel pathways. In the retina, this strategy is implemented by splitting neural signals for light onset and offset via distinct synapses connecting photoreceptors to ON and OFF bipolar cells, respectively. It remains poorly understood which molecular cues establish the architecture of this synaptic configuration to split light onset and offset signals. A mutant with reduced synapses between photoreceptors and one bipolar cell type, but not the other, could reveal a critical cue. From this approach, we report a novel synaptic role for pregnancy associated plasma protein aa (pappaa) in promoting the structure and function of cone synapses that transmit light offset information. Electrophysiological and behavioral analyses indicated pappaa mutant zebrafish have dysfunctional cone to OFF bipolar cell synapses and impaired responses to light offset, but intact cone to ON bipolar cell synapses and light onset responses. Ultrastructural analyses of pappaa mutant cones showed a lack of presynaptic domains at synapses with OFF bipolar cells. pappaa is expressed postsynaptically to the cones during retinal synaptogenesis and encodes a secreted metalloprotease known to stimulate insulin-like growth factor 1 (IGF1) signaling. Induction of dominant negative IGF1 receptor expression during synaptogenesis reduced light offset responses. Conversely, stimulating IGF1 signaling at this time improved pappaa mutants' light offset responses and cone presynaptic structures. Together, our results indicate Pappaa-regulated IGF1 signaling as a novel pathway that establishes how cone synapses convey light offset signals to guide behavior.

10:30-11:00 Session : AM Break

AM Break

LOCATION: Mayo Civic Center 104/105

11:00-12:30 Session 3: Outstanding Travel Awardees

Junior Faculty, Postdoctoral, and Student Outstanding Travel Awardees

CHAIR: *Mark Rutledge-Gorman*

LOCATION: Mayo Civic Center 102/103

11:00 *Anna Warden*

Sex-dependent effects of toll-like-receptor 3 activation on alcohol intake

SPEAKER: *Anna Warden*

ABSTRACT. It is unclear if sex differences in immune response can influence alcohol drinking behavior. We tested the possibility that toll-like receptor 3 (TLR3) activation regulates alcohol consumption in a sex-dependent manner. Male and female mice were injected with the TLR3 agonist polyinosinic:polycytidylic acid (poly I:C; 5 mg/kg) and brain proinflammatory responses and alcohol consumption (every-other-day two-bottle choice) were measured. In males, poly I:C produced a peak cytokine response at 3 hours post-injection. In contrast, poly I:C injection resulted in a delayed and prolonged cytokine response lasting up to 72 hours post-injection in females. Access to ethanol at peak cytokine activation decreased ethanol consumption in both sexes; whereas, access to ethanol after peak cytokine activation resulted in an increase in ethanol consumption in males, with no change in ethanol consumption in females. Repeated poly I:C and ethanol exposures altered innate immune transcript abundances. Decreased levels of the Myeloid Differentiation response gene 88 (MyD88)-dependent pathway correlated with decreased alcohol intake in females; whereas, increased levels of the TIR-domain-containing adapter-inducing interferon β (TRIF)-dependent pathway correlated with increased alcohol intake in males. We validated that the effect of poly I:C was mediated through MyD88-dependent signaling in females by testing in knockout female mice lacking Myd88. Poly I:C did not alter alcohol intake in these animals. Our results provide novel evidence that there are sex-dependent differences in TLR3 activation and that enhanced TRIF-dependent pathway expression may regulate escalation of alcohol intake. Supported by: U01 AA020926, P01 AA020683, AA013520, AA006399, F31 AA025499-02.

1Waggoner Center for Alcoholism and Addiction Research, University of Texas at Austin, Austin, TX 78712, USA 2Institute for Neuroscience, University of Texas at Austin, Austin, TX 78712, USA 3Department of Neurology, Dell Medical

School, University of Texas at Austin, Austin TX 78712, USA.

11:23 *Dana Zeid*

Transgenerational effects of nicotine exposure on fear learning, gene expression, and cholinergic signaling

SPEAKER: *Dana Zeid*

ABSTRACT. Recent work has shown that the effects of nicotine use may be transgenerationally transmitted through epigenetic modifications. For the present study, we examined the effects of paternal nicotine exposure on fear learning in subsequent generations. Male adult C57BL6/J mice received either chronic nicotine or saline exposure and were crossed to naïve female C57BL/6J mice. The offspring of nicotine (Nic-Sired) and saline (Sal-Sired) exposed mice were tested in contextual and cued fear conditioning. Results indicated that paternal nicotine exposure resulted in enhanced cued and contextual fear learning in F1 and F2 generations compared to Sal-Sired mice, and this effect was reversed when F1 generation, but not F2 generation, mice received acute nicotine injections. Furthermore, Nic-Sired mice also showed more pronounced spontaneous recovery of fear when re-tested following extinction. We additionally examined general cholinergic activity in the Nic-Sired mice using nicotinic acetylcholine receptor (nAChR) binding and potassium and nicotine-evoked acetylcholine release in the dorsal and ventral hippocampus. Reduced cholinergic functioning was found in ventral, but not dorsal, hippocampus in the Nic-Sired mice along with global hippocampal increases in nAChR binding. In parallel, we also found increased DNA methylation in the ventral hippocampi of Nic-Sired mice. Finally, RNA-sequencing indicated differential expression of cholinergic synapse associated genes in Sal-Sired and Nic-Sired mice. Together, our results suggest that paternal nicotine exposure may result in alterations in the epigenome, which, in turn, leads to exaggerated fear learning and abnormal cholinergic function in subsequent generations.

1Department of Biobehavioral Health, Penn State University, 2Department of Pharmacology, Vanderbilt University, 3Department of Psychology, Temple University Funding: DA017949; 1U01DA041632 (NIDA, T.G.), DA037421 (NIDA, V.P.)

11:46 *Amy Dunn*

Gene x Diet Interactions Modify Symptoms of Alzheimer's Disease

SPEAKER: *Amy Dunn*

ABSTRACT. Alzheimer's disease (AD) is complex, with both genetic (G) and environmental (E) factors regulating disease progression. Identification of GxE interactions that modulate AD pathogenesis is critical to developing novel and personalized treatments. However, extracting GxE effects is challenging in humans due to human genome complexity and difficulty controlling environmental factors. To overcome these barriers, we developed a panel of genetically diverse mice carrying human familial AD mutations (AD-BXDs). Because the AD-BXDs model some genetic heterogeneity of humans, they are ideally suited to investigate translationally-relevant GxE interactions. Here, we used AD-BXDs to determine how genetics and diet interact to modify AD-related pathogenesis. We fed a chronic high fat diet (HFD) to 10 strains of AD-BXDs and monitored metabolic and cognitive function before and after HFD. Control groups included AD-BXDs on chow, and nontransgenic BXDs on chow and HFD. We observed accelerated working memory decline in AD-BXDs on HFD compared to controls. However, this was dependent on genetic background, with some AD-BXD strains maintaining cognitive function on HFD. Subsequent analyses indicated gene-by-diet interactions accounted for 18% of individual variation in memory decline. Higher body weight and adiposity were protective against working memory decline in nontransgenic BXDs, but not in AD-BXDs. Our results suggest that diet and genetic background interact to mediate vulnerability to AD pathogenesis, and that metabolic factors may contribute to cognitive decline differentially in normal aging versus AD. Future analyses will identify genetic and molecular targets contributing to AD pathogenesis that may be exploited to delay, prevent or treat AD.

(1) The Jackson Laboratory, Bar Harbor, ME, USA, (2) The University of Tennessee Health Science Center, Memphis, TN, USA Funding support: NIA 1 R01 AG057914-01 to C.C.K.; NIA 1 R01 AG054180-01A1 to C.C.K; JAX Director's Innovation Fund to K.M.S.O; Alzheimer's Association AARF-18-565506 to A.R.D.

12:08 *Sarah Bergen*

Joint contributions of rare CNVs and common SNPs to risk for schizophrenia

SPEAKER: *Sarah Bergen*

ABSTRACT. Sarah E. Bergen, PhD1,2; Alexander Ploner, PhD1; Daniel Howrigan, PhD3; CNV Analysis Group and the Schizophrenia Working Group of the Psychiatric Genomics Consortium; Michael C. O'Donovan4, MD, PhD; Jordan W. Smoller, MD, ScD5; Patrick F. Sullivan, MD, FRANZCP1,6; Jonathan Sebat, PhD7; Benjamin Neale, PhD3; Kenneth S. Kendler, MD8

Both rare copy number variants (CNVs) and common single nucleotide polymorphisms (SNPs) contribute to liability to schizophrenia, but their etiological relationship has not been fully elucidated. We evaluated an additive model, whereby risk of schizophrenia requires less contribution from common genetic variation in the presence of a rare CNV and test for interactions. Individual genetic data from 21,094 schizophrenia cases and 20,227 controls from the Psychiatric Genomics Consortium were used. We assessed three classes of rare CNVs: those previously associated with schizophrenia, large deletions ≥500kb, and total CNV burden. We compared mean polygenic risk scores (PRS) between subjects with and without rare CNVs and modeled the joint effects of PRS and CNVs on schizophrenia liability using logistic regression. Schizophrenia cases carrying risk CNVs have lower polygenic risk than without CNVs, but still higher than controls. For cases carrying known risk CNVs, the PRS was diminished proportional to the effect size of the CNV. The strongly-associated 22q11.2 deletion required little added PRS to produce schizophrenia. Large deletions and increased CNV burden were also associated with lower polygenic risk in cases. However, the results for controls suggested more complex relationships. We found evidence for interactive effects of PRS and previously associated CNVs for risk for schizophrenia, while our results for large deletions and total CNV burden support an additive model. These findings offer insights into the genetic architecture of schizophrenia by illuminating how different established genetic risk factors act and interact to influence liability to schizophrenia.

1 Department of Medical Epidemiology and Biostatistics, Karolinska Institutet, Stockholm, Sweden 2 Stanley Center for Psychiatric Research, Broad Institute of MIT and Harvard, Cambridge, MA, USA 3 Analytic and Translational Genetics Unit, Massachusetts

General Hospital, Boston, MA, USA 4 MRC Centre for Neuropsychiatric Genetics and Genomics, Division of Psychological Medicine and Clinical Neurosciences, Cardiff University, Cardiff, UK 5 Harvard Medical School, Department of Psychiatry, Massachusetts General Hospital, Boston, MA 02114, USA; Psychiatric and Neurodevelopmental Genetics Unit, Center for Human Genetics Research, Massachusetts General Hospital, Boston, MA 02114, USA 6 Departments of Genetics and Psychiatry, University of North Carolina, Chapel Hill, NC, USA 7 Departments of Psychiatry and Cellular and Molecular Medicine, University of California, San Diego, CA, USA 8 Virginia Institute for Psychiatric and Behavioral Genetics, Department of Psychiatry, Virginia Commonwealth University, Richmond, VA, USA

12:30-13:30 Session L1: Lunch

Lunch

LOCATION: Mayo Civic Center 104/105

13:30-15:30 Session 4: Selected Talks I

Selected Talks 1

CHAIR: *Elissa Chesler*

LOCATION: Mayo Civic Center 102/103

13:30 *Sean Farris*

Molecular and behavioral assessment of long non-coding RNAs in alcohol use disorder

SPEAKER: *Sean Farris*

ABSTRACT. S.P. Farris1, CM Borghese1, E.A Osterndorff-Kahanek1, Y.A. Blednov1, G.E. Homanics2, R.D. Mayfield1, and R.A. Harris11Waggoner Center for Alcohol and Addiction Research The University of Texas at Austin, Austin, TX, 78712-1095, U.S.A.; 2 Departments of Anesthesiology and Pharmacology and Chemical Biology, University of Pittsburgh PA, 15261

Transcriptional regulation of gene expression is an important aspect of human health and diseases. The human transcriptome contains both protein-coding and non-coding RNAs (ncRNAs), with ncRNAs greatly outnumbering protein-coding transcripts. Despite the large number, few ncRNAs have been assigned names, biologically characterized, or implicated in disease. A large fraction of ncRNAs, particularly long non-coding RNAs (lncRNAs), are specifically expressed in brain. To test the contribution of altered lncRNA expression in

alcohol use disorder (AUD) we conduced RNA-Seq profiling of multiple human postmortem brain regions (n=385 samples). Focusing on lncRNAs with evidence for evolutionary conservation, our analyses identified over 100 unique cross-validated lncRNAs associated with AUD. Using Xenopus oocytes as an expression system, we evaluated the potential biological impact of candidate human lncRNAs on known alcohol-responsive ion-channels. One lncRNA, markedly increased in AUD, decreased NMDAR subunit GRIN1 and GRIN2B expression ($p < 1e-03$, n=10/group). Two-electrode voltage clamp recordings showed an ~80% reduction in maximum glutmate currents, without affecting other ion-channels (e.g. GABA-A or Glycine). To test the potential role of this lncRNA in vivo we generated a knockout mouse for the predicted homolog using CRISPR/Cas9. The lncRNA KO mouse showed a significant increase for alcohol-induced loss of righting response (LORR) ($p < 0.01$, n=8/group). Consistent with NMDAR oocyte studies, lncRNA KOs also showed enchanced ketamine-induced LORR; however, no differences were observed for the GABAergic sedative gaboxadol. Overall, our work supports the functional importance of lncRNAs, and validates a novel candidate for an alcohol-related behaivor affecting NMDARs. Supported NIAAA (K99AA024836 and U01AA020926).

13:45 *Sa-Ik Hong*

Adenosine A2A Receptor in the Dorsomedial Striatum Regulates Alcohol-Seeking Behavior though Top-Down Inhibitory Pathway

SPEAKER: *Sa-Ik Hong*

ABSTRACT. Sa-Ik Hong1, Seungwoo Kang1, and Doo-Sup Choi1, 2, 3

Top-down control facilitates cortico-limbic circuits for the decision-making process. Dysregulation of this process is implicated in addiction including alcohol use disorder (AUD). Activation of striatopallidal neurons is known to suppress hierarchical response in reward-seeking behavior. Adenosine A2A receptor (A2AR) is expressed in the indirect inhibitory circuit. Although A2AR is co-expressed with dopamine D2 receptor (D2R) in the medium spiny neurons (MSN) in the striatum, the precise role of A2AR in the dorsomedial striatum (DMS) in reward-seeking behaviors through top-down inhibitory pathway has not been investigated. In our study, to investigate a DMS-specific role of A2AR in top-down ethanol-seeking behavior, first, we trained

mice to voluntarily seek ethanol in the nose-poke operant chambers. Then, to examine the effect of A2AR, we measured neuronal activity in the DMS and a major output targeting neurons in the external part of globus pallidus (GPe). Using in vivo pharmacologic and optogenetic techniques, we manipulated neural activities in the A2AR expressing DMS neurons to GPe. We found that mice pre-exposed to ethanol exhibited increased top-down reward-seeking behavior. Pharmacological activation of A2AR and optogenetic stimulation to A2AR-containing indirect pathway suppressed top-down response. In contrast, A2AR-inhibition and optogenetic suppression of DMS-GPe circuit reversed top-down inhibitory response. Taken together, activation of striatopallidal A2AR and indirect pathway dampens top-down ethanol-seeking behavior, indicating that activation of A2AR signaling in the striatal indirect pathway could be a potential therapeutic target for AUD.

1Department of Molecular Pharmacology and Experimental Therapeutics, 2Neurobiology of Disease Program, 3Department of Psychiatry and Psychology, Mayo Clinic College of Medicine, Rochester, Minnesota 55905, USA.
Funding Support: NIH AA018779, Samuel C. Johnson for Genomics of Addiction Program at Mayo Clinic, Ulm Foundation, Godby Foundation, David Lehr Research Award from American Society for Pharmacology and Experimental Therapeutics

14:00 *Spencer Huggett*

Cocaine'omics: The Genetic and Neurological Underpinnings of Cocaine Use and Dependence – An Unorthodox Replication

SPEAKER: *Spencer Huggett*

ABSTRACT. Spencer B. Huggett1,2 & Michael C. Stallings1,2

1Department of Psychology and Neuroscience, University of Colorado 2Institute for Behavioral Genetics, University of Colorado Supported in part by: NIDA P60 DA11015

We investigated the genetic and transcriptional landscape of cocaine dependence (CD) and chronic cocaine use. We performed and integrated popular genome-wide and transcriptome-wide analyses using data from the largest genome wide association study (GWAS) on CD to date (Gelernter et al. 2014), 3,176 European Americans (EAs), and human post-

mortem brain tissue from seven cocaine users and eight drug free controls. First, linkage disequilibrium (LD) score regression analyses was performed and detected a significant genomic heritability of 28% (s.e = 0.14) for CD and gene-based association tests found three novel genes underlying this heritability: the C1QL2, KCTD20 and STK38 genes. Tissue specificity analyses indicated robust enrichment in numerous brain regions, including the hippocampus, padj = 2.02e-06. Therefore using RNA-sequencing (RNA-seq) analyses we performed differential expression and weighted gene covariance network analyses (WGCNA) on post-mortem hippocampal brain tissue from Zhou et al. 2011. Differentially expressed genes or transcripts between chronic cocaine users versus drug free controls were enriched for genes associated with CD ($p < 0.05$), OR = 1.34, p = 0.031, and were used to find various potential therapeutic compounds for cocaine use/toxicity. Lastly, we found that KCTD20 was a central part of a hippocampal gene network strongly associated with cocaine use and thus, might be contributing to the genetic liability of CD by disrupting intricate gene networks in the brain. Overall, our study elucidates the biological architecture of cocaine use/dependence, proposes various novel therapeutic compounds for cocaine use and includes an alternative framework to validate/provide biological meaning to genome-wide findings.

14:15 *Amy Lasek*

Genome-wide transcriptional changes in the rat hippocampus during withdrawal from chronic alcohol drinking identifies altered neuroimmune signaling

SPEAKER: *Amy Lasek*

ABSTRACT. WY Chen1, H Chen1, Y Chen1, H Zhang1, HR Krishnan1, C Liu1, DR Grayson1, SC Pandey1,2, & AW Lasek1 Chronic alcohol drinking and withdrawal causes epigenetic and transcriptional changes in the brain that may contribute to relapse. In order to find novel genes altered during withdrawal, we performed a genome-wide analysis of transcripts in the rat hippocampus. Male adult Sprague Dawley rats were fed an ethanol or control liquid diet for 15 days and withdrawn for 24 hours. Hippocampal RNA was isolated and subjected to RNA-Seq. Weighted gene co-expression network analysis (WGCNA) was used to identify modules of co-expressed genes and demonstrated that the genes fit into 53 co-expression modules. Genes

in module 1 were significantly higher during withdrawal and were enriched in the “TNF signaling pathway” and “Epstein Barr virus Infection” by gene ontology analysis, indicating disruptions in neuroimmune signaling during withdrawal. To validate the RNA-Seq findings, hippocampal RNA and chromatin were isolated for qPCR and chromatin immunoprecipitation (ChIP) analyses, respectively. Expression of Tnfrsf1a, Stat3, Relb, Plat, Serpine1, and Timp1 were increased during withdrawal, similar to the RNA-Seq findings. Decreased acetylation of histone H3 lysine 9/14 was also observed at the promoters of these genes. Notably, treatment with the histone deacetylase inhibitor suberoylanilide hydroxamic acid (SAHA) during withdrawal normalized expression and histone acetylation at the promoters of the Tnfrsf1a, Stat3, and Relb genes. Our results demonstrate that transcriptional changes in neuroimmune genes occur in the hippocampus during withdrawal from chronic ethanol drinking and that some of these changes can be reversed by SAHA treatment, suggesting epigenetic regulation of neuroimmune genes and possible involvement in pathophysiology of alcoholism.

1University of Illinois at Chicago, Center for Alcohol Research in Epigenetics and Department of Psychiatry & 2Jesse Brown VA Medical Center, Chicago, IL 60612 USA, NIAAA P50 AA022538 (to AWL and SCP) and U01 AA020912 (to AWL).

14:30 *David Linsenbardt*

Encoding of the Intent to Drink Alcohol by the Prefrontal Cortex is blunted in Rats with a Family History of Excessive Drinking

SPEAKER: *David Linsenbardt*

ABSTRACT. David N. Linsenbardt, Nicholas M. Timme, & Christopher C. Lapish

Indiana Alcohol Research Center and Department of Addiction Neuroscience - Psychology, Indiana University – Purdue University Indianapolis, Indianapolis, IN 46202.

Aberrant decision-making is both a risk factor for, and the result of, an Alcohol Use Disorder (AUD). The decision to use alcohol can be robustly influenced by exposure to stimuli associated with the drug, and these stimuli are a critical underlying factor that contributes to compulsive drinking and relapse. Additionally, the identification of risk factors for AUD’s in human epidemiological studies and animal models has

led to the view that there is a strong genetic component to the disease. Therefore, identifying heritable alterations in computation that occur in brain regions that guide the decision to seek and consume alcohol is critical to develop novel intervention strategies. The prefrontal cortex plays a central role in guiding decision-making, and is altered by alcohol use and familial (genetic) risk for excessive drinking. Thus, we chose to record neural signals from the medial Prefrontal Cortex (mPFC) of animals given the opportunity to drink alcohol. We used an information theoretic statistical approach to quantify and compare the amount of information neurons provided about trial-to-trial alcohol drinking decisions. Animals without familial risk (Wistar rats) exhibited patterns of neural activity consistent with the intention to drink or abstain, whereas these patterns were blunted or absent in animals ('P' rats) with high risk. These data indicate that computations guiding drinking decisions are not encoded by the mPFC in populations with increased familial risk, possibly indicating a lack of control over decision-making by this otherwise well-validated supervisory brain region.

Acknowledgments: This work was supported by NIAAA grant #'s AA022268 (DNL), AA025120 (DNL), AA007462 (NMT), AA022821 (CCL), AA023786 (CCL), and the Indiana Alcohol Research Center P60AA007611 (D. Kareken). This research was supported in part by Lilly Endowment, Inc., through its support for the Indiana University Pervasive Technology Institute, and in part by the Indiana METACyt Initiative. The Indiana METACyt Initiative at IU is also supported in part by Lilly Endowment, Inc.

14:45 *Jacob Beierle*

Systems genetics, fine mapping, and validation of candidate genes involved in opioid and psychostimulant addiction traits in a reduced complexity cross

SPEAKER: *Jacob Beierle*

ABSTRACT. Jacob A. Beierle 1,2,,3, Lisa R. Goldberg 1,2, Julia C. Kelliher 1, Kimberly P. Luttik 1, Julia L. Scotellaro 1, Alex M. Luong 1,4, Jiayi Wu 3,5, Eric R. Reed 6, David F. Jenkins 6,7, Qiu T. Ruan 1,2,3, Ali Al Abdullatif 8, Stacey L. Kirkpatrick 1, Cory Parks 9, Christine Watkins 9, Morgan Dickerson 9, Sufiya Khanam 9, Sydney B. Crotts 1, Timothy A. Drescher 1, Neema Yazdani 1,2,3, Robert W. Williams 9, Gregg E. Homanics 10, William E. Johnson 7,

Benjamin Wolozin 8, Megan K. Mulligan 9, Camron D. Bryant 1

Murine forward genetic studies of addiction traits can identify genetic factors and biological pathways relevant to humans. Reduced Complexity Crosses (RCC) facilitate gene identification by decreasing the number of candidate variants by up to 300-fold. We used an RCC between C57BL/6J (B6J) and C57BL/6NJ (B6NJ) substrains to map opioid (oxycodone; OXY) and psychostimulant (methamphetamine; MA) behaviors in our Multi-Stage Addiction Assessment Protocol (MSAAP). We identified a major QTL on distal chr. 1 underlying OXY-induced locomotor activity and anxiety-like withdrawal. We fine mapped this locus to 167-174 Mb by backcrossing select recombinant F2 mice. Cis-expression (eQTL) combined with transcript/behavior correlation identified Pcp4l1, Atp1a2, and Cadm3 as positional/functional candidate genes. Transcriptome analysis of the chr. 1 locus identified a dual-hub network of trans-downregulated genes comprising neurodegenerative proteins APP and TAU. Mapt (TAU) knockout mice show preliminary enhanced OXY tolerance and withdrawal, suggesting TAU protects against chronic opioid-induced neurobehavioral plasticity. We also mapped a QTL on medial chr. 5 for increased MA-induced locomotor activity that contains the cis-modulated α2 subunit of the GABA-A receptor (Gabra2). The B6J allele harbors an intronic deletion resulting in decreased mRNA and protein levels. The causal role of Gabra2 on MA-induced locomotor activity was confirmed using gene editing to repair the deletion in B6J. Lower Gabra2 levels associated with the B6J allele confer a greater MA behavioral response compared to the repair allele. Studies are underway to validate Gabra2 in conditioned aversive behaviors induced by the opioid receptor antagonist naloxone that also mapped to chr 5.

1. Laboratory of Addiction Genetics (R01DA039168, R21DA038738), Departments of Pharmacology and Experimental Therapeutics and Psychiatry, Boston University School of Medicine, Boston, MA USA 2. Biomolecular Pharmacology Training Program (T32GM008541), Boston University School of Medicine, Boston, MA USA 3. Transformative Training Program in Addiction Science (1011479), Burroughs Wellcome Fund 4. Masters Program in Biomedical Sciences, Boston University, Boston, MA USA 5. Genetics and Genomics, Program in Biomedical Sciences, Boston University, Boston, MA USA 6. Ph.D. Program in Bioinformatics, Boston University,

Boston, MA USA 7. Computational Biomedicine, Boston University School of Medicine, Boston, MA USA 8. Laboratory of Neurodegeneration, Departments of Pharmacology and Experimental Therapeutics and Neurology, Boston University School of Medicine, Boston, MA USA 9. Departments of Genetics, Genomics, and Informatics and Anatomy and Neurobiology, University of Tennessee Health and Science Center, Memphis, TN USA 10. Departments of Anesthesiology, Neurobiology, and Pharmacology and Chemical Biology, University of Pittsburgh, Pittsburgh, PA USA

15:00 *Clarissa Parker*

Using genome-wide association and RNA sequencing to identify genes associated with ethanol sensitivity in Diversity Outbred mice

SPEAKER: *Clarissa Parker*

ABSTRACT. CC Parker1, VM Philip2, DM Gatti3, A Holmes4, EJ Chesler3 1Department of Psychology and Program in Neuroscience, Middlebury College, VT 05753 2Center for Computational Sciences, The Jackson Laboratory, 600 Main Street, Bar Harbor, ME 04609 3Center for Mammalian Genetics, The Jackson Laboratory, 600 Main Street, Bar Harbor, ME 04609 4Laboratory of Behavioral and Genomic Neuroscience, NIAAA, NIH, Rockville MD 20852

A strong predictor for the development of alcohol use disorders (AUDs) is altered sensitivity to the intoxicating effects of alcohol. Individual differences in the initial sensitivity to alcohol are controlled at least in part by genetic factors. Mice offer a powerful tool for elucidating the genetic basis of behavioral and physiological traits relevant to AUDs; but conventional experimental crosses have only been able to identify large chromosomal regions rather than specific genes. Genetically diverse, highly recombinant mouse populations allow for the opportunity to observe a wider range of phenotypic variation, offer greater mapping precision, and thus increase the potential for efficient gene identification. We have taken advantage of the Diversity Outbred (DO) mouse population to identify and map narrow quantitative trait loci (QTL) associated with ethanol sensitivity. We phenotyped 778 JAX DO mice for three measures of ethanol sensitivity: ataxia, hypothermia, and loss of the righting response. We used high density MEGAMuga and GIGAMuga arrays to obtain genotypes ranging from 77,808 – 143,259 SNPs. We also measured

striatal gene expression using RNA sequencing. We identified multiple QTLs associated with ethanol sensitivity related traits. With the inclusion of RNA-Seq we are able to apply a systems genetic strategy to construct the network of correlations that exist between DNA sequence, gene expression values and ethanol-related phenotypes. This information can in turn be used to identify alleles that contribute to AUDs in humans, elucidate causative biological mechanisms, or assist in the development of putative treatment strategies.

15:15 *David Jentsch*

Evaluation of Nav1 as a candidate gene influencing intravenous cocaine self-administration in inbred mice

SPEAKER: *David Jentsch*

ABSTRACT. JD Jentsch1, M Zheng2, A Arslan2, G Peltz2 Cocaine use disorder is a substantially heritable trait in humans, yet relatively few candidate genes have been associated with it. To address this issue, we examined intravenous cocaine self-administration in adult male mice derived from a panel of >70 inbred mouse strains. Most strains readily acquired operant responding reinforced by delivery of 0.5 mg/kg/infusion of cocaine. After 10 days of testing, levels of intake were relatively stable at an individual level but were highly variable across strains. The broad sense heritability of levels of intake at the end of the acquisition period was >0.8. Haplotype-based computational genetic mapping (HBCGM) was conducted, using the cocaine self-administration phenotypes of 21 inbred strains for which whole genome sequence data exists. This analysis revealed that multiple genes within a region (134-7MB) on mouse chromosome 1 show an allelic pattern strongly associated with strain differences in cocaine self-administration. Based upon the presence of associated non-synonymous coding SNPs within it and its brain expression profile, Nav1, a member of the neuron navigator gene family, was evaluated further. Its expression in striatal tissue in BxD mice was analyzed using Genenetwork.org (record ID: ILM6620129). The Nav1 eQTL mapped to the Nav1 locus. Strain level variation in Nav1 expression correlated negatively with dopamine D2 receptor mRNA expression (record ID:15186; r=-.78; p=4.23e-08) and positively with locomotor response to cocaine (record ID:10320; r=0.59, p=4.34e-03). These studies suggest that variation within Nav1 may powerfully influence the tone of

dopaminergic transmission in brain, in turn producing heritable variation in cocaine-evoked behaviors.

1Department of Psychology (Behavioral Neuroscience), Binghamton University; 2Department of Anesthesia, Stanford University School of Medicine

15:30-16:00 Session Break: PM Break

PM Break

LOCATION: Mayo Civic Center 104/105

16:00-18:00 Session P1: Poster Session I

Poster Session 1

LOCATION: Mayo Civic Center 104, 105, 106

16:00 *Jeyeon Lee*

1. Mapping neuro-pharmacological effect of harmaline in pig

SPEAKER: *Jeyeon Lee*

ABSTRACT. Jeyeon Lee[1], Inyong Kim[2], Hoonki P. Min[3], In MyungHo[3], Hang Joon Jo[2], and Su-Youne Chang[1, 4]

Harmaline induced tremor is the most commonly utilized disease model for essential tremor. However, underlying mechanisms of harmaline-induced tremor have not yet been fully elucidated. Therefore, understanding tremorgenesis mechanism of harmaline will be crucial to develop a novel treatment for essential tremor. In this study, we tried to define the harmaline effect at the system level. To do so, we performed whole brain imaging via fMRI (functional magnetic resonance image) in swine and analyzed an acute effect of harmaline injection in BOLD (blood-oxygen level dependent) signal. As results, we observed significant BOLD changes in inferior olivary nucleus (ION), cerebellum, and thalamus, which are known as tremor-related regions (n= 5; $p < 0.0005$, one-sample *t*-test). In addition, significant increase of inter-regional correlation between cerebellum and deep cerebellar nuclei and between cerebellum and thalamus was also found (n= 5, $p < 0.001$, Wilcoxon signed-rank test). Our results of this study suggest ph-fMRI (pharmachological fMRI) as an applicable method to evaluate novel therapeutic agents and/or neuromodulatory therapies for essential tremor and the swine model of tremor as an acute animal model for essential tremor.

[1]Department of Neurologic Surgery, Mayo Clinic

[2]Department of Neurology, Mayo Clinic

[3]Department of Radiology, Mayo Clinic

[4]Department of Physiology and Biomedical Engineering, Mayo Clinic

This study was funded by the National Institutes of Health, National Institute of Neurological Disorders and Stroke (NS 88260).

16:00 *Kyle Flippo*

3. Mapping a novel endocrine circuit regulating alcohol consumption

SPEAKER: *Kyle Flippo*

ABSTRACT. KH Flippo1,2, SO Idiga1,2, KE Claflin1,2, MC Naber1,2, MJ Potthoff1,2

In the United States alcohol use disorder (AUD) affects ~15% of adults with the prevalence of binge drinking on the rise in adolescents and young adults. AUD represents a major issue to healthcare given that chronic excessive alcohol consumption in humans is associated with cardiovascular disease, metabolic syndrome, and cancer while acute alcohol intoxication can prove lethal. Economically, AUD represents a massive burden due to loss of productivity and associated healthcare costs. Recently, the endocrine hormone fibroblast growth factor 21 (FGF21), known for its potent metabolic effects, was illustrated to significantly reduce alcohol consumption via an undescribed mechanism requiring expression of the obligate FGF21 co-receptor ß-klotho (KLB) in the brain. Importantly, single nucleotide polymorphisms (SNPs) in both FGF21 and KLB genomic loci are highly associated with increased alcohol consumption in humans. Here we extend those findings illustrating that FGF21 can reverse alcohol consumption even in mice chronically administered ethanol prior to FGF21 administration. Furthermore, excessive alcohol consumption promotes FGF21 secretion from the liver perhaps representing a homeostatic feedback loop to regulate alcohol consumption. However, the target of FGF21 in the brain mediating these effects remains unclear. Excitingly, we have observed FGF21 dependent activation of neurons in the piriform cortex. Additionally, deletion of KLB in glutamatergic neurons significantly increases alcohol

consumption in mice. These findings represent a novel endocrine circuit regulating alcohol consumption in an FGF21 dependent manner. Future studies will focus on mapping this circuit and characterization of neurons expressing KLB.

1Department of Pharmacology, University of Iowa Carver College of Medicine, Iowa City, IA, 2Fraternal Order of Eagles Diabetes Research Center, University of Iowa Carver College of Medicine, Iowa City, IA

16:00 *Leonard Schalkwyk*

5. A histone acetylome-wide association study of Alzheimer's disease: neuropathology-associated regulatory variation in the human entorhinal cortex

SPEAKER: *Leonard Schalkwyk*

ABSTRACT. Sarah J. Marzi1, Szi Kay Leung2, ^, Teodora Ribarska3, ^, Eilis Hannon2, Adam R. Smith2, Ehsan Pishva2,4, Jeremie Poschmann2,5, Karen Moore3, Claire Troakes1, Safa Al-Sarraj1, Stephan Beck6, Stuart Newman7, Katie Lunnon2, Leonard C. Schalkwyk7+, Jonathan Mill2+*

1 Institute of Psychiatry, Psychology & Neuroscience, King's College London, London, UK. 2 University of Exeter Medical School, University of Exeter, Exeter, UK. 3 German Cancer Research Center (DKFZ), Heidelberg, Germany. 4 Department of Psychiatry and Neuropsychology, Maastricht University Medical Centre, Maastricht, The Netherlands.5 Centre de Recherche en Transplantation et Immunology, Inserm, Université de Nantes, Nantes, France. 6 UCL Cancer Institute, University College London, London, UK. 7 University of Essex, Colchester, UK ^ / + Equal contributions

Alzheimer's disease (AD) is a chronic neurodegenerative disorder characterized by the progressive accumulation of amyloid-β (Aβ) plaques and neurofibrillary tangles in the neocortex. Recent studies have implicated a role for regulatory genomic variation in AD progression, finding widespread evidence for altered DNA methylation associated with neuropathology. To date, however, no study has systematically examined other types of regulatory genomic modifications in AD. In this study, we quantified genome-wide patterns of lysine H3K27 acetylation (H3K27ac) - a robust mark of active enhancers and promoters that is strongly correlated with gene expression and transcription factor binding - in entorhinal cortex samples from AD cases and matched controls using chromatin

immunoprecipitation followed by highly parallel sequencing (ChIP-seq). Across ~182,000 robustly detected H3K27ac peak regions, we found widespread acetylomic variation associated with AD neuropathology, identifying 4,162 differential peaks (FDR < 0.05) between AD cases and controls. These differentially acetylated peaks were enriched in disease-specific biological pathways and include regions annotated to multiple genes directly involved in the progression of Aβ and tau pathology (e.g. APP, PSEN1, PSEN2, and MAPT), as well as genomic regions containing variants associated with sporadic late-onset AD. Partitioned heritability analysis highlighted a highly-significant enrichment of AD risk variants in entrohinal cortex H3K27ac peak regions. Finally, targeted gene expression analysis showed that variable H3K27ac is associated with transcriptional variation at proximal genes including CR1, GPR22, KMO, PIM3, PSEN1 and RGCC. This is the first study of variable H3K27ac yet undertaken in AD and the largest study investigating this modification in the entorhinal cortex. In addition to identifying molecular pathways associated with AD neuropathology, we present a framework for genome-wide studies of histone modifications in complex disease, integrating our data with results obtained from genome-wide association studies as well as other epigenetic marks profiled on the same samples.

16:00 *John Henley*

7. Glucocorticoid regulation of ependymal glia and regenerative potential after vertebrate spinal cord injury
SPEAKER: *John Henley*

ABSTRACT. CM Nelson1,2, H Lee3, RG Krug3, A Kamilova1, NN Madigan4, KJ Clark3, VA Lennon2,4,5, AJ Windebank4, JR Henley1,6

Following injury, the mammalian spinal cord forms a glial scar and fails to regenerate. In contrast, spinal cord tissue of vertebrate fish regenerates and swimming movements recover. The mechanisms underlying functional regeneration are not fully understood. Here we report that the glucocorticoid pathway regulates functional neural regeneration by directly affecting ependymal glia. Cord transection in larval zebrafish (Danio rerio) causes paralysis and neural cell death, with subsequent ependymal glial proliferation, extension of bipolar glia across the lesion, and neurogenesis. Functional connectivity is restored by axons extending from spared and nascent neurons along trans-lesional glial bridges. Studies in the

transgenic SR4G reporter zebrafish reveal downregulation of both the glucocorticoid receptor Nr3c1 and glucocorticoid signaling activity in ependymal glia follow injury. Functional recovery is impaired by dexamethasone (Dex) treatment, which attenuates injury-induced ependymal glial proliferation, bridging, and neural tissue regeneration, and is independent of haematopoietic-derived immune cells. Loss-of-function mutagenesis of nr3c1 reverses functional impairment by Dex. By contrast, in the adult rat, NR3C1 levels and signaling activity in ependymal glia are upregulated following spinal cord transection. The unanticipated negative regulation of neural regeneration by glucocorticoid signaling via a direct effect on ependymal glia calls into question the putative benefit of corticosteroid therapy in early management of spinal cord injury. Indeed, therapeutic down-regulation of CNS glucocorticoid receptors might improve patient outcomes.

Departments of 1Neurological Surgery, 2Laboratory Medicine and Pathology, 3Biochemistry and Molecular Biology, 4Neurology, 5Immunology, and 6Physiology and Biomedical Engineering, Mayo Clinic, College of Medicine, Rochester, MN, USA 55905. Present address, AK: Vanderbilt Brain Institute, Vanderbilt University School of Medicine, Nashville, TN, USA 37232. Funding Support: NIH-NINDS (USA) R01 NS67311.

16:00 *Svitlana Bach*

9. CRISPR/dCas9 manipulation of Bdnf transcription in rat primary hippocampal neurons

SPEAKER: *Svitlana Bach*

ABSTRACT. SV Bach, D Hosein, D Williams, NV Gallus, FA Sultan, KD Bunner, KE Savell & JJ Day

Brain-derived neurotrophic factor (Bdnf) plays a critical role in brain development, neuronal differentiation, dendritic growth and synaptic plasticity. Rodent Bdnf gene consists of nine 5' non-coding exons (I-IXa) and one 3' coding exon (IX). Each non-coding exon has its own promoter region where transcription of different variants is initiated. To investigate specific roles of the activity-regulated Bdnf variants I and IV we used a CRIPSR/dCas9 – VPR system, in which a strong transcriptional activator, VPR, is targeted to Bdnf I and IV promoter regions with the help of specific guide RNAs (gRNAs). Using this system in primary rat hippocampal neurons we are able

to selectively upregulate Bdnf variants I and IV from their endogenous gene loci while leaving other variants mostly unaffected. To assess functional significance of selective Bdnf variant upregulation, we used Multichannel Electrode Arrays (MEAs) to perform single-unit recordings from neurons treated with CRISPR constructs. Upregulation of select Bdnf variants causes an increase in the spike frequency as well as the number of spontaneously active neurons. To assess subcellular localization of Bdnf mRNAs, single-molecule RNA fluorescent in situ hybridization (FISH) was used to visualize individual Bdnf I, IV and IX transcripts, which occupy diverse cellular compartments upon neuronal depolarization with potassium chloride. With this work we demonstrate the unprecedented precision of the endogenous Bdnf transcript variant upregulation using CRIPSR/dCas9 tools, the functional significance of Bdnf transcript variant upregulation for neuronal physiology, and the subcellular localization of Bdnf mRNAs with unparalleled resolution.

Department of Neurobiology, University of Alabama at Birmingham, Birmingham, AL 35294, USA

16:00 *Amy Lasek*

11. Genome-wide transcriptional changes in the rat hippocampus during withdrawal from chronic alcohol drinking identifies altered neuroimmune signaling

SPEAKER: *Amy Lasek*

ABSTRACT. Chronic alcohol drinking and withdrawal causes epigenetic and transcriptional changes in the brain that may contribute to relapse. In order to find novel genes altered during withdrawal, we performed a genome-wide analysis of transcripts in the rat hippocampus. Male adult Sprague Dawley rats were fed an ethanol or control liquid diet for 15 days and withdrawn for 24 hours. Hippocampal RNA was isolated and subjected to RNA-Seq. Weighted gene co-expression network analysis (WGCNA) was used to identify modules of co-expressed genes and demonstrated that the genes fit into 53 co-expression modules. Genes in module 1 were significantly higher during withdrawal and were enriched in the “TNF signaling pathway” and “Epstein Barr virus Infection” by gene ontology analysis, indicating disruptions in neuroimmune signaling during withdrawal. To validate the RNA-Seq findings, hippocampal RNA and chromatin were isolated for qPCR and chromatin immunoprecipitation (ChIP) analyses,

respectively. Expression of Tnfrsf1a, Stat3, Relb, Plat, Serpine1, and Timp1 were increased during withdrawal, similar to the RNA-Seq findings. Decreased acetylation of histone H3 lysine 9/14 was also observed at the promoters of these genes. Notably, treatment with the histone deacetylase inhibitor suberoylanilide hydroxamic acid (SAHA) during withdrawal normalized expression and histone acetylation at the promoters of the Tnfrsf1a, Stat3, and Relb genes. Our results demonstrate that transcriptional changes in neuroimmune genes occur in the hippocampus during withdrawal from chronic ethanol drinking and that some of these changes can be reversed by SAHA treatment, suggesting epigenetic regulation of neuroimmune genes and possible involvement in pathophysiology of alcoholism.

1University of Illinois at Chicago, Center for Alcohol Research in Epigenetics and Department of Psychiatry & 2Jesse Brown VA Medical Center, Chicago, IL 60612 USA, NIAAA P50 AA022538 (to AWL and SCP) and U01 AA020912 (to AWL).

16:00 *Sa-Ik Hong*

13. Adenosine A2A Receptor in the Dorsomedial Striatum Regulates Alcohol-Seeking Behavior though Top-Down Inhibitory Pathway

SPEAKER: *Sa-Ik Hong*

ABSTRACT. Sa-Ik Hong1, Seungwoo Kang1, and Doo-Sup Choi1, 2, 3

Top-down control facilitates cortico-limbic circuits for the decision-making process. Dysregulation of this process is implicated in addiction including alcohol use disorder (AUD). Activation of striatopallidal neurons is known to suppress hierarchical response in reward-seeking behavior. Adenosine A2A receptor (A2AR) is expressed in the indirect inhibitory circuit. Although A2AR is co-expressed with dopamine D2 receptor (D2R) in the medium spiny neurons (MSN) in the striatum, the precise role of A2AR in the dorsomedial striatum (DMS) in reward-seeking behaviors through top-down inhibitory pathway has not been investigated. In our study, to investigate a DMS-specific role of A2AR in top-down ethanol-seeking behavior, first, we trained mice to voluntarily seek ethanol in the nose-poke operant chambers. Then, to examine the effect of A2AR, we measured neuronal activity in the DMS and a major output targeting neurons in the external part of globus pallidus (GPe). Using in vivo pharmacologic and optogenetic techniques, we manipulated neural activities in the A2AR

expressing DMS neurons to GPe. We found that mice pre-exposed to ethanol exhibited increased top-down reward-seeking behavior. Pharmacological activation of A2AR and optogenetic stimulation to A2AR-containing indirect pathway suppressed top-down response. In contrast, A2AR-inhibition and optogenetic suppression of DMS-GPe circuit reversed top-down inhibitory response. Taken together, activation of striatopallidal A2AR and indirect pathway dampens top-down ethanol-seeking behavior, indicating that activation of A2AR signaling in the striatal indirect pathway could be a potential therapeutic target for AUD.

1Department of Molecular Pharmacology and Experimental Therapeutics, 2Neurobiology of Disease Program, 3Department of Psychiatry and Psychology, Mayo Clinic College of Medicine, Rochester, Minnesota 55905, USA.
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16:00 *Mohammed Althobiti*

15. Hospital Pharmacists' Knowledge of Pharmacogenetics in Saudi Arabia
SPEAKER: *Mohammed Althobiti*

ABSTRACT. Mohammed A. AlThobiti1 and Rayan M. AlSaadi2

1 PhD. Molecular Genetics, King Fahd Security College, Riyadh, Kingdom of Saudi Arabia 2 PharmD, Collage of Pharmacy, Taif University, Taif, Kingdom of Saudi Arabia

OBJECTIVE: To explore the practice and knowledge of hospital pharmacists in Saudi Arabia regarding pharmacogenetics.

METHODS: This was a cross-sectional study conducted in 2018 that included hospital pharmacists' from the five regions in Saudi Arabia. A validated self-administered survey collected demographics and information on the pharmacist's knowledge regarding pharmacogenetics.

RESULTS: Four hundred and forty-six questionnaires were returned with a full of response rate. Only 6.5% of the participants they assessed themselves as having knowledge of pharmacogenetics. Most participants (82.6%)

believed that genetic variation affects the body's response to the medication. More than half of the respondents (60.9%) said they believe that pharmacogenetics affects the provision of patient counseling. Most participants (76.4%) reported that they had never studied pharmacogenetics. All participants (100%) stated that they had never trained in pharmacogenetics. The majority of participating pharmacists (80.4%) believe that they should have knowledge of pharmacogenetics. About two-thirds (63%) of the participants believe that pharmacogenetics reduces the economic losses resulting from medication substitution.

CONCLUSION: Pharmacists at hospital pharmacies had lacking knowledge of pharmacogenetics. Lack of pharmacogenetics training and education has been identified as the major barrier to knowledge. We recommend that pharmacy colleges be required to introduce pharmacogenetics into the curriculum and to increase the number of pharmacogenetics courses for hospital pharmacists'.

16:00 *Alejandro Ferrer*

17. Functional evaluation of a novel mutation in DOCK3 associated with neurological symptoms

SPEAKER: *Alejandro Ferrer*

ABSTRACT. A Ferrer1, MT Zimmermann2, K Namekata3, T Harada3, MA Cousin1, JL Kemppainen4, KJ Mack5, M Shinawi6, D Babovic-Vuksanovic4, EW Klee1.

Affordable next generation sequencing has increased drastically the number of variations linked to neurologic diseases; however, functional studies remain the gold standard to establish the causal genotype-phenotype link. Here, we describe the functional validation of a homozygous mutation in the gene for the Dedicator Of CytoKinesis 3 (DOCK3) found in a patient with a neurological phenotype.

The proband is a 3-year-old female with macrocephaly, global developmental delay, hypotonia and autism spectrum disorder. High arched palate and dental anomalies were also noted. Brain and spinal MRI showed ventriculomegaly and hippocampal head atrophy, and diffuse atrophy of the thoracic spinal cord. Other tests were unremarkable. Using whole exome sequence (WES) in blood samples from the trio, we uncovered a homozygous mutation in DOCK3 (c.5020A>T; p.Met1674Leu). Her healthy parents were both heterozygous.

DOCK3 induces axonal growth in the brain, where it is mainly expressed. This variant is not described in mutation databases (ClinVar and HGMD), and represented at low percentage (0.12%) in gnomAD. In silico tools predict the variant as benign, although DOCK3 is statistically intolerant to missense variation. Protein 3D modeling indicated that the residue change occurred in the hydrophobic core of the protein's catalytic domain, and in vitro transfection experiments concluded that the catalytic activity was impaired as a consequence. This information made us conclude that this variant is responsible for the proband's neurologic symptoms.

This study highlights the relevance of using functional experiments to complement and validate the connection between WES findings and the phenotype from the patient tested.

1Center for Individualized Medicine. Mayo Clinic, Rochester MN. USA 2Genomics Science & Precision Medicine Center. Medical College of Wisconsin, Milwaukee WI. USA 3Visual Research Project. Tokyo Metropolitan Institute of Medical Science, Tokyo. Japan 4Clinical Genomics. Mayo Clinic, Rochester MN. USA 5Pediatric Neurology. Mayo Clinic, Rochester MN. USA 6Division of Genetics and Genomic Medicine. Washington University School of Medicine, St. Louis MO. USA

16:00 *Louis El Khoury*

19. Sink or Swim: Impacts of prolonged stress on zebrafish physiology and behavior

SPEAKER: *Louis El Khoury*

ABSTRACT. LY El Khoury1, HB Lee1, RG Krug1,2, AN Sigafoos1, KJ Clark1 1Department of Biochemistry and Molecular Biology, Mayo Clinic, MN, USA 2Mayo Clinic School of Medicine, Mayo Clinic, MN, USA

The hypothalamic-pituitary-adrenal (HPA) axis mediates vertebrate-specific stress responses. Alterations in HPA axis activity are a causative and critical prognostic factor in many psychiatric disorders including depression. Stress during the early critical period is deemed to have a long-lasting effect throughout an organism's lifespan. However, early life stressors can either lead to increased susceptibility or resilience to future stressors. To better understand gene-environment interactions during these critical developmental periods, we are developing assays to mimic chronic stress in the rapidly developing zebrafish. Currently, we have subjected larval zebrafish (5 days post

fertilization) to prolonged, unpredictable stress (10-hr random on/off shaking overnight). Unpredictable shaking increased levels of glucocorticoid receptor (GR) activation and decreased innate immune response to a wound site. We will be further testing parameters of the unpredictable shaking assay, including the impact of various HPA axis mutants, and the impact of this early stressor on adult response to forced beach test (FBT), which measures coping in a similar manner as the forced swim test in rodents. Our assay suites will characterize the effect of early life stress on the physiology and behavioral of larval fish and the coping styles of adults, thereby providing a platform for genetic screening on potential susceptibility or resilience factors.

16:00 *Price Dickson*

21. Systems genetics discovery of genetic, genomic, and gene-by-environment mechanisms driving substance use and sensation seeking

SPEAKER: *Price Dickson*

ABSTRACT. Price E. Dickson1, Tyler A. Roy1, Troy D. Wilcox1, Guy Mittleman2, Elissa J. Chesler1

1 The Jackson Laboratory, 600 Main Street, Bar Harbor, ME 04609, USA 2 Department of Psychological Science, Ball State University, Muncie, IN 47306, USA

Substance abuse is a critical public health issue with genetic and environmental causes. Sensation seeking is a multifaceted, heritable trait which predicts the development of substance use and abuse in humans, and similar phenomena have been observed in rodents. Genetic correlations among substance use and sensation seeking indicate shared biological mechanisms. Environmental enrichment attenuates both traits suggesting that effects occur through these shared mechanisms. The molecular and neurobiological mechanisms underlying these relationships remain elusive. We used a systems genetics approach in BXD recombinant inbred (RI) mice to identify (1) genetic mechanisms driving intravenous cocaine self-administration and (2) shared genetic mechanisms underlying operant sensation seeking and alcohol preference. To assess the feasability of using the BXD RI panel to discover the mechanisms through which environmental factors influence the shared mechanisms

underlying substance use and sensation seeking, we quantified the effects of environmental enrichment on operant sensation seeking, preference for a novel environment, and locomotion in a novel environment in the C57BL/6J and DBA/2J inbred strains, the founder strains of the BXD RI panel. We identified strain-dependent effects of housing condition on each of these distinct indexes of sensation seeking. Collectively, these data provide novel and, in some cases, shared biological mechanisms driving substance use and sensation seeking in the BXD RI mouse panel and provide evidence of genotype-dependent effects of environmental enrichment on sensation seeking traits in the BXD founder strains.

This work was supported by NIDA K99 DA043573 to PED and NIDA R01 DA037927 to EJC.

16:00 *Christopher Kliethermes*

23. Genetic analysis of ethanol-stimulated locomotion in food-deprived and fed flies

SPEAKER: *Christopher Kliethermes*

ABSTRACT. Acute food deprivation results in increased voluntary locomotion in a variety of vertebrate and invertebrate species, and increases the subjective, behavioral, and motivational effects of multiple drugs of abuse. The current experiments leveraged the locomotor stimulating effects of food deprivation and low-dose ethanol in an attempt to disentangle genes that regulate food- and drug-related behaviors in D melanogaster. Male and female flies from 37 Recombinant Inbred (RI) lines were ad lib fed or acutely food deprived prior to testing for basal and ethanol-stimulated locomotion. Food deprivation resulted in a marked increase in basal locomotion and a modest increase in ethanol-stimulated locomotion overall, with RI line accounting for 30-40% of the total phenotypic variation in these measures. Genome-wide association analyses identified 36 variants associated with basal locomotion in food deprived flies at LOD > 6, 13 of which were also associated with ethanol-stimulated locomotion, and two of which, VMAT and DAT, have been implicated previously in the expression of drug-related behaviors. Analysis of results from fed flies identified 3 variants associated with ethanol-stimulated locomotion, including one, fili, that also associated with basal and ethanol-stimulated locomotion in food deprived flies, as well as with body weight. These results suggest that food deprivation and low-dose ethanol induce locomotion via pathways that are largely

genetically distinct in D melanogaster, but which might share some underlying genetic modifiers. Ongoing studies are examining the function of several of the identified genes in basal and ethanol-stimulated locomotion, as well as in other food- and drug-related behavioral assays. 1Drake University, Department of Psychology and Neuroscience, Des Moines IA

16:00 *Elizabeth Catudio Garrett*

25. Regulation of neuron communication and development by the matricellular protein dCCN

SPEAKER: *Elizabeth Catudio Garrett*

ABSTRACT. EL Catudio Garrett1,2, T Pallister1, S Dufner1, and SJ Certel1,2

The extracellular matrix (ECM) provides critical biochemical and physical signals to initiate and support a diverse array of cellular functions. Within the nervous system, both neurons and glia secrete molecules that contribute to the ECM. The CCN (CYR61, CTGF, NOV) family of proteins functions in a primarily regulatory role rather than structural. Specific members are highly expressed in the cerebral cortex, hippocampus, and cerebellum, contributing to neuron development, differentiation, and synaptic plasticity. However, the contributions of CCN family members and matricellular proteins to neuron communication and synaptic transmission remain understudied. Here we examine the role of the sole Drosophila CCN (dCCN) family member in the developing and mature nervous system. dCCN is expressed in motorneurons and interneurons throughout embryonic, larval, and adult stages. In the adult, dCCN expression is found in peripheral neurons located in the legs, wing, and proboscis that respond to sensory information and in the ventral nerve cord in neurons that innervate the reproductive system and motorneurons. In the central brain, our results demonstrate dCCN is expressed in octopamine (OA) neurons, and neurons that express the male form of Fruitless, a key regulator of male-specific behavior. Therefore, we examined the requirement for dCCN function in OA neurons using male aggression and courtship as readouts of neuron output. Our observations indicate that a reduction of dCCN in OA or all neurons results in decreased male aggression. Taken together, this work will contribute to understanding the role of matricellular proteins and the ECM in regulating neuron function and communication.

1Division of Biological Sciences, The University of Montana, Missoula, MT, United States
2Cellular, Molecular and Microbial Biology Graduate Program, The University of Montana, Missoula, MT, United States, Funded by NIH RO1 GM115510, USA

16:00 *Lisa Goldberg*

27. Utilizing the BXD genetic reference panel to identify the causal genetic variants of nicotine withdrawal deficits in hippocampal learning

SPEAKER: *Lisa Goldberg*

ABSTRACT. L.R. Goldberg1, M.G. Kutlu1, D. Zeid1, S., Gadiwalla1, T.J. Gould1

Cognitive deficits, such as disrupted learning, are a major symptom of nicotine withdrawal. These deficits are heritable, yet the genetic basis is unknown. Mice are valuable for identifying novel genes that contribute to variation in traits associated with various stages of addiction, including withdrawal. Our lab has developed a mouse model of nicotine withdrawal deficits in hippocampus-dependent learning, using chronic nicotine exposure via osmotic minipumps and fear conditioning. Here, we are using the recombinant inbred BXD genetic reference panel to identify novel genetic variants related to nicotine withdrawal deficits in learning. Male and female mice (n=4-8 per sex per strain, 30 strains) received either chronic saline or nicotine (6.3 mg/kg per day for 12 days), and then were tested for hippocampus-dependent learning deficits using fear conditioning. Preliminary QTL analyses using Genenetwork (1000 permutations) identified a significant QTL on chromosome 4 (96.9 Mb, LRS =35.7, $p<0.05$). A key advantage to utilizing the BXD lines is the wealth of available data. Upon completion of behavioral phenotyping, we will begin to prioritize candidate genes using behavioral and gene expression correlations in Genenetwork. Finally, RNA-sequencing in the BXD lines exhibiting extreme phenotypic variation will be used to identify hippocampal transcriptome changes associated with nicotine withdrawal.

1Department of Biobehavioral Health, Penn State University, University Park, PA, USA Funding Support: U01DA04163202

16:00 *Wim Crusio*

29. Effects of prenatal stress on the behavior of Fmr1 knock-out mice
SPEAKER: *Wim Crusio*

ABSTRACT. W.E. Crusio1, V. Lemaire-Mayo1, W. Fyke1,2, M. Premoli1,3, E. Subashi1, A. Delprato1,4, S. Pietropaolo1.

Fragile X Syndrome (FXS), the most common heritable cause of mental retardation, is due to a triplet repeat in the X-linked FMR1 gene that results in a loss of expression of the protein, FMRP. There exists a mouse model for the disorder, in which the homologous mouse gene, Fmr1, has been knocked out, also resulting in a loss of expression of the FMRP protein. This model thus has a good construct validity and therefore likely also has good predictive validity. One drawback of the mouse model is that the behavioral deficits that it displays are much milder than those observed in humans. We therefore wanted to see whether adding an environmental insult, prenatal stress, would result in more severe deficits. We subjected pregnant dams (genetic background C57BL/6J) that had been housed with a male for 2 weeks to unpredictable chronic mild stress and tested the offspring (WT and KO males, WT and heterozygous females) at the age of 7 weeks. Stress altered activity in the open field and social interaction of WT mice only. Stress also induced the appearance of cognitive deficits in the Y maze in KO mice, usually absent at this young age. These effects were sex dependent (mostly observed in males) and demonstrate interesting gene-environment interactions in the development of FXS in this mouse model.

1CNRS and University of Bordeaux, Institut de Neurosciences Cognitives et Integratives d'Aquitaine- UMR5287, Pessac, France. 2SUNY Downstate Medical Center - Neural & Behavioral Science Program, Department of Pathology, Brooklyn- NY, USA. 3University of Brescia, Department of Molecular & Translational Medicine, Brescia, Italy. 4BioScience Project, Wakefield, MA, USA.

16:00 *Philipp Pottmeier*

31. A function investigation of novel Y chromosome encoded long non-coding RNAs expressed in human male CNS during early development
SPEAKER: *Philipp Pottmeier*

ABSTRACT. The prevalence, age of onset, and clinical symptoms of many neuropsychiatric disorders - including alcoholism - differ substantially between males and females. We hypothesize, that the sexual differentiation of the human brain during development, is a major factor contributing to the difference in susceptibility to neurological disorders between males and females. The current concept of central nervous system sexual differentiation during development includes, not only the action of gonadal hormones, but also genes encoded on the sex chromosomes. We are investigating whether gametologous regions of the X and Y chromosome are involved in the formation of sex differences in the human brain, especially during very early central nervous system development (less than 12 weeks after gestation), before the production of sex hormones by the primordial sex organs. To do this, we differentiate human embryonic and neuronal stem cells to mature neurons/glia and investigate differences in gene expression patterns between male and female cells. In addition, marker genes for differentiation, as well as parameters for proliferation, motility and morphology are being observed. We are also using CRISPR/Cas9 gene editing for a functional investigation of novel long non-coding RNAs encoded on the Y chromosome, which have been previously identified by our group and are implicated in the early development of the nervous system in human male (manuscript under revision). Our study will shed light on differences between male and female brain development, and thus sets the basis for advances in one of the most neglected issues in medical science, sex differences.

Uppsala University, Department of Organism Biology, Unit for Evolution and Development, Uppsala, Sweden. Funding support: Swedish Research Foundation, Grant name: Sex determination factors in the brain encoded in the Y chromosome (Project number K2012-61X-22089-01-3)

16:00 *Kayla Townsley*

33. The role of Phosphodiesterase type 4 (Pde4) in chronic binge-like drinking

SPEAKER: *Kayla Townsley*

ABSTRACT. Townsley, K.G. 1, 2, 3, Tran, A.T.D. 1, 2, Firsick, E.J. 1, 2, Hack, W. 1, 2, Batish, T. 1, 2, Kanadibhotla, S. 1, 2, Crabbe, J.C. 1, 2, Ozburn, A.R. 1,2

Recent studies provide strong evidence regarding a potential role for phosphodiesterase (PDE) inhibitors in the regulation of alcohol drinking in mice, rats, and humans. PDE4 inhibitors increase cAMP signaling and striatal activity in the brain. Rolipram and apremilast target multiple PDE4 isoforms with varying affinities. PDE4A and PDE4B are highly expressed in the nucleus accumbens (NAc), an important point of convergence in stress/reward-pathways. We investigated the role of PDE4 in high-intensity, binge-like alcohol drinking in High Drinking in the Dark mice (HDID-1).

HDID-1 female mice experienced 8 weeks of a 4-days/week Drinking in the Dark (DID; ethanol or water) paradigm. NAc tissue was collected and processed for multiplex qRT-PCR (n=6 mice/treatment/time point) to determine whether ethanol altered the expression of PDE4. Next, we tested whether rolipram (0, 5, 7.5, or 10mg/kg) or apremilast (0, 20, or 40 mg/kg) reduced ethanol drinking during DID (n=11-12/sex/dose). To determine whether inhibition of PDE4 in the NAc was sufficient to reduce DID, we administered apremilast intra-accumbens (0 or 2ug, via bilateral cannulae; n=15-17/group). We also tested the effects of apremilast on intake of other fluids and tastants.

Chronic binge-like drinking increased Pde4b expression in the NAc ($p<0.01$). Rolipram and apremilast reduced binge-like drinking (both drugs $p<0.0001$) and BALs (rolipram $p<0.05$; apremilast $p<0.0001$). Further, intra-accumbens administration of apremilast selectively reduced ethanol intake ($p<0.05$). These data provide support for the use of apremilast as a potential new therapeutic for the treatment of high-intensity binge-like drinking.

1Department of Behavioral Neuroscience, Oregon Health & Science University, Portland, Oregon, 97239 USA; 2Portland Alcohol Research Center, Research and Development, VA Portland Health Care System, Portland, Oregon, 97239 USA; 3Department of Biology, Portland State University, Portland, Oregon, 97201 USA

Supported by NIH grants (AA13519; AA10760; AA020245), US Department of Veterans Affairs Grants (BX000313; CDA2 BX002488), NIH Common Fund and Office of Scientific Workforce Diversity (UL1GM118964; TL4GM118965; RL5GM118963), and the John R. Andrews Family.

16:00 *Jessica Huebschman*

35. Fragile X mental retardation protein regulates dendritic branching and spine morphology in the striatum following repeated cocaine administration.

SPEAKER: *Jessica Huebschman*

ABSTRACT. Jessica Huebschman1, Miles Fontenot2, Feba Thomas2, Chris Cowan2,3, Laura N. Smith1,2

Cocaine, and other drugs of abuse, cause synaptic changes in the brain, altering circuitry to produce potentially long-lasting effects. Such exposure in rodents causes an increase in dendritic spine density and synaptic strength on medium spiny neurons (MSNs) in both the nucleus accumbens (NAc) and dorsal striatum – brain regions important for goal-oriented and habitual behaviors that are likely involved in the development of addiction. In this study, we identify a role for the fragile X mental retardation protein (FMRP), an RNA binding protein, in regulating these changes. FMRP controls the translation of hundreds of brain RNAs, many of which are involved in synaptic function. Loss of this protein, as seen in fragile X syndrome (FXS), results in an increase in dendritic spines, particularly immature spine types, in cortical and hippocampal brain regions, suggesting that FMRP is involved in regulating spine maturation and elimination. Here we show that lack of FMRP in Fmr1 knockout (KO) mice allows cocaine-induced increases in dendritic branching and spine density in NAc at a time point when it is not yet observed in wild-type animals, suggesting that FMRP limits this process. Interestingly, the observed increase primarily involves thin spine types, typically considered more immature and labile. We also observed a significant cocaine-induced increase in overall spine density in the dorsal lateral striatum of Fmr1 KO animals at this time point. Ongoing work in our lab is investigating the role of FMRP more specifically in regulating synaptic morphology and function.

1 Department of Neuroscience and Experimental Therapeutics, College of Medicine Texas A&M Health Science Center, 2 Department of Psychiatry, The University of Texas Southwestern Medical Center, 3 Departments of Neuroscience and Psychiatry & Behavioral Sciences, Medical University of South Carolina

16:00 *Susan Bergeson*

37. Translational Model Improvement for Alcohol Use Disorder: Voluntary Consumption Behavior in Hybrid Swine
SPEAKER: *Susan Bergeson*

ABSTRACT. B.L. Backus1, J.M. Martinez2, D.C. Curtis2, J.E. Bertrand2, P.J. Syapin2, and S.E. Bergeson2

1Texas Tech University and 2Texas Tech University Health Sciences Center, Lubbock, Texas 79430

Only three pharmacotherapeutics for Alcohol Use Disorder (AUD) are FDA approved and none are widely used (<10%) or show a strong effect to reduce alcohol consumption in the long-term (<20% see sustained outcomes). Unfortunately, ~10% of the population suffers from AUD and over 5% of all medical morbidities share risky ethanol consumption as an underlying issue. As a consequence, intoxication, in general, and 'alcohol addiction' (severe AUD), in particular, are important clinical problems with a compelling need for new treatment. Although the age of 'big data' and high throughput genomics has increased potential targets for medications development, overwhelming rodent use is an increasing research concern. We recently showed that tetracycline analogs reduced drinking, and wished to test a species with more biological similarity to humans. Swine have successfully been used for medical purposes and share better genetic similarity to humans than rodents. At TTU's research farm we used a Large White x Landrace hybrid cross to test the overarching hypothesis that swine would effectively model AUD. Escalating % ethanol, two-bucket choice showed preference levels of ~70% and intoxication. Pharmacokinetic elimination was similar to humans. Finally, we tested whether naltrexone and minocycline would reduce consumption; both drugs reduced drinking, and minocycline decreased preference. Voluntary consumption to biologically relevant BACs, human-relevant alcohol elimination, and drug reduction of 'risky' drinking support the hypothesis that swine may be a useful translational model for AUD.

Supported by the TTU/TTUHSC Presidential Initiative and The Laura W. Bush Institute for Women's Health.

16:00 *Alyssa Moore*

39. Effects of genetic deletion of Syn3 on operant reversal learning in mice
SPEAKER: *Alyssa Moore*

ABSTRACT. AN Moore1, J Linden1, JD Jentsch1

There is substantial evidence to suggest a relationship between a lack of inhibitory control and an increased susceptibility to drug abuse and addiction. Previous research suggests that an individual's capacity for inhibitory control is heritable, and genome-wide linkage studies in BxD mice identified Syn3, which encodes the synaptic phosphoprotein synapsin III, as a potential candidate gene. Specifically, low Syn3 expression was found to be genetically correlated with poor inhibitory control. We hypothesize that mice underexpressing Syn3 will exhibit poor reversal learning performance, signaling diminished inhibitory control. Methods. Male and female mice, aged 2-9 months, that were homozygous for a deletion of the Syn3 gene (Syn3 -/-), heterozygous (Syn3 -/+), or wild-type (Syn3 +/+) were tested in a reversal learning task. Results. Genotype was not found to significantly alter reversal learning performance in terms of trials to criterion, omissions, or feedback learning. Although genotype did not significantly alter feedback learning, sex did, with males outperforming females on their ability to use previous outcomes to optimize future choice behavior. Additionally, genotype was found to have a significant sex-dependent effect on the ability to complete a sustained, variable duration observing response, with homozygous Syn3 deleted males having the greatest difficulty completing the observing response under the longest time requirement. Taken together, these findings suggest that Syn3 may have a complex effect on impulsivity, altering waiting but not action impulsivity. Further investigation is required to clarify the full effects of underexpressing Syn3 on motivated and impulsive behaviors.

1 Department of Psychology (Behavioral Neuroscience), Binghamton University, Binghamton NY 13902.

16:00 *Jared Bagley*

41. Heritable variation in voluntary alcohol drinking in a genetically diverse inbred mouse panel
SPEAKER: *Jared Bagley*

ABSTRACT. J.R. Bagley1, L. S. Bailey1, J. D. Jentsch1,2

Alcohol consumption and associated subjective effects are individually variable, and genetic factors account for a substantial proportion of that variance. Most forward genetic research to date has originated from reduced complexity intercrosses with limited genetic and phenotypic variability. The collaborative cross (CC) recombinant inbred (RI) panel, its inbred founders and the diversity outbred (DO) populations, are a cutting-edge tool for genetic and genomic research in part because of their remarkable genetic diversity. For these reasons, we assessed both sexes of all eight inbred CC/DO founder strains for voluntary alcohol drinking (20% ethanol, 2-bottle choice with water) during the active phase of the mouse circadian cycle (in the dark). The CC/DO founder strains demonstrate substantial and statistically significant strain differences in terms of alcohol lick preference scores, as well as total alcohol intake, with the high drinking strain consuming up to 10 times the body-weight adjusted amount of alcohol relative to the low drinking strain. Furthermore, strain mean distributions suggest this is a quantitative trait in both sexes, however substantial sex effects are present in some strains with females drinking more alcohol. This work has established heritability of voluntary ethanol drinking in the CC founder strains and will serve as a foundation for further characterization of CC RI strains. This research is performed in the context of the Center for Systems Neurogenetics of Addiction (CSNA) project and will be integrated into expansive data sets that will allow for in depth analysis of genetic relationships of addiction related behaviors and gene expression.

1. State University of New York at Binghamton, Binghamton, New York 13902 USA, 2. The Jackson Laboratory, Bar Harbor, Maine 04609 USA

16:00 *Gang Chen*

43. Modeling the subtypes of depression on the basis of identification of syndrome patterns from two inbred mouse strains

SPEAKER: *Gang Chen*

ABSTRACT. Current treatment of major depression is not medically satisfying, partly due to the fact that the heterogeneity of the disease

was not clearly defined, and thus treatment with differential drugs was not plausible. Traditional Chinese Medicine (TCM) is a personalized medicine in which the diagnosis and treatment is based on syndrome pattern identification for a given condition. It has been found that most prevalent TCM syndrome patterns in depression clinically include Qi stagnation, the stress responsiveness-like syndrome and Qi deficiency, fatigue-like syndrome. Balb/cJ and 129/S1 both were depression-prone, but they appeared to have different phenotypes of syndrome. Here, to further validate the plausible subtypes of depression, we used the formula that specifically treats the TCM syndrome and characterized the associated gene networks. Balb/cJ and 129/S1 received same protocol of chronic mild stress. Stressed or non-stressed mice also received administration of Qi stagnation alleviating TCM drug Yueju, Qi tonifying drug Sijunzi. After stress, both strain of mice reduced the sucrose preference, indicating a depression-like condition. However, only Balb/cJ mice showed early-onset and lasting Qi deficiency phenotype, tested with grip strength of forelimbs and loaded swimming. In Balb/cJ mice, treatment with Sijunzi and Yueju both improved performance in sucrose preference, although Sijunzi but not Yueju improved Qi-deficiency; In 129/S1 mice, Yueju improved performance in both sucrose preference and Oi-stagnation, whereas neither was improved by Sijunzi. Finally, syndrome specific gene networks were dissected using RNA-seq on the hypothalamus in these mice to identify the molecular signatures of subtypes of depression. Together, this study suggests genetic predisposition of Balb/cJ and 129/S1 to different syndromes may be useful to model the subtypes of depression for better and more precise treatment of depression.

1, Center for Translational Systems Biology and Neuroscience, 2, key Laboratory of Integrative Biomedicine of Brain Diseases, Support: by the National Science Foundation of China (81673625) and the Priority Academic Program Development of Jiangsu Higher Education Institutions (PAPD).

16:00 *Anna Delprato*

45. Structure-function analysis of Slitrk proteins and the consequences of gene variants

SPEAKER: *Anna Delprato*

ABSTRACT. Anna Delprato1 and Wim Crusio2,3
1BioScience Project, Wakefield, MA, USA
2University of Bordeaux, Institut de

Neurosciences Cognitives et Intégratives d'Aquitaine (UMR 5287), Pessac Cedex, France 3CNRS, Institut de Neurosciences Cognitives et Intégratives d'Aquitaine (UMR 5287), Pessac Cedex, France.

Slitrk family members function in nervous system development and synapse dynamics. Several Slitrk gene variants have been linked to human neurological disorders such as Tourette syndrome, schizophrenia, autism, and obsessive compulsive disorder. Slitrks are transmembrane proteins that have extracellular N-terminal tandem leucine-rich repeats (LRR domains), a single transmembrane domain, and distinct C-terminal, cytoplasmic regions of varying length. In this study, we describe a Slitrk family-wide characterization using structural modeling and other in silico methods to predict the functional consequences of Slitrk gene variants on the fidelity of the proteins and their known interactions. These data will be used to forecast additional mutations that may be risk factors for neuropsychiatric disorders as well as identify conserved and divergent patterns of naturally-occurring variation. Results: For Slitrk1, 79% of the residues were modeled with greater than 90% accuracy which enables the visualization of most of the protein and the orientation of its domains with respect to one another. The prediction of a ligand binding site occurring in the 2nd LRR domain was also detected. For the other Slitrk proteins, the structural modeling was largely confined to the N-terminal region, including both LRR domains, and ranged from 58% (Slitrk3 and Slitrk5) to 66% (Slitrk4) of the residues modeled with greater than 90% accuracy. For Slitrk3 and Slitrk4, a portion of the C-terminal region was modeled with 80% confidence. Taken together, the structural modeling results for the 6 Slitrk family members, provides a foundation for rationalizing the gene variants based on the three dimensional protein structures.

16:00 *Cheryl Reed*

47. Selection for MA intake in mice with a non-functional trace amine-associated 1 receptor and examination of genetically correlated MA-related traits

SPEAKER: *Cheryl Reed*

ABSTRACT. C. Reed 1, H. Baba1, J. Erk1, T.J. Phillips1,2

The trace amine-associated receptor 1 gene (Taar1) impacts MA intake and other MA-related behaviors. A mutation in Taar1 (Taar1m1J) codes for a non-functional receptor, and Taar1m1J/m1J mice exhibit greater levels of MA intake than mice that possess at least 1 copy of the alternative allele (Taar1+). This mutation spontaneously arose in DBA/2J (D2J) mice and its impact has been studied on D2J and C57BL/6J strain backgrounds, which may not possess all genetic factors that could impact MA intake and other MA-related traits. Thus, the mutation was crossed onto heterogeneous stock-collaborative cross (HS-CC) mice that capture 90% of the genetic diversity in Mus musculus. Although Taar1m1J/m1J mice from this population consumed more MA than mice with at least 1 copy of Taar1+, some Taar1m1J/m1J mice had low MA intake, suggesting the presence of genetic modifiers of the Taar1m1J/m1J genotype effect. To identify these modifiers, selective breeding of high and low MA intake lines from D2JxHS-CC-Taar1m1J/m1J individuals is underway. Parent means (±SEM) of the S1 generation were 8.1±0.7 and 1.7±0.2 mg/kg, for the high and low MA intake lines, respectively. Response to selection was significant by S2; offspring means for MA intake were 5.2±0.4 vs. 4.2±0.3 mg/kg. Acute MA-induced stimulation and sensitization were significantly greater in Taar1m1J/m1J vs. Taar1+/+ mice of the originating population, but did not differ between the Taar1m1J/m1J S1 MA intake lines. If S2 mice differ for these traits, this will indicate that modifiers of the Taar1m1J/m1J effect for MA intake are also relevant to MA-induced locomotor stimulation and sensitization.

1Oregon Health & Science University, and 2Veterans Affairs Portland Health Care System Portland, OR, 97239 USA Supported by the Department of Veterans Affairs I01BX002106, NIH NIDA U01DA041579, NIH NIDA P50DA018165, and NIH NIAAA R24AA020245.

16:00 *Anjelica Rodriguez*

49. C. elegans as a Neurological Model for Duchenne Muscular Dystrophy

SPEAKER: *Anjelica Rodriguez*

ABSTRACT. Duchenne muscular dystrophy (DMD) is a lethal degenerative disease that affects 1 in 3,500 males. DMD is caused by mutations in the dystrophin gene, which is expressed in muscle and nervous tissue. About one-third of DMD patients show developmental

delays, among other neurological and muscular phenotypes. C. elegans is unique among animals used in DMD research in its ability to model not only the genetic insult, but also the behavioral, and cellular phenotypes observed in patients. To determine if Caenorhabditis elegans can also be used to model the neurological deficits of DMD, we ran dystrophic (dys-1) worms through a battery of neurological tests. Wild type animals are attracted to low concentrations of an attractant (1% diacetyl) but are repelled by high concentrations (100% diacetyl). Dystrophic worms detected and oriented normally towards low appetitive concentrations of a chemical cue. However, knockout mutants or worms with dystrophin specifically suppressed in nervous tissue also exhibited positive chemotaxis rather than being repelled by high concentrations. These findings suggest that lack of neuronal dys-1 is responsible for the ability of the animals to be repelled by noxious concentrations of diacetyl. The current experiments suggest that, in addition to modeling the muscular aspects of this disease, C. elegans may be useful to model the neurological impairments associated with DMD as well.

16:00 *Kiley Hughes*

51. Effectiveness of exercise in a nematode model of Duchenne Muscular Dystrophy: improvements in mobility following an endurance exercise regimen

SPEAKER: *Kiley Hughes*

ABSTRACT. Duchenne muscular dystrophy (DMD) is an x-linked degenerative disease that affects one out of every 3,500 males. This disease is produced by a mutation in the dystrophin gene that results in an absence of the dystrophin protein. The result is progressive muscle weakness, leading to loss of ambulation in late adolescence and premature death. Currently, there is no cure for DMD, and no well accepted exercise regime for DMD patients. Studies have suggested minimal improvement in strength following exercise, but the effect of type and duration of exercise has not been considered until now. Previously, we demonstrated that the dys-1(eg33) C. elegans mutant is the most faithful animal model for Duchenne muscular dystrophy. It models the disease genetically, behaviorally, and anatomically, without requiring the secondary (sensitizing) mutations common in other animal models of the disease. To determine the effects of exercise on muscle integrity we subjected dys-1 animals to control, strength, or

endurance exercise regiments of multiple durations. We find that while swimming did not increase longevity in dystrophic animals, it did temporarily improved their mobility. Our results represent the most complete assessment of exercise effect on dystrophic musculature to date, and demonstrate the complex interactions taking place between factors differentially affecting dystrophic musculature health, function, and longevity. Patients often face the daunting challenge of engaging in therapeutic treatments lacking experimental basis. It is our hope that our work will enable DMD patients and physicians to make more informed decisions regarding potential treatment approaches.

16:00 *Christiann Hill*

53. Behavioral characterization of a mouse model of Wolfram Syndrome 2
SPEAKER: *Christiann Hill*

ABSTRACT. Wolfram syndrome (WFS) is a rare autosomal recessive disorder characterized by diabetes mellitus and insipidus, progressive optic atrophy, and sensorineural deafness. An increased risk of psychiatric disorders has also been reported in WFS patients. There are two subtypes of WFS. Type 1 (WFS1) is caused by mutations in the WFS1 gene and type 2 (WFS2) is characterized by mutations in the CISD2 gene.

Existing mouse models for WFS exhibit similar phenotypes to those observed in WFS patients including diabetic nephropathy, metabolic disruptions and optic atrophy. We identified a mouse mutant, Chirper, with a spontaneous mutation in the Cisd2 gene. Chirper mice emit frequent sonic vocalizations that are audible to the human ear, exhibit rapid respiration and have decreased body size and weight compared to unaffected littermates. Unlike WFS patients, Chirper mice do not appear to be diabetic.

Although behavioral phenotypes have been characterized in Wfs1 knockout mice, similar studies have been lacking for Cisd2. We tested Chirper mice in a battery of behavioral assays that model phenotypes related to neurological and psychiatric disorders including anxiety, sensorimotor gating, stress response, social interaction, learning and memory. We observed that homozygous and heterozygous mutant mice exhibit increased stress response. Homozygous mutants also show deficits in spatial learning and memory compared to wildtype littermates. Future studies will assess auditory brainstem response to observe the cochlear function of Chirper mice.

Our data indicate that the Chirper mouse strain could be a useful model to investigate the neurological and psychiatric symptoms observed in WFS.

1Department of Genetics, 2Neurobiology Curriculum, 3Division of Pharmacotherapy and Experimental Therapeutics, Eshelman School of Pharmacy, University of North Carolina, Chapel Hill, NC.

16:00 *Qiu Ruan*

65. Triangulating on the neuroanatomical and cell biological mechanisms by which hnRNP H1 haploinsufficiency results in reduced methamphetamine-induced dopamine release and behavior

SPEAKER: *Qiu Ruan*

ABSTRACT. Heterogeneous nuclear ribonucleoprotein H1 (Hnrnph1) is a quantitative trait gene underlying reduced methamphetamine (MA) sensitivity. We subsequently showed that Hnrnph1 haploinsufficiency reduces MA-induced reward and reinforcement and MA-induced dopamine release in the nucleus accumbens. To explore the mechanism, we used immunoblotting and immunohistochemistry (IHC) to assay the level of tyrosine hydroxylase (TH), a precursor for dopamine synthesis and a marker of dopaminergic neurons, in the striatum and medial prefrontal cortex where the dopaminergic fibers terminate as well as the ventral midbrain where dopaminergic neuronal cell bodies originate. We hypothesized that a decrease in the number or innervation of dopaminergic neurons could underlie the neurobehavioral results. We found a small increase in TH staining in the dorsal striatum and nucleus accumbens. Immunoblot confirmed an increase in TH protein in the dorsal striatum and ventral midbrain. However, IHC of the ventral midbrain did not provide any evidence for an increase in the number of TH-positive neurons or in the amount of TH staining. Stereology of TH-positive puncta within the forebrain did not show any evidence for a difference in the number of dopaminergic fibers. These data suggest an alternate, drug-induced cell biological mechanism by which hnRNP H1 deletion affects MA neurobehavioral responding. We are investigating synaptic levels of the dopamine transporter (DAT), DAT function, and vesicular monoamine transporter expression. We are also using CLIP-seq and co-

immunoprecipitation combined with mass spectrometry to identify changes in hnRNP H1 RNA targets and protein complexes following MA treatment. These studies will provide insight for future mechanistic validation.

1 Laboratory of Addiction Genetics, Departments of Pharmacology and Experimental Therapeutics and Psychiatry, Boston University School of Medicine 2 NIGMS Biomolecular Pharmacology Training Program (GRANT), Boston University School of Medicine (GRANT) 3 Boston University Transformative Training Program in Addiction Science (TTPAS) Burroughs Wellcome Fund, (GRANT) 4 Department of Psychological and Brain Sciences, University of California, Santa Barbara 5 Ph.D. Program in Bioinformatics, Boston University 6 Computational Biomedicine, Boston University School of Medicine 7 Department of Anatomy and Neurobiology, Boston University School of Medicine

16:00 *Ryan Lusk*

55. Dynamic alternative polyadenylation events in the nucleus accumbens of alcohol consuming rats

SPEAKER: *Ryan Lusk*

ABSTRACT. Our expanding knowledge about the influence of alternative polyadenylation (APA) on health and disease has created a new avenue for genetics research. APA is a tightly regulated mechanism by which a single gene encodes RNA isoforms with different polyadenylation sites. The present study utilized publicly available RNA sequencing (RNA-Seq) data from the read sequence archive in a secondary analysis to investigate APA changes in the nucleus accumbens of alcohol consuming male outbred Wistar rats in two separate experiments. For the first experiment, animals were either given continuous access to ethanol or only received water. In the second experiment, all animals were given continuous access with the exception of a period of forced consumption via gavage of either ethanol or water in between continuous access periods. Differences in APA between the groups in each experiment were determined by analyzing the RNA-Seq data using the bioinformatics tool DaPars (Dynamic analysis of Alternative PolyAdenylation from RNA-seq). Several genes displayed differences in polyadenylation site usage between groups. Some of these genes, such as Gfap, Psma4, and Ndufv3, have previously been associated with alcohol consumption. For these genes, the results here may provide an alternative understanding for the effect of ethanol on their

expression. Other genes lacked any previous connection with alcohol, indicating possibly novel genetic factors were identified and, due to the nature of the analysis used for their identification, ethanol's effect on their expression and structure. Our results indicate that APA may improve our understanding of ethanol's effects on brain beyond expression differences.

1Department of Pharmaceutical Sciences, University of Colorado Anschutz Medical Campus, CO, USA

16:00 *Meejung Ko*

57. Behavioral characterization of β-arrestin-1 knockout mice in anxiety-like, seizure, and alcohol-drinking behavior

SPEAKER: *Meejung Ko*

ABSTRACT. ß-arrestin proteins (ß-arrestin-1 and 2) are ubiquitously expressed signaling molecules heavily implicated in the desensitization of G protein-coupled receptor signaling. While genetic knockout of a single ß-arrestin isoform has previously revealed distinct roles of these isoforms in drug-related behaviors, the majority of previous research has focused on the role of ß-arrestin-2. To better understand the role of ß-arrestin-1 in neurological behavior, we evaluated baseline and drug-related behavioral differences in ß-arrestin-1 wild-type (ßarr1 +/+), heterozygous (ßarr1 +/-), and knockout (ßarr1 -/-) male and female C57BL/6 mice. Compared with wild-type and heterozygous mice, ß-arrestin-1 knockout mice demonstrated higher baseline locomotor activity. Knockout mice also displayed higher Modified Racine Scale (MRS) scores for seizure behavior for delta-opioid agonist SNC80-induced seizures, suggesting that seizure threshold is reduced upon a global knockout of β-arrestin-1. For natural reward intake, no differences in sucrose preference were observed between genotypes or sexes. However, female ß-arrestin-1 knockout mice consumed more 10% alcohol than heterozygous females in a limited access, two-bottle choice model. In a binge-like 20% alcohol model, female ß-arrestin-1 knockout mice consumed significantly more alcohol than both heterozygous and wild-type females. A significant sex-effect was observed with females consuming more alcohol than males in both drinking models. Increased sensitivity to latency to loss of righting reflex was also observed in ß-arrestin-1 knockout mice, although no differences in duration of loss of righting reflex were observed. Overall, our findings suggest that expression of ß-arrestin-1 may be protective

against seizure, hyperlocomotion in both sexes, and alcohol consumption in female mice.

1Department of Medicinal Chemistry and Molecular Pharmacology, College of Pharmacy, Purdue University, West Lafayette, Indiana 47907, 2Purdue Institute for Integrative Neuroscience, Purdue University, West Lafayette, Indiana 47907, 3Interdisciplinary Life Science PhD Program (PULSe), Purdue University, West Lafayette, Indiana 47907

16:00 *Casey Gährs*

59. Transgenic crustaceans: adaptation of modern molecular technology to the study of neural function in decapods

SPEAKER: *Casey Gährs*

ABSTRACT. Modulatory transmitter systems are major contributors to nervous system plasticity and behavioral flexibility. To understand the mechanisms of neuromodulation, we must characterize modulator origins, targets, molecular pathways, and physiological responses. This is challenging using existing model organisms due to lack of identified neurons, access to cellular and circuit dynamics, or molecular tools. We seek to overcome these limitations by establishing transgenic labeling of neuronal structures, including modulatory transmitters and their receptors, in decapod crustaceans with characterized modulatory systems and neuronal physiology. We are using marbled crayfish - the first decapod crustaceans to have both genome and transcriptome sequenced. We have shown that offspring of this parthenogenetic species show little genetic variability, indicating a high probability for genetic manipulations to be carried to future generations1. As a first step toward transgenesis, we utilized our genome and transcriptome to perform BLAST analyses against related species to identify candidate genes of interest. We first developed a positive transgenesis marker by fusing GFP with the ubiquitously-activated promoter for actin-1 (Pactin-1), as actin-1 is broadly expressed early in development. Co-injection of Pactin-1::GFP will facilitate determination of transgenesis in subsequent injections of GFP-fusion products of neuronal genes. The marbled crayfish actin-1 transcript consists of 1132 bases with no introns; it is 92% homologous to cytoplasmic-type-actin-1 from Homarus americanus and 97.6% homologous to actin-1 from Penaeus monodon. We are now microinjecting Pactin-1::GFP plasmids into stage-1 oocytes to create the first transgenic line of crayfish.

1 Gutekunst, J., Andriantsoa, R., Falckenhayn, C., Hanna, K., Stein, W., Rasamy, J., and Lyko, F. Clonal genome evolution and rapid invasive spread of the marbled crayfish. Nature Ecology & Evolution. (2018). doi:10.1038/s41559-018-0467-9

16:00 *Chang-Chih Huang* and *San-Yuan Huang*

61. The SLC6A3 gene variants and reduction of dopamine transporter availability may have a role in susceptibility to alcohol use disorder

SPEAKER: *San-Yuan Huang*

ABSTRACT. **Chang-Chih Huang[1,2], San-Yuan Huang[1,3*]**

[1] Graduate Institute of Medical Sciences, National Defense Medical Center, Taipei, Taiwan[2] Department of Psychiatry, Buddhist Tzu Chi General Hospital, Taipei Branch, Taipei, Taiwan, R.O.C.[3] Department of Psychiatry, Tri-Service General Hospital, National Defense Medical Center, Taipei, Taiwan

Dopaminergic dysfunction is an important role in the pathogenesis of alcohol use disorder (AUD) and major depression (MD). Gene variants of the dopamine transporter (DAT) (also known as SLC6A3 gene) may influence expression of DAT. However, imaging studies on brain DAT availability in patient with AUD are limited, and the association of DAT availability with SLC6A3 gene variants in patients with AUD has not been analyzed. Hence, this study examined the relationship between brain DAT availability, SLC6A3 gene variants, cognitive function, and depressive symptoms in different subgroups of AUD. Single-photon emission tomography imaging with 99mTc-TRODAT-1 as a ligand was used to measure striatal DAT availability in 103 patients with AUD (55 pure AUD and 48 AUD/MD) and 42 age and sex-matched healthy volunteers. Each subject was genotyped for the DAT polymorphism, and the Wisconsin Card Sorting Test (WCST), Hamilton Depression Rating Scale (HDRS) were used to assess neurocognitive function and severity of depression prior to brain imaging. Patients with AUD showed a significant reduction of DAT availability in three brain regions ($p < 0.001$), and this reduction was more pronounced in the pure AUD patients compared to healthy controls. The A allele of rs6350 have a greater risk to develop AUD ($P < 0.05$), and AUD patient with A allele shows a significant reduction trend of DAT

availability in striatum region. The study results indicate that the DAT availability and SLC6A3 gene may have a role in susceptibility to AUD, and the SLC6A3 variants may influence the DAT availability.

16:00 *Carley Miller*

63. Age and genetic background influence initial nicotine sensitivity in C57BL/6J and DBA/2J mice

SPEAKER: *Carley Miller*

ABSTRACT. CN Miller1,2, MJ Caruso3, and HM Kamens2

The initial response to nicotine is an important predictor of subsequent abuse. Multiple factors may alter this response including genetic background and age of first use. Here we investigated the influence of age, genetic background, and their interaction on nicotine sensitivity. We then examined whether these factors influence the relationship between initial behavioral responses and voluntary nicotine consumption in adulthood in male C57BL/6J and DBA/2J mice. The initial response to nicotine was measured during early adolescence (PND 31), middle adolescence (PND 41), late adolescence (PND 51), or adulthood (PND 71). We measured nicotine-induced changes in locomotor activity and body temperature to assess behavioral and physiological sensitivity to an acute injection of nicotine. Thirty-five days after behavioral testing, all animals were assessed for voluntary oral nicotine consumption. Results demonstrated that adult C57BL/6J mice were more sensitive to nicotine-induced hypothermia compared to early adolescence. In DBA/2J mice age and treatment interacted such that early adolescents were insensitive to nicotine's hypothermic effects, but this response developed in later age. Locomotor sedation also differed by strain. Locomotor depression increased with age in C57BL/6J animals, but remained constant across time in DBA/2J mice. Finally, our data suggest that an acute nicotine exposure has long lasting effects on 100 ug/uL nicotine consumption in DBA/2J, but not C57BL/6J, mice. By understanding how age and genetic background influence initial behavioral responses to nicotine, we have a greater understanding of factors that promote nicotine abuse later in life.

1Department of Science, 2Department of Biobehavioral Health Penn State University Old Main, University Park, PA 16802, USA
3Department of Systems Pharmacology and Translational Therapeutics, Perelman School of

Medicine, University of Pennsylvania, Philadelphia, PA 19104, USA
Acknowledgements: The Broadhurst Career Development Professorship for the study of Health Promotion and Disease Prevention (HMK).

18:00-21:00 Session : Dinner on your own

Dinner on your own

2018 IBANGS MEETING: THE 20TH ANNUAL GENES, BRAIN & BEHAVIOR MEETING

WELCOME PROGRAM INDEXES

PROGRAM FOR SATURDAY, MAY 19TH

Days: previous day next day all days

View: session overview talk overview

08:30-09:30 Session Reg: Registration

Meeting Registration

LOCATION: Mayo Civic Center Grand Lobby West

09:00-09:10 Session 5: Mayo Clinic Welcome

CHAIR: *Doo-Sup Choi*

LOCATION: Mayo Civic Center 102/103

09:00 *Mark Frye*

Mayo Clinic Welcomes the 20th Annual Genes, Brain, and Behavior Meeting

SPEAKER: *Mark Frye*

09:10-11:10 Session 6: Symposium 2

Circadian roles in psychiatric and neural disorders

CHAIR: *Han Wang*

LOCATION: Mayo Civic Center 102/103

09:10 *Bethany Stahl*

Eaat2 functions in ensheathing glia to regulate of sleep and metabolic function

SPEAKER: *Bethany Stahl*

ABSTRACT. Bethany A. Stahl1,2#, Emilie Peco3,4#, Sejal Davla3,5, Kazuma Murakami1,2, Don J. van Meyel3,4*, and Alex C. Keene1,2*

Sleep is critical for many aspects of brain function, and chronic sleep disturbances are associated negative health outcomes including heart disease, diabetes and metabolic syndrome. Growing evidence suggests that glia contribute to diverse aspects of sleep regulation, including neuronal and metabolic homeostasis. Moreover, glial cells regulate synaptic function and neurotransmitter clearance, raising the possibility that glial cells affect sleep by modulating neurotransmitter signaling. The fruit fly, Drosophila melanogaster, provides a powerful system for interrogating genetic mechanisms underlying sleep regulation and function. At the molecular and cellular levels, sleep is highly conserved from mammals to flies, and powerful genetic tools allow for targeted manipulation of

genes in discrete cellular populations. We identify that disruption of the transporter Excitatory amino acid transporter 2 (Eaat2) in glia, but not neurons, increases sleep. In mammals, EAATs non-preferentially clear excitatory neurotransmitters from the extracellular region at synapses; however in flies, Eaat2 demonstrates selective high-affinity of aspartate and taurine. Eaat2 is exclusively expressed in ensheathing glia, and spatio-temporal manipulation reveals Eaat2 functions during adulthood in these cells to regulate sleep. The increased sleep observed in Eaat2 deficient flies is accompanied by reduced metabolic rate, revealing a novel role for ensheathing glia in metabolic regulation. Together, these findings reveal a novel wake-promoting role for Eaat2 and ensheathing glia in sleep regulation.

1. Jupiter Life Science Initiative, Florida Atlantic University, Jupiter, FL 33458, USA 2. Department of Biological Sciences, Florida Atlantic University, Jupiter, FL 33458, USA 3. Centre for Research in Neuroscience, Department of Neurology and Neurosurgery, McGill University, Montreal, Quebec, Canada 4. Research Institute of the McGill University Health Centre, Montreal, Quebec, Canada 5. McGill Integrated Program in Neuroscience McGill University, Montreal, Quebec, Canada # Denotes equal contributions * Correspondence to don.vanmeyel@mcgill.ca or keenea@fau.edu

09:40 *Han Wang*

Circadian roles of in psychiatric diseases – insights from zebrafish

SPEAKER: *Han Wang*

ABSTRACT. Previous studies showed that circadian abnormalities often lead to human psychiatric disorders. The underlying mechanisms, however, are far from certain. We found that zebrafish per1b mutants display hyperactive-, impulsive-, and attention deficit-like behaviors and low levels of dopamine, reminiscent of human Attention Deficit Hyperactivity Disorder (ADHD) patients. Per1b directly regulates dopamine-related genes monoamine oxidase and dopamine β hydroxylase, and acts through genes important for the development or maintenance of dopaminergic neurons to regulate their number and organization in the ventral diencephalic posterior tuberculum. Using a series of behavioral assays, we show that zebrafish per2 mutant fish display lower locomotor activities, more time staying in the center region of the tank and less time for social interactions, in

comparison with wild types, indicating a clear depression phenotype of the per2 mutant zebrafish. Quantitative RT-PCR shows that glucocorticoid receptor (gr) is significantly down-regulated in both the per2 mutant larvae and adult male fish brain. Luciferase reporter assays show that Per2 can enhance Rorαa-mediated expression of gr, and ChIP assays show that Per2 binds to the three RORE elements in the gr promoter, suggesting that Per2 positively regulates gr in zebrafish. Further, cortisol, and expression of pomc (proopiomelanocortin) and crh (corticotropin releasing hormone) are significantly up-regulated in the per2 mutant male fish, reminiscent of human depression patients with disruptive activities of the hypothalamic-pituitary-adrenal (HPA) axis. Taken together, our analyses of zebrafish circadian mutants shed light on new roles of the circadian clock in psychiatric disorders.

Center for Circadian Clocks, Soochow University, Suzhou 215123, Jiangsu, China Funding Support: the National Basic Research Program of China (973 Program) (2012CB947600) and the National Natural Science Foundation of China (NSFC) (31030062, 81070455, 81570171).

10:10 *Angela Ozburn*

NPAS2 Regulation of Anxiety-Like Behavior

SPEAKER: *Angela Ozburn*

ABSTRACT. Abnormal circadian rhythms and circadian genes are strongly associated with several psychiatric disorders. Neuronal PAS Domain Protein 2 (NPAS2) is a core component of the molecular clock that acts as a transcription factor and is highly expressed in reward- and stress-related brain regions, such as the striatum. However, the mechanism by which NPAS2 is involved in mood-related behaviors is unclear. We measured anxiety-like behaviors in mice with a null mutation in Npas2 and found these mice exhibit less anxiety-like behavior than wild-type littermates (in elevated plus maze, light/dark box and open field assays). We assessed the effects of acute and chronic stress on striatal Npas2 expression, and found that both stressors increased Npas2. Further characterization revealed Npas2 expression is restricted to Drd1-expressing neurons. Moreover, knockdown of Npas2 in the ventral striatum resulted in a similar reduction of anxiety-like behaviors as seen in the Npas2 mutant mouse. Using ChIP-Seq, we observed that NPAS2 has distinct temporal patterns of DNA binding in ventral striatum and identified novel binding sites for NPAS2 at genes known to be important for neurotransmission

(e.g. dopamine receptor Drd3, and several Gabra genes). We found that Npas2 mutant mice exhibit reduced sensitivity to the GABAa positive allosteric modulator, diazepam, and that knockdown of Npas2 reduced Gabra1 expression and response to diazepam in the ventral striatum. These results implicate Npas2 in the response to stress and the development of anxiety and provide functional evidence for the regulation of GABAergic neurotransmission by NPAS2 in the ventral striatum.

Speaker: Angela Ozburn Talk title: NPAS2 Regulation of Anxiety-Like Behavior Speaker institutional affiliations: 1. Portland Veterans Affairs Medical Center, Research and Development Service, Portland, OR, United States. 2. Department of Behavioral Neuroscience, Oregon Health and Science University, Portland, OR, United States.

Funding: NARSAD Young Investigator Award, Veterans Affairs Career Development Award 2: IK2 BX002488, and NIH grants: DA07290, AA020452 and AA010760.

10:40 *Gi Hoon Son*

Impact of circadian nuclear receptor NR1D1 on central dopaminergic system and its implications for bipolar disorder
SPEAKER: *Gi Hoon Son*

ABSTRACT. Most physiology and behaviors in mammal exhibit daily oscillations generated by an internal time-keeping system composed of interacting circadian clock proteins. Although circadian dysfunctions in affective disorders have been well recognized, the underlying molecular mechanisms still remain to be further clarified. Accumulating evidence suggests that molecular components of the circadian system are heavily involved in mood control. We have recently demonstrated that the circadian nuclear receptor NR1D1 (also known as REV-ERBα) constituting a stabilizing loop of the mammalian clock, has a key role in the cyclic regulation of central dopaminergic system as well as mood-related behaviors. Abrogation of Nr1d1 gene or pharmacological inhibition of NR1D1 activity induced bipolar mania-like phenotypes in mice such as hyperlocomotion, reduced depression/anxiety-like behaviors and increased aggression. In accordance with animal models, our findings in human subjects also suggest dysregulated NR1D1 function in bipolar disorder patients and imply its diagnostic applications. In conclusion, circadian molecular clock is directly and functionally linked with the midbrain

dopaminergic system, thereby having an impact on mood regulation and dysregulation.

1Department of Biomedical Sciences, 2Department of Legal Medicine, Korea University College of Medicine, Seoul, South Korea.

11:10-11:35 Session Break: AM Break

Break

LOCATION: Mayo Civic Center 104/105

11:35-12:35 Session 7: Young Investigator Awards

Young Investigator Awardees

CHAIRS: *Elissa Chesler* and *Lisa Tarantino*
LOCATION: Mayo Civic Center 102/103

11:35 *Catherine Kaczorowski*

Systems genetics reveals novel mechanisms underlying resilience to Alzheimer's Disease
SPEAKER: *Catherine Kaczorowski*

ABSTRACT. Sarah M. Neuner, BS[1,2], Sarah Heuer, BS[1], Timothy J. Hohman, PhD[3], Ryan Richholt, BS[4], Matthew J. Huentelman, PhD[4], Kristen M.S. O'Connell, PhD[2], **Catherine C. Kaczorowski**, PhD[1]

1. The Jackson Laboratory, Bar Harbor, Maine, United States
2. The Neuroscience Institute, University of Tennessee Health Science Center, Memphis, Tennessee, United States
3. Vanderbilt Memory and Alzheimer's Center, Department of Neurology, Vanderbilt University Medical Center, Nashville, Tennessee, United States
4. Neurogenomics Division, Translational Genomics Research Institute, Phoenix, Arizona, United States

In Alzheimer's disease (AD), the age of symptom onset is highly variable, with some patients exhibiting cognitive symptoms several decades later than predicted based on family history and genetic status. This variability cannot be explained by simple clinical or environmental factors, suggesting that additional genetic factors modify disease onset. The identification of modifier genes that confer resilience in high-risk patient populations would reveal new mechanisms and thus therapeutic strategies to delay disease onset. Disease-relevant protective variants are difficult to identify in human populations, primarily because asymptomatic individuals rarely enter the clinic. Mouse models represent an ideal complement to human studies, as they present many advantages, such as defined genotypes, early access to brain tissue, and precise environmental control. However, the traditional mouse models of AD have failed to translate into successful treatments that improve cognition in humans. Here, we employ a novel AD mouse genetic reference panel, designed to overcome some of the barriers presented by current AD models. Analyses of whole-genome RNA expression from the hippocampus using a variety of

bioinformatics approaches revealed key hub genes as specific targets that may be leveraged to promote resilience. We also identified a strong negative association between microglia transcriptional signatures and cognitive resilience to AD. Overall, work here introduces a humanized mouse population as an innovative and reproducible resource for the study of AD and identifies potential mechanisms and candidate genes that may be targeted in at-risk individuals to promote resilience. Ongoing and future work will investigate their utility as therapeutic targets.

12:05 *Karla Kaun*

Circuits and molecules driving cue-induced alcohol preference in Drosophila

SPEAKER: *Karla Kaun*

ABSTRACT. E Petruccelli1, KM Scaplen1, M Feyder1, N Ledru1, N Savory1, KR Kaun1

The ability to associate a rewarding stimulus with a sensory cue from the environment is critical for an animal's ability to find food and mates. Mapping the circuits in which these associations are formed, then initiate an output response provides a scaffold for understanding how experiences can influence decisions. Investigating the molecular makeup of this scaffold is key to understanding how memories for rewarding stimuli are remembered, and can be manipulated. The genetic accessibility afforded in Drosophila provides an ideal platform to understand how molecules act within reward circuits to form appetitive memories, and how drugs of abuse such as alcohol can manipulate these circuits to induce cravings. Here we describe a how memories for the intoxicating properties of alcohol are acquired and expressed through different mushroom body circuits. Intriguingly expression of these memories requires a remarkably complex multilevel circuit whereby dopamine directly, and indirectly via the mushroom body, modulates the activity of glutamatergic and cholinergic output neurons. We then reveal molecular changes that occur within mushroom body neurons to chance activity within these circuits required for memory retention. We show the importance of Scabrous/Notch signaling in formation of alcohol memory in this circuit, and demonstrate how alcohol can disrupt this highly conserved signaling cascade in the adult brain to induce transcriptional changes that persist in a reward memory circuit. Together this work provides a snapshot of how alcohol can affect the dynamic molecular and circuit mechanisms required for behavioral decisions.

12:35-13:30 Session L2: Lunch

Lunch

13:30-15:30 Session 8: Symposium 3

Rapid, non-genomic glucocorticoid signaling and its impact on behaviors

CHAIR: *Karl Clark*

LOCATION: Mayo Civic Center 102/103

13:30 *Jeffrey Tasker*

Acute stress-induced desensitization of CRH neurons to norepinephrine via rapid glucocorticoid regulation of adrenoreceptor trafficking

SPEAKER: *Jeffrey Tasker*

ABSTRACT. JG Tasker1,2, GW Weiss1, C Chen1, Z Jiang1

Noradrenergic afferent inputs stimulate corticotropin releasing hormone (CRH) neurons of the hypothalamic paraventricular nucleus (PVN) to activate the hypothalamic-pituitary-adrenal (HPA) axis in response to systemic stress. Glucocorticoids secreted in response to HPA activation feed back to the hypothalamus to inhibit HPA activation. Patch-clamp recordings of CRH neurons in brain slices revealed a rapid glucocorticoid-induced, endocytosis-dependent desensitization of alpha1 adrenoreceptors (AR1a) in PVN CRH neurons following acute stress. The glucocorticoid regulation of AR1a sensitivity is dependent on the nuclear glucocorticoid receptor (GR), as it is lost in a conditional GR knockout. Live-cell imaging of the AR1a in a hypothalamic cell line revealed that corticosterone does not itself cause internalization of the AR1a, but facilitates the norepinephrine (NE)-induced AR1a internalization by causing the accumulation of the AR1a in the late endosome. Thus, corticosterone increased AR1a interaction with the late endosomal marker Rab 11, but had no effect on AR1a interaction with the early endosomal marker Rab 5. Corticosterone also caused a reduced interaction of AR1a with the rapid recycling endosomal marker Rab4, which suggested that it caused redirection of the AR1a from the rapid recycling pathway into the late endosome. Therefore, glucocorticoids prevent rapid AR1a recycling to the membrane following ligand-mediated internalization by routing the receptor into the late endosome. These findings demonstrate a stress desensitization of CRH neurons to noradrenergic activation that likely contributes to the rapid negative feedback regulation of the HPA axis by glucocorticoids.

1Department of Cell and Molecular Biology, 2Tulane Brain Institute, Tulane University, New Orleans, Louisiana, USA

Funding support: National Institutes of Health MH066958.

14:00 *Paul Gasser*

Roles for organic cation transporter 3 (OCT3), a glucocorticoid-sensitive monoamine transporter, in stress-induced regulation of dopamine clearance and behavior

SPEAKER: *Paul Gasser*

ABSTRACT. Department of Biomedical Sciences, Charles E Kubly Mental Health Research Center, Marquette University, Milwaukee, WI

Glucocorticoid hormones exert powerful influences on behavior and brain function through a variety of cellular mechanisms. In addition to actions mediated by the intracellular glucocorticoid receptors, accumulating evidence suggests that many glucocorticoid effects, particularly those that appear with short latencies, involve interactions with other, membrane-associated, proteins. One such protein is organic cation transporter 3 (OCT3), a bidirectional transporter for norepinephrine, dopamine, serotonin and histamine. In contrast to the presynaptic norepinephrine (NET), dopamine (DAT) and serotonin transporters (SERT), OCT3 has higher capacity and lower affinity for substrates, is insensitive to inhibition by cocaine and antidepressants, and is inhibited directly by glucocorticoid hormones. Thus, OCT3 represents a stress hormone-sensitive monoamine transport mechanism that may mediate context-dependent effects of stress on monoaminergic neurotransmission and behavior. We have demonstrated that OCT3 is expressed widely throughout the brain, localized to plasma membranes in axonal, dendritic and astrocytic processes, and on neuronal somata, consistent with a role in regulating extracellular monoamines. Indeed, we have recently demonstrated that injection of rats with stress levels of corticosterone acutely decreases dopamine clearance in the nucleus accumbens via a DAT-independent mechanism likely involving the inhibition of OCT3-mediated transport. Treatment of rats with stress levels of corticosterone led to increases in the duration and amplitude of both naturally occurring dopamine transients and electrically evoked dopamine signals. We further provided evidence that, through this mechanism, corticosterone potentiates the actions of cocaine on dopamine

signaling and reinstatement of drug-seeking behavior in rats and mice.

14:30 *Henk Karst*

Rapid actions of corticosterone as function of the circadian rhythm and the implication for fear learning

SPEAKER: *Henk Karst*

ABSTRACT. The hypothalamic-pituitary-adrenal axis regulates plasma glucocorticoid levels. Under basal conditions, the synthesis and release of glucocorticoids occur in a circadian rhythm overarching ultradian pulses. Peak levels of glucocorticoids are seen at the start of the active phase. Limbic areas express high levels of corticosteroids (CORT) receptors and are affected by changes in CORT levels. We demonstrated earlier that repeated CORT application results in immediate (non-genomic) and long-lasting (genomic) changes in glutamate transmission in the basolateral amygdala (BLA), an area that regulates fear behavior. To study the effect of circadian fluctuations of CORT levels on glutamatergic neurotransmission in the BLA, we investigated spontaneous glutamate transmission in BLA slices of mice sacrificed at the start of the inactive and the start of the active phase. To mimic the ultradian glucocorticoid pulses, ‘inactive phase slices’ were exposed to pulses with increasing concentrations of CORT. Active phase slices were exposed to decreasing concentrations of CORT. Results showed that the basal miniature excitatory postsynaptic current (mEPSCs) frequency was increased in inactive phase slices with pulses of increasing concentration, while the basal mEPSC frequency was reduced in active phase slices exposed to the paradigm. Furthermore, tone-cued fear conditioning experiments showed that mice at the start of the inactive phase show more freezing behavior, a measure of fear, than mice at the start of the active phase. Mice treated with metyrapone, normally causing a reduction in the basal CORT level, showed more freezing behavior than controls, suggesting that fear learning depends on circadian variations in CORT levels.

15:00 *Han Lee*

Locomotor response to acute stressors requires hypothalamic-pituitary-interrenal axis activation and glucocorticoid receptors in zebrafish

SPEAKER: *Han Lee*

ABSTRACT. HB Leea,1, TL Schwabb,1, AN Sigafoosb, JL Gauerkeb, RG Krug, IIa, MR Serresb, DC Jacobsb, RP Cotterb, B Dasb,2, MO Petersenb, CL Dabyb, RM Urbanb, BC Berryb, and KJ Clarka,b,3

aNeurobiology of Disease program, Mayo Clinic Graduate School of Biomedical Sciences, 200 First St. SW, Rochester, MN 55905; and bDepartment of Biochemistry and Molecular Biology, Mayo Clinic, 221 Fourth Ave. SW, Rochester, MN 55902

When vertebrates face acute stressors, their bodies rapidly undergo a repertoire of physiological and behavioral adaptations, which is termed the stress response (SR). Rapid physiological changes in heart rates and blood sugar levels occur via the interaction of glucocorticoids and their cognate receptors following hypothalamic-pituitary-adrenal (HPA) axis activation. These physiological changes are observed within minutes of encountering a stressor and the rapid time domain rules out genomic responses that require gene expression changes. Although behavioral changes corresponding to physiological changes are commonly observed, it is not clearly understood to what extent HPA axis activation dictates adaptive behavior. We hypothesized that rapid locomotor response to acute stressors in zebrafish requires HPI axis activation. In teleost fish, interrenal cells (I) are functionally homologous to the adrenal gland cortical layer. We derived 8 frameshift mutants in genes involved in HPI axis function: two mutants in exon 2 of mc2r (adrenocorticotropic hormone receptor), two in each of exon 2 and exon 5 of nr3c1 (glucocorticoid receptor), and two in exon 2 of nr3c2 (mineralocorticoid receptor). Using larval zebrafish and mild environmental stressors, acute changes in salinity or light illumination, we demonstrate that rapid locomotor response to acute stressors requires a functioning HPI axis via the action of mc2r (adrenocorticotropic hormone receptor) and the canonical glucocorticoid receptor encoded by nr3c1 gene, but not mineralocorticoid receptor (nr3c2). Our rapid behavioral assay paradigm based on HPI axis biology may prove useful to screen for genetic and pharmacological modifiers of the HPA axis.

15:30-16:00 Session Break: PM Break

PM Break

LOCATION: Mayo Civic Center 104/105

16:00-18:00 Session P2: Poster Session II

Poster Session 2

LOCATION: Mayo Civic Center 104, 105, 106

16:00 *William Jons*

2. Investigating the Association of X Chromosome Genetic Variants with Sex-Specific Symptoms of Bipolar Disorder

SPEAKER: *William Jons*

ABSTRACT. William Jons1, Colin Colby1, Sue McElroy2, Mark Frye3, Joanna Biernacka1,3, Stacey Winham1

1Department of Health Sciences Research, Mayo Clinic, Rochester, MN, USA, 2Lindner Center of HOPE/University of Cincinnati, Cincinnati, Ohio, USA, 3Department of Psychiatry and Psychology, Mayo Clinic, Rochester, Minnesota, USA

Bipolar disorder (BD) affects 3% of both men and women in the US, but has sex-specific differences in its symptoms. We previously discovered that cycle acceleration, rapid cycling, and an increased severity of mood episodes are more prevalent among women. Using data from the Mayo Clinic Bipolar Disorder Biobank, we now investigate whether X chromosome genetic variants might be associated with sex-specific differences in symptoms or comorbid behaviors of BD (cycle acceleration, increased severity of mood episodes, rapid cycling, attempted suicide, substance use disorder, or binge eating behavior). Logistic regression was used to test the association of each X chromosome variant with each phenotype using three approaches: (1) an approach that ignores X chromosome inactivation (XCI), (2) an approach that allows for XCI at all locations, and (3) our new approach that uses biological data regarding which regions are subject to XCI to inform the statistical model selected. None of the approaches identified significant associations of the phenotypes with X chromosome variants. Results approaching the significance threshold with our approach ($p<3.3x10-6$) included the association of substance use disorder with rs787088 ($p= 8.77x10-6$), and association of binge eating behavior with rs5915827 ($p= 4.41x10-06$). Both of these variants are intergenic, the first being located near DDX3X, NYX, USP9X and a gene encoding the calcium signaling kinase (CASK) protein, and the second being located between PRKX and NLGN4X, which has been implicated in autism and possibly schizophrenia. Further work will be needed to validate our findings in a larger, independent sample.

Funding Support: NIH R25 GM075148, and Marriott Foundation, Bethesda, Maryland, USA

16:00 *Dushyant Mehra*

4. Dynamic Mapping of Genomic Architecture in vivo.

SPEAKER: *Dushyant Mehra*

ABSTRACT. Dushyant Mehra1*, Thomas Burghardt1,2*, Joshua D.Trzasko1, Han B. Lee2,, Tanya L. Schwab2, Wil A. Gendron2,, Brandon W. Simone2, Stephen E. Ekker2,, Tamas Ordog2, and Karl J. Clark2,

*Denotes Equal Contribution 1Department of Biomedical Engineering and Physiology, Mayo Clinic Rochester MN 2Department of Biochemistry and Molecular Biology, Mayo Clinic, Rochester, MN

Understanding the role that epigenetics plays in stress response is important for studying how stressors affect the body. DNA based epigenetic events often involve a change in chromatin conformation prior to an alteration in gene expression and capturing these dynamics can potentially provide a mechanism for understanding how different stressors affect gene regulation. Current conformation capture methods such as 3C, 4C, and Hi-C that are used map chromatin architecture only capture chromatin architecture in a snapshot and are difficult to use to characterize different chromatin conformational states over time. This makes it difficult for these methods to capture dynamic chromatin changes that can occur during transcription. Conformation capture based methods are further constrained by the location of the restriction enzyme sites and the depth of sequencing which currently limits inter loci distance to approximately 500 bp. Current dynamic imaging based methods are diffraction limited to approximately 500 bp as well due to conventional microscopy resolution limits. By combining a Crispr dCas9 protein fused to a photoactivatable GFP protein, a multiplex gRNA delivery system, and photoactivatable localization microscopy (PALM) for single molecule imaging, we are developing methods to quantitatively track dynamics and distances between individual loci at resolutions below conventional microscopy limits. This method can be used be used to determine how chromatin architecture changes in response to stress events and should be applicable to transparent model organisms such as C. Elegans and Zebrafish.

16:00 *Nicole Nelson*

6. A mechanism of public misunderstanding of animal behavior genetics research
SPEAKER: *Nicole Nelson*

ABSTRACT. Media coverage of animal behavior genetics research often misrepresents the complex etiology of behavioral disorders, typically portraying these disorders as arising from the effects of single genes rather than multiple genetic and environmental causes. This paper explores one mechanism through which these misrepresentations develop: the separation of knowledge about environmental contributors to behavior from knowledge about genetic contributors during the publication process. Using ethnographic research from animal behavior genetics laboratories, I describe how researchers informally acquire knowledge about how environmental factors (noise, smells, other stressors) alter mouse behavior, and how the designation of this knowledge as "methodological" or "tacit" limits its circulation outside of the laboratory. The limited circulation of environmental knowledge, as compared to the much wider circulation of genetic findings, contributes to a distorted public perception of disease etiology. As a counterfactual, I present media coverage of published instances of environmental knowledge (Crabbe, Wahlsten and Dudek's well-known 1999 paper on mouse behavior and interactions with the laboratory environment), demonstrating that alternative media narratives about behavioral disorders are possible. This research has important implications for the way that animal behavior geneticists communicate their findings publically, suggesting that more effort to circulate environmental knowledge is needed to counterbalance genetic knowledge.

16:00 *Alexandra Goetjen*

8. GABRA2 genetic variants and chromosome conformation in induced pluripotent stem cell-derived neural cultures
SPEAKER: *Alexandra Goetjen*

ABSTRACT. Approximately 8.5% of American adults are afflicted by either moderate or severe alcohol use disorder (AUD), defined as excessive alcohol use within the last twelve months that impedes the safety of oneself and others, while being unable to reduce one's drinking. The Collaborative Study on the Genetics of Alcoholism used linkage analysis to suggest, in European Americans, a significant association between alcohol dependence and a 140kb haplotype block in GABRA2. Synonymous SNP rs279858 tags this haplotype block, and has a

minor allele frequency of 0.45. In addition to AUD, rs279858 has been associated with a number of neuropsychiatric phenotypes including comorbid illicit drug use and childhood conduct disorder. Neuro-endophenotypes such as increased activation of the insular cortex and nucleus accumbens in reward anticipation and differential activation of the ventral tegmental area and medial frontal cortex in response to alcohol cues are also associated with this haplotype block. The chr4p12 locus codes for γ1, α2, α4, and β1 GABA receptor subunits; iPSC lines carrying the minor allele at rs279858 have reduced expression not only of GABRA2, but the other three genes within this cluster. Virtual chromatin conformation capture (4C) data supports this hypothesis of cis regulation of GABA gene expression. Identification and characterization of allele-specific variants involved in mediating long-range intrachromosomal interactions in this locus is a step forward in the process of elucidating genetic risk variants for AUD and subsequently developing more-specific therapies for those at increased genetic risk.

16:00 *Annie Park*

10. Alcohol-Intoxicated Flies Become Aggressive

SPEAKER: *Annie Park*

ABSTRACT. Alcohol-induced violence causes an immense social and economic burden worldwide. Despite the pervasiveness of this phenomenon, it is an understudied behavior and it's neurogenetic underpinnings are unknown. In this study, we describe a novel fly alcohol behavior; male Drosophila melanogaster become more aggressive after being exposed to a single low dose of alcohol. After this same low dose of alcohol female flies become more receptive to courtship, but do not exhibit alcohol-induced aggression. We predicted that alcohol was cross-potentiating the endogenous cVa (11-cis-vaccenyl acetate) sex-pheromone pathway, which resulted in potentiation of aggression and receptivity to courtship. The cVa sensitive olfactory receptor neurons (ORNs) express both FruM and Or67d. Here we show that both FruM and Or67d are necessary for alcohol-induced aggression. When flies are exposed to high levels of alcohol they have lower levels of FruM and have specifically downregulated FruM production in the Or67d expressing ORNs that project to the DA1 glomerulus. Flies previously treated with this higher dose of alcohol show reduced aggression. Taken together, these results suggest that

ethanol regulates FruM to produce changes in aggression in a dose dependent manner.

16:00 *Spencer Huggett*

12. Cocaine'omics: The Genetic and Neurological Underpinnings of Cocaine Use and Dependence – An Unorthodox Replication

SPEAKER: *Spencer Huggett*

ABSTRACT. Spencer B. Huggett1,2 & Michael C. Stallings1,2

1Department of Psychology and Neuroscience, University of Colorado 2Institute for Behavioral Genetics, University of Colorado Supported in part by: NIDA P60 DA11015

We investigated the genetic and transcriptional landscape of cocaine dependence (CD) and chronic cocaine use. We performed and integrated popular genome-wide and transcriptome-wide analyses using data from the largest genome wide association study (GWAS) on CD to date (Gelernter et al. 2014), 3,176 European Americans (EAs), and human post-mortem brain tissue from seven cocaine users and eight drug free controls. First, linkage disequilibrium (LD) score regression analyses was performed and detected a significant genomic heritability of 28% (s.e = 0.14) for CD and gene-based association tests found three novel genes underlying this heritability: the C1QL2, KCTD20 and STK38 genes. Tissue specificity analyses indicated robust enrichment in numerous brain regions, including the hippocampus, padj = 2.02e-06. Therefore using RNA-sequencing (RNA-seq) analyses we performed differential expression and weighted gene covariance network analyses (WGCNA) on post-mortem hippocampal brain tissue from Zhou et al. 2011. Differentially expressed genes or transcripts between chronic cocaine users versus drug free controls were enriched for genes associated with CD ($p < 0.05$), OR = 1.34, $p = 0.031$, and were used to find various potential therapeutic compounds for cocaine use/toxicity. Lastly, we found that KCTD20 was a central part of a hippocampal gene network strongly associated with cocaine use and thus, might be contributing to the genetic liability of CD by disrupting intricate gene networks in the brain. Overall, our study elucidates the biological architecture of cocaine use/dependence, proposes various novel therapeutic compounds for cocaine use and includes an alternative framework to validate/provide biological meaning to genome-wide findings.

16:00 *Thilini Wijesekera*

14. Drosophila NfKB Dif shows splice variant specificity in ethanol response
SPEAKER: *Thilini Wijesekera*

ABSTRACT. TP Wijesekera1 and NS Atkinson1

Chronic alcohol consumption is shown to affect conserved gene networks in the human brain; one such network being the innate immune system. The innate immune system in Drosophila melanogaster consists of the Toll and IMD pathways, which culminate in NfKB proteins. The NfKB proteins connected to the Toll pathway are Dorsal and Dif (Dorsal like immunity factor). The functioning of the Toll pathway, including Dorsal and Dif, in ethanol response was previously demonstrated in flies. Dif is expressed in alternative splice isoforms. Based on the presence or absence of a nuclear localization signal in the genomic sequence, it is hypothesized that Dif A is nuclear and Dif B is non-nuclear. This study demonstrates that the functioning of Dif on ethanol response is splice variant specific. The function of the two isoforms was tested using fly lines that are Dif null, while engineered to express a specific (A or B) isoform. Flies deficient in Dif B show an increase in sensitivity to ethanol induced sedation as indicated by a significant decrease in KD50 (time for 50% sedation). Absence of the Dif A isoform does not change sensitivity to ethanol. Additionally, we demonstrate the significance of the Dif splice isoforms in other adult behaviors such as male courtship, circadian rhythm and sleep. Cellular localization of Dif B was seen to be non-nuclear. We document the expression of Dif B in the mushroom bodies and the antennal lobes of the adult brain, further strengthening the significance of the isoform in ethanol response.

1Department of Neuroscience and Waggoner Center for Alcohol and Addiction Research, University of Texas at Austin, Austin, TX, USA.

16:00 *Margot Cousin*

16. First cohort of patients with novel KCNK9 variants: gain and loss of function cause Birk-Barel mental retardation dysmorphism syndrome
SPEAKER: *Margot Cousin*

ABSTRACT. MA Cousin1, 2, †, EL Veale3, †, MT Zimmermann4, 5, †, M Bialer6, MR Bekheirnia7, C Boelman8, G Douglas9, D Doummar10, C Gilissen11, J Juusola9, B Keren12, T Kleefstra11,

TM Kruisselbrink2, 13, MS Leduc7, K Machol7, S Mahida14, C Mignot12, J McLaughlin6, V Narayanan15, C Nava12, R Pfundt11, K Ramsey15, F Scaglia7, C Smith-Hicks14, AS Trentesaux16, R Willaert9, N Zadeh17, 18, D Babovic-Vuksanovic2, 13, RA Urrutia5, 19, A Mathie3, EW Klee2, 11, 13

The paternally imprinted KCNK9 gene encodes a two-pore-domain potassium ion transporter expressed primarily in the brain. A variant (G236R) in KCNK9 has been reported to cause Birk-Barel mental retardation dysmorphism syndrome (BBMRDS) in several families, but no additional variants have been reported to date. Here, we describe a cohort of 15 affected individuals from 11 families, one with the G236R variant and 10 affected by 9 additional novel KCNK9 alterations. The phenotypes in all patients are generally consistent with BBMRDS. Protein modeling showed that variants affect protein structure, dynamics, and K+ ion distribution. Compared to WT, a subset of variants caused an increased probability of K+ near the channel pore and were associated with more K+ transport events. This is in stark contrast to the decrease seen with G236R, suggesting distinct channel dysregulation by these novel variants. Patient variant characterization using electrophysiological techniques was consistent with the modeling results. Four of five variants assessed by these methods caused a gain of conductance compared to WT. All five variants were outwardly rectifying with reversal potentials close to the WT equilibrium potential. This is in contrast to the severely reduced current amplitude and pronounced inward rectification seen with G236R. We describe a cohort of patients with novel KCNK9 variants causing BBMRDS and with functional impact of these variants that is distinct from the classical G236R. Based on this observation, treatment with channel agonists, with reported positive outcomes in patients with the G236R variant, may not benefit patients with gain-of-function variants.

1Department of Health Sciences Research, Mayo Clinic, Rochester, MN, USA, 2Center for Individualized Medicine, Mayo Clinic, Rochester, MN, USA, 3Medway School of Pharmacy, University of Kent and University of Greenwich, Central Avenue, Chatham Maritime, Kent, ME4 4TB, United Kingdom, 4Clinical and Translational Sciences Institute, Medical College of Wisconsin, Milwaukee, WI 53226, USA, 5Bioinformatics Research and Development Laboratory, Genomics Sciences and Precision Medicine Center, Medical College of Wisconsin,

Milwaukee, WI 53226, USA, 6Division of Medical Genetics, Northwell Health, Manhasset, NY 11030, USA, 7Department of Molecular and Human Genetics, Baylor College of Medicine, Houston, TX, USA, 8Division of Neurology, BC Children's Hospital, Vancouver, British Columbia, Canada, 9GeneDx, 207 Perry Parkway, Gaithersburg, MD 20877, USA, 10APHP, Department of Neuropediatrics, National Reference Center for Neurogenetic Disorders, Hôpital Armand-Trousseau, GHUEP, Paris, France, 11Department of Human Genetics, Donders Centre for Neuroscience, Radboud University Medical Center, Nijmegen, the Netherlands, 12APHP, Département de Génétique et Centre de Référence Déficiences Intellectuelles de Causes Rares, Hôpital de la Pitié-Salpêtrière, Assistance Publique - Hôpitaux de Paris, Paris 75651, France, 13Department of Clinical Genomics, Mayo Clinic, Rochester, MN, USA, 14Department of Neurogenetics, Kennedy Krieger Institute, Baltimore, MD, USA, 15Center for Rare Childhood Disorders, Translational Genomics Research Institute, Phoenix, AZ, USA, 16Service de Néonatologie, CHU de Caen, France, 17Genetics Center, Orange, CA, USA, 18Division of Medical Genetics, CHOC Children's Hospital, Orange, CA, USA, 19Department of Surgery, Medical College of Wisconsin, Milwaukee, WI 53226, USA, †Authors contributed equally. Funding Support: Center for Individualized Medicine, Mayo Clinic, Rochester, MN, USA.

16:00 *Adeeb Sebai*

18. The Genetic Landscape of Familial Congenital Hydrocephalus
SPEAKER: *Adeeb Sebai*

ABSTRACT. Ranad Shaheen, PhD,1 Mohammed Adeeb Sebai, MD,1 Nisha Patel, PhD,1 Nour Ewida, BSc,1 Wesam Kurdi, MD,2 Ikhlass Altweijri, MD,3 Sameera Sogaty, MD,4 Elham A lmardaw i, MD,2 Mohammed Zain Seidahmed, MD,5 Abdulrahman Alnemri, MD,6 Sateesh Madirevula, PhD, 1Niema Ibrahim, BSc, 1Firdous Abdulwahab, BSc,1Mais Hashem, BSc,1Tarfa Al-Sheddi, BSc,1Rana Alomar, MSc,1Eman Alobeid, BSc,1Bahauddin Sallout, MD,7Badi AlB aqawi, MD,7Wajeih AlAali, MD,7,8Nouf Ajaji, MD,7Harry Lesmana, MD,9Robert J. Hopkin, MD,9Lucie Dupuis, MD,10Roberto Mendoza-Londono, MD,10Hadeel Al Rukban, MD,10Grace Yoon, MD,10,11Eissa Faqeih, MD,12andFowzan S. Alkuraya, MD1,13,14

Objective: Congenital hydrocephalus is an important birth defect, the genetics of which remains incompletely under-stood. To date, only 4 genes are known to cause Mendelian diseases in which congenital hydrocephalus is the main orsole clinical feature, 2 X-linked (L1CAM and AP1S2) and 2 autosomal recessive (CCDC88C and MPDZ). In this study, we aimed to determine the genetic etiology of familial congenital hydrocephalus with the assumption that these cases repre-sent Mendelian forms of the disease. Methods: Exome sequencing combined, where applicable, with positional mapping. Results: We identified a likely causal mutation in the majority of these families (21 of 27, 78%), spanning 16 genes,none of which is X-linked. Ciliopathies and dystroglycanopathies were the most common etiologies of congenital hydrocephalus in our cohort (19% and 26%, respectively). In 1 family with 4 affected members, we identified a homo-zygous truncating variant in EML1, which we propose as a novel cause of congenital hydrocephalus in addition to its suggested role in cortical malformation. Similarly, we show that recessive mutations in WDR81, previously linked to cerebellar ataxia, mental retardation, and disequilibrium syndrome 2, cause severe congenital hydrocephalus. Fur-thermore, we confirm the previously reported candidacy of MPDZ by presenting a phenotypic spectrum of congenital hydrocephalus associated with 5 recessive alleles. Interpretation: Our study highlights the importance of recessive mutations in familial congenital hydrocephalus andexpands the locus heterogeneity of this condition.

1. Departments of Genetics and 2. Department of Obstetrics and Gynecology, King Faisal Specialist Hospital and Research Center, Riyadh, SaudiArabia; 3. Department of Surgery, College of Medicine, King Saud University, Riyadh, Saudi Arabia; 4. Department of Medical Genetics, King FahadGeneral Hospital, Jeddah, Saudi Arabia; 5. Department of Pediatrics, Security Forces Hospital, Riyadh, Saudi Arabia; 6. Department of Pediatrics, Collegeof Medicine, King Saud University, Riyadh, Saudi Arabia; 7. Maternal Fetal Medicine Department, Women's Specialized Hospital, King Fahad MedicalCity, Riyadh, Saudi Arabia; 8. Bnoon Medical Center, Riyadh, Saudi Arabia; 9. Division of Human Genetics, Cincinnati Children's Hospital Medical Center,Cincinnati, OH; 10. Division of Clinical and Metabolic Genetics and 11. Neurology, Department of Paediatrics, Hospital for Sick Children, University ofToronto, Toronto, Ontario, Canada; 12. Department of Pediatric Subspecialties, Children's Hospital, King Fahad

Medical City, Riyadh, Saudi Arabia; 13. Department of Anatomy and Cell Biology, College of Medicine, Alfaisal University, Riyadh, Saudi Arabia; and 14. Saudi Human Genome Program, KingAbdulaziz City for Science and Technology, Riyadh, Saudi Arabia

16:00 *Kerry Preston*

20. A relatively enriched environment delays binge onset but does not affect binge size
SPEAKER: *Kerry Preston*

ABSTRACT. KE Preston1, RL Corwin2, JO Bader1, SL Crimmins1.

The goal of this study was to use the limited access (LA) model of binge eating to evaluate the impact of relatively enriched housing conditions on binge-type behavior in rats. In the LA model, bingeing is established when a group with intermittent access to a palatable food consumes significantly more of the food during the access period than a group with daily access. Historically, the LA model is used in nonenriched housing conditions. However, from an animal care perspective and with respect to the etiology of binge eating, it is important to determine how enrichment affects binge eating. Rats were divided into four groups: Control-Enriched, Control-Nonenriched, Intermittent-Enriched, and Intermittent-Nonenriched. Control groups received daily 30-minute access to vegetable shortening, while intermittent groups received 30-minute access to shortening on Monday, Wednesday, and Friday only. Enriched groups were housed with bedding, nesting material, toys, and a solid floor, while nonenriched groups were housed on wire flooring with no additions to cages. Conditions were reversed for intermittent groups halfway through the study. Results indicate that a relatively enriched environment delays binge onset, but does not significantly alter the amount of fat consumed during binge sessions or reverse established binge behavior. Intermittent access to vegetable shortening induces greater consumption of shortening than does daily access regardless of enrichment condition; however, rapid establishment of enduring binge-type eating appears to require austerity in housing conditions. Also, it is not likely that the level of enrichment provided in this study can prevent or reverse binge-type eating in rats.

1Dept of Clinical Investigation, William Beaumont Army Medical Center, El Paso, TX, USA

2Nutritional Sciences Dept, The Pennsylvania State University, University Park, PA, USA.
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16:00 *Ryan Cotter*

22. SSRIs and serotonin transporter impact on bone: How zebrafish inform personalized medicine

SPEAKER: *Ryan Cotter*

ABSTRACT. RP Cotter, HB Lee, AN Sigafoos, BL Clarke, MI Lapid, KJ Clark

Selective serotonin reuptake inhibitors (SSRIs) are commonly used antidepressant medications that regulate mood and alleviate depression by increasing amounts of circulating serotonin in the synaptic cleft. The serotonin transporter, encoded by SLC6A4 gene in humans, recycles serotonin back to the axonal boutons of pre-synaptic neurons. However, pleiotropic roles that serotonin plays in different brain tissues are largely unknown. Moreover, we are only now becoming aware of potential long-term adverse effects of SSRIs involving other tissues than the nervous system since the first prescribed SSRI, fluoxetine, was released in 1987. Recent studies proposed a link between long-term exposure to SSRIs and decreased bone mineral density and increased fracture risk. In addition, the short form of the polymorphic region in the serotonin transporter gene (SLC6A4) is implicated in decreased bone density. Importantly, the relationship between the functions of SLC6A4, SSRIs, and bone have not been systematically investigated. We hypothesize that SLC6A4 regulates bone density by modulating osteoblast formation and/or differentiation. Using zebrafish, we are generating revertible slc6a4a mutant alleles by targeted integration of a gene-break cassette to interrupt gene function and test the role of slc6a4a on bone mineralization. We will also test the effect of commonly prescribed SSRIs on bone mineralization in conjunction with genetic manipulations. This project may demonstrate a link between slc6a4a levels and the propensity of individual SSRIs to impact bone metabolism that could inform patients and clinicians as they weigh the benefits and risks of SSRIs.

16:00 *Kyle Schaefbauer*

24. Neuronal Expression Represented in The Gene Breaking Transposon Library
SPEAKER: *Kyle Schaefbauer*

ABSTRACT. Kyle Schaefbauer, Hirotaka Ata, Mark Wishman, Karl J Clark and Stephen C. Ekker

The gene break transposon (GBT) cassette is a unique insertional mutagen that reports endogenous gene expression including protein and RNA, causes a 99+% knockdown of the tagged gene, and is revertible by Cre recombinase. We have generated a library of 1000+ GBT alleles, searchable by gene name and expression pattern (www.zfishbook.org). A number of lines show expression in diverse neural circuits including known and novel patterns. In addition, a subset of GBT alleles are in nuclearly encoded mitochondrial genes, loci that play critical roles in neurodegenerative diseases through unknown or understudied mechanisms. The GBT system presents a unique opportunity to study genes that are associated with Parkinson's Disease (PD) and other neurodegenerative disorders. Notably, of the over 10 mitochondrial GBT loci we have identified to date, GBT 599 is integrated in the mitochondrial calcium uniporter (mcu) gene. Finally, the recently developed VALET gene editing-based approach enables the targeted integration of this gene break insertional cassette. We will report our efforts to generate engineered GBT alleles in pink1 and park2. Together, these new genetic lines represent new revertible zebrafish models suitable for mechanistic analyses and high-throughput drug screening applications of PD and related disorders.

International Protein Trap Consortium Mayo Clinic, Rochester, MN 55906

16:00 *Elizabeth Litkowski*

26. Differential Brain Gene Co-expression Between Saline and Ethanol Treated Recombinant Inbred Mice
SPEAKER: *Elizabeth Litkowski*

ABSTRACT. EM Litkowski1, RA Radcliffe2, WJ Shi3, LM Saba1,2, K Kechris1

To increase our understanding of genetic effects on acute ethanol sensitivity as it relates to alcohol consumption, we examined the differential brain co-expression of genes between saline and ethanol treated LXS recombinant inbred mice. Differential co-expression can be defined as a

change in the relationship between two genes after a perturbation, such as exposure to ethanol. We estimated posterior probabilities for differential co-expression by employing a mixture model that bins gene pairs based on their relationship before and after the perturbation, e.g., no correlation between the two genes in the saline group and a positive correlation between genes in the ethanol group. Preliminary results unveiled approximately 450 unique genes in co-expressed gene pairs after ethanol exposure that had no relationship in the saline group. Furthermore, approximately 900 unique genes were in co-expressed gene pairs in the saline group that had no relationship after ethanol exposure. Gene enrichment analysis applied to these gene classes highlights several pathways known to have roles in ethanol exposure and alcohol use disorders, such as dopaminergic-synapse, alcoholism, thyrotropin-releasing hormone receptor signaling, and endogenous cannabinoid signaling. Performing differential co-expression analysis allowed us to distinguish between treatment groups, furthering our knowledge of the genetics of acute ethanol sensitivity and how that might relate to alcohol consumption.

1Department of Biostatistics and Informatics, Colorado School of Public Health, 2 Department of Pharmaceutical Sciences, Skaggs School of Pharmacy and Pharmaceutical Sciences, 3Computational Bioscience Program, School of Medicine, University of Colorado, Anschutz Medical Campus, 13001 E. 17th Place, Aurora, CO 80045, Funding Support: NIH grant R01-AA016957, NIH grant R01-AA021131, NIH grant T15LM009451

16:00 *Ashley Sigafoos*

28. Functional testing of patient DNA variants implicated in neuronal phenotypes
SPEAKER: *Ashley Sigafoos*

ABSTRACT. Ashley N. Sigafoos1,2, Nicole J. Boczek1, Han B. Lee2, Tanya L. Schwab 1,2 , Patrick R. Blackburn1, Margot A. Cousin1, Eric W. Klee1, and Karl J. Clark1,2

1Center for Individualized Medicine and 2Department of Biochemistry and Molecular Biology, Mayo Clinic, Rochester, MN USA

Precision medicine is conceptualized as a medical care and research model to provide individualized and tailored care for each patient

based on one's genetic make-up. By far, the most common phenotypic category of undiagnosed diseases is neurologic disorders, accounting for 53% of all accepted patients to the NIH. Arguably, neuronal phenotypes are the most difficult to study since the tissue is not easily accessible. Our Translational Genomics Core (TGC) within the Center for Individualized Medicine at Mayo Clinic have been developing functional and behavioral assay suites using models including zebrafish. The overarching goal and hypothesis of TGC is that we enhance the diagnosis and understanding of neuronal phenotypes of rare disorders via functional characterization and validation of the roles that Variants of Unknown Significance (VUS) play. Leveraging the high degree of conservation of gene functions to humans and rapid development of the fish model, we are characterizing the functions of VUSs of human GABBR2, SLC2A10, SOX5, and DNM1L. We revealed that zebrafish gabbr2 knockouts show profoundly decreased locomotor response while overexpression of human GABBR2 in transgenic fish lines leads to moderately decreased locomotion to light change stimulation. Additional functional work is required to understand how patient VUSs influence the observed phenotypes. We envision our pioneering functional and behavioral work will both aid in patient diagnosis and provide options for patient care and wellbeing.

16:00 *Elena Jazin*

30. Novel Y chromosome encoded long non-coding RNAs (Y-lncs) expressed in human male CNS during early development
SPEAKER: *Elena Jazin*

ABSTRACT. M M Johansson1, P Pottmeier1, P Suciu1, T Ahmad1, A Zaghlool2, J Halvardson2, E Darj3, 4, L Feuk2, C Peuckert1, 5 and Elena Jazin1*

1Department of Organismal Biology, EBC, Uppsala University, Sweden 2Department of Immunology, Genetics and Pathology, Science for Life Laboratory, Uppsala University, Sweden 3Department of Women's and Children's Health, International Maternal and Child Health (IMCH), Uppsala University, Sweden 4Department of Public Health and General Practice, Norwegian University of Science and Technology, Trondheim, Norway 5 Department of Molecular Biology, Stockholm University, Stockholm, Sweden

Global microarray gene expression analyses demonstrated differences in female and male

embryos during neurodevelopment. In particular, before sexual maturation of the gonads, the differences seem to concentrate on the expression of genes encoded on the X and Y chromosomes. To investigate the genome-wide differences in expression during this early developmental window, we combined high resolution RNA sequencing with qPCR to analyse brain samples from human embryos during the first trimester of development. Our results show that the largest biased group consisted of genes encoded on the sex chromosomes and the majority of all differentially expressed genes were male-biased. 10 out of 13 expressed gametolog pairs showed unbalanced gene dosage and as a consequence, a male biased expression. Among X-chromosome genes, three genes had higher expression in female embryos but a balanced gene dosage due to the Y gametolog expression. In addition, we found 6 novel non-annotated long non-coding RNAs on the Y-chromosome with conserved expression patterns in new-born chimpanzees. The tissue specific and time restricted expression of these long non-coding RNAs strongly suggests important functions during central nervous system development in human males.

*Uppsala University, Department of Organism Biology, Unit for Evolution and Development, Uppsala, Sweden. Funding support: Swedish Research Foundation, Grant name: Sex determination factors in the brain encoded in the Y chromosome (Project number K2012-61X-22089-01-3)

16:00 *Chris Stojanik*

32. An Investigation of Genes Involved in the Regulation of Novel Alcohol Behaviors (Alcohol Induced Aggression and Courtship Receptivity) in Drosophila melanogaster
SPEAKER: *Chris Stojanik*

ABSTRACT. A Park, C Stojanik, T Tran, N Atkinson Department of Neuroscience, University of Texas at Austin

Despite the pervasiveness of alcohol-induced violence, little is understood about what genes contributes to this behavior. Previous work has identified serotonin and monoamine oxidases as being important for regulating alcohol-induced aggression. However, there are extremely few studies investigating novel genes that contribute to alcohol-induced aggression, likely due to the lack of model organisms with robust genetic toolsets. Using Drosophila melanogaster we investigate the role of NPF (Neuropeptide F) and

TH (Tyrosine Hydroxylase) in mediating alcohol-induced aggression. NPF is a neuropeptide that has previously been implicated in sexually dimorphic alcohol behaviors. Tyrosine Hydroxylase, which is an enzyme necessary for dopamine synthesis has also been previously implicated in alcohol behaviors. Both of these genes have mammalian orthologues with nearly identical pathways and functions. Investigating the role of these genes in Drosophila will enable us to better understand the unknown basis of how alcohol-induced aggression is regulated in humans.

16:00 *Rajani Maiya*

34. Differential regulation of excessive alcohol consumption by the transcriptional regulator LMO4

SPEAKER: *Rajani Maiya*

ABSTRACT. R. Maiya1,2, A. Beckham1,2, G.N. Tiwari1, S.P. Farris1, R.D. Mayfield1, and R.O. Messing1,2.

Repeated alcohol exposure leads to changes in gene expression that are thought to underlie the transition from moderate to excessive drinking. Gene expression profiling studies have identified a multitude of alcohol-responsive gene networks, but the mechanisms by which these networks are mobilized to a neuroadaptive response are not well understood. One mechanism could involve alcohol regulation of transcriptional co-regulators that bind and modulate the activity of several transcription factors. Our results indicate that the transcriptional regulator LMO4 is one such candidate regulator. Mice harboring a gene trap insertion at the Lmo4 (Lmo4gt/+) locus consumed significantly more and showed enhanced preference for alcohol in a 24-h intermittent access procedure. shRNA-mediated knockdown of LMO4 in the NAc significantly enhanced whereas knockdown in the BLA decreased alcohol consumption and preference and reduced conditioned place preference to alcohol. To ascertain the molecular mechanisms that underlie the contrasting effects of LMO4 knockdown in the BLA and NAc, we performed unbiased transcriptome profiling of these two brain regions in WT and Lmo4gt/+ mice using RNASeq. We identified approximately 1000 genes that were differentially expressed in both brain regions of which only 48 were common between the two brain regions. We validated several of these differentially expressed genes by quantitative real time PCR. Weighted gene co-

expression network analysis implicated genes related to the extracellular matrix in the BLA and genes related to phosphatidylinositol-3,4,5-triphosphate binding in the NAc as primary transcriptional targets of LMO4. Future experiments will determine the effects of perturbation of these networks on alcohol consumption.

1Waggoner Center for Alcohol and Addiction Research, 2Department of Neuroscience, The University of Texas at Austin, Austin, TX, 78712

16:00 *Katheryn Wininger*

36. The Role of Astrocytic Glutamate Transporter 1 (GLT1) in Psychiatric Disorders
SPEAKER: *Katheryn Wininger*

ABSTRACT. Authors: Katheryn M Wininger1, Yun Fang Jia2, Lee Peyton2, Seungwoo Kang2, Daniel Lindberg1, Doo-Sup Choi1,2,3

1Neurobiology of Disease Program, Mayo Clinic Graduate School of Biomedical Sciences, Rochester, MN 55905, USA. 2Department of Molecular Pharmacology and Experimental Therapeutics, Mayo Graduate School of Biomedical Sciences, Rochester, MN 55905, USA.3Department of Psychiatry and Psychology, Mayo Clinic, Rochester, MN 55905, USA.

Glutamate transporter 1 (GLT1) is responsible for roughly 90% of glutamate clearance from the synaptic environment. Consequently, dysregulation of GLT1 results in hyper or hypo-glutamatergic states, which have been implicated in many psychiatric disorders such as anxiety, depression, addiction, and post-traumatic stress disorders. Although GLT1 is expressed by neurons and plays an important role in regulating neural physiology and glutamatergic signaling, numerous studies have suggested that GLT1 is predominantly expressed by astrocytes. Therefore, to investigate the astrocytic function of GLT1, we generated a line of astrocyte-specific GLT1 knockout mice by crossing floxed GLT1 mice with astrocyte-specific protein GFAP (glial fibrillary acidic protein) driven Cre recombinase expressing transgenic mice. As anticipated, these animals displayed marked deficiencies in GLT1 mRNA and protein expression in brain regions associated with psychiatric disorders, including the medial prefrontal cortex, nucleus accumbens, caudate putamen, and hippocampus. Importantly, mice lacking astrocytic GLT1 exhibited significantly reduced depressive and anxiety-like

behavior, as well as decreased freezing behavior to both context and cue in fear conditioning. Interestingly, astrocytic GLT1 deficient mice displayed reduced behavioral sensitivity to acute ethanol exposure without changes in ethanol preference. No differences were observed in in cognition, memory, or overall movement, although GLT1 deficient mice reared significantly more than their wild type counterparts. Combined, this data suggests that astrocytic GLT1-deficient animals may be less sensitive to the effects of aversive stimuli or stress. However, because stress and corticosterone are known to regulate GLT1 expression, further investigation is necessary to clarify the region or circuit-specific roles of GLT1 and to more fully understand how GLT1 may contribute to psychiatric disorders.

16:00 *Gaston Risi*

38. Development of a novel assay to identify genes involved in host infection by parasitic nematodes

SPEAKER: *Gaston Risi*

ABSTRACT. G Risi1,2, E Ausmus3, G Salinas1,2, AG Vidal-Gadea3*

About one-quarter of the world population is estimated to be infected with parasitic nematodes. There is widespread resistance to currently used drugs in the veterinary field, and concern is growing that resistance may arise in humans parasitic nematodes. C. elegans is a free living nematode that has been especially suggested as a good model for anthelmintic drug, and target discovery. This project takes advantage of C. elegans' burrowing behavior to model the need of parasitic worms to cross biological barriers during infections. We are searching for new molecular targets to prevent nematodes from infecting livestock and other hosts. Worms will be placed in multi-well plates and fed bacteria using RNA interference to target a specific gene in each well. We are targeting genes with no homology in humans, but with homology in the parasitic nematode Haemonchus contortus. This will allow us to identify genes likely to be involved in parasitic nematode biology, but with no similar targets in humans (or livestock). Burrowing will be assessed in RNAi silenced animals. This assay will allow us to identify potential drug targets that can be used to impede nematocidal infections.

1Pasteur Institute of Montevideo, Montevideo, Uruguay, 2School of Chemistry, University of the Republic, Montevideo, Uruguay, 3School of Biological Sciences, Illinois State University,

Normal, IL *avidal@ilstu.edu Funding support: National Agency for Investigation and Innovation (ANII) of Uruguay internship to GR. (MOV_CA_2017_1_137068); NIH 1R15AR068583-01A1 grant to AGVG.

16:00 *Tsung-Han Kuo*

40. Altruistic allogrooming in subordinate male mice

SPEAKER: *Tsung-Han Kuo*

ABSTRACT. T-H Kuo, Y-S. Su Institute of Systems Neuroscience, National Tsing Hua University, Hsinchu, Taiwan (ROC), Funding support: Ministry of Science and Technology, Taiwan (ROC)

Animals perform a wide range of social behaviors for survival and reproduction. Understanding these behavior may provide a framework of social interactions in human and eventually help us to elucidate abnormalities in social behaviors under various psychiatric disorders. Allogrooming, or social grooming, is defined as grooming behavior between members of the same species and can be widely observed throughout animal kingdoms. This behavior plays an important role in social bonding and is generally believed to be an altruistic behavior because groomers face several immediate costs to benefit recipients. Although allogrooming has been documented in mice, the functions and mechanisms have never been well characterized due to intensive male-male aggression, which limits the observations of other social behaviors during resident-intruder assay. By comparing dominant and subordinate males under resident-intruder assay, we found that subordinate males mainly perform allogrooming instead of aggression while there is no change in other social behaviors. Covering exotic materials, like mineral oil, on a intruder significantly enhanced the allogrooming behavior from a groomer (resident), supporting the function of allogrooming in hygienic purpose. We also observed increase of immobile time of intruder during allogrooming interaction, suggesting that grooming recipients are directly benefited from this social interaction. Our study showed that allogrooming in mice is altruistic and can be easily observed in subordinate animals. This study established a new platform to investigate the biological mechanisms of altruistic behaviors for the future study.

16:00 *Lauren Bailey*

42. Heritable variation in reward sensitivity and impulsive action and choice in a

genetically diverse inbred mouse panel
SPEAKER: *Lauren Bailey*

ABSTRACT. Authors: Lauren Bailey, Samantha Cermak, Jared Bagley, J. David Jentsch

Affiliation: Department of Psychology (Behavioral Neuroscience), Binghamton University, Binghamton NY 13902

Drugs of abuse, including alcohol and stimulants like cocaine, produce subjective effects that are subject to individual variability, and genetic variation accounts for at least a portion of those differences. Notably, research in both animal models and human subjects point towards reward sensitivity and impulsivity as being trait characteristics that predict relatively greater positive subjective responses to stimulant drugs. Unfortunately, past efforts have yet to yield convincing insights into underlying genetic influences on these traits due to the characteristics of the mouse panels used. The Collaborative Cross (CC) recombinant inbred mouse strains, their inbred founders, and the Diversity Outbred (DO) mice that are derived from them are a powerful genetic reference panel that has potential as a tool for revealing genetic contributions to cocaine abuse and related traits. Here we describe use of the eight CC/DO founder strains to examine the heritability of reward sensitivity and impulsivity traits, as well as genetic correlations between these measures and existing addiction-related phenotypes. Methods. Founder strains were all tested for activity in an open field and reward sensitivity (intake of chocolate BOOST®). Mice were then divided into two counterbalanced groups and underwent reversal learning (impulsive action) or delay discounting (impulsive choice). Results. The founder mice demonstrate significant heritability for premature responding within the reversal task, locomotor activity, and reward sensitivity. At this preliminary stage, significant strain differences for delay discounting are unclear. This research was conducted within the broader, inter-laboratory effort of the Center for Systems Neurogenetics of Addiction (CSNA) to characterize CC and DO mice for multiple, cocaine abuse related traits. These data will facilitate the discovery of genetic correlations between predictive traits, which will then guide discovery of genes and genetic variants that contribute to addictive behaviors.

Funding: These studies were supported in part by PHS Grant P50-DA041602.

16:00 *Shkelzen Shabani*

44. TAAR1 mediates the aversive effects of methamphetamine in a genetic mouse model of low methamphetamine intake

SPEAKER: *Shkelzen Shabani*

ABSTRACT. 1Shabani S, 1Houlton S, 1Ghimire B, 1Casiquin C, 2,3Phillips TJ

Avidity for methamphetamine (MA) is genetically associated with the balance in sensitivity to the rewarding and aversive effects of MA. Studies using bidirectional selective breeding of mice for high and low voluntary MA drinking (MADR) identified a quantitative trait locus on chromosome 10 associated with MA intake. Further study identified the trace amine-associated receptor 1 (Taar1) gene as a relevant gene in that region. The gene product, TAAR1, is a receptor that regulates monoamine neurotransmission. The MADR mice were selectively bred from a founding population of C57BL/6J x DBA/2J F2 (B6D2F2) mice, using a two-bottle choice drinking procedure. The D2 strain and MA high drinking (MAHDR) mice are homozygous for a Taar1 allele that codes for a non-functional receptor and are insensitive to the aversive and hypothermic effects of MA. B6 and MA low drinking (MALDR) mice have at least one copy of the alternative Taar1 allele that codes for functional TAAR1, are sensitive to these MA effects and have low MA consumption. MA is a direct agonist at TAAR1, but also impacts monoamine levels via transporter actions. Whether MA induces aversion and hypothermia via TAAR1 stimulation is not known. To study this, we used the MALDR mice to determine whether the effects of a TAAR1 full agonist would be similar to those of MA. The agonist, RO-5256390, was studied for its ability to induce a conditioned taste aversion (CTA), a conditioned place aversion (CPA), and hypothermia. The ability of IP 2 or 4 mg/kg RO-5256390 to induce a CTA to sodium chloride was measured, and the ability of IP 0.05, 0.1, 0.5 or 4 mg/kg RO-5256390 to induce CPA was examined. One day following the end of each procedure, the mice were tested with the same RO-5256390 doses for hypothermic effects. Rectal temperatures were measured at time 0 and then at times 60, 120 and 180 min after injection. Both doses of the TAAR1 agonist induced robust CTA, compared to vehicle treatment, but there were no differences between the 2 and 4 mg/kg dose groups. Significant and robust hypothermic effects of both TAAR1 agonist doses were found at 60 min post injection, with no significant difference between these two groups. In the place conditioning

procedure, even the lowest dose of 0.05 mg/kg RO-5256390 induced robust CPA. Hypothermic effects were apparent after all doses of the agonist and were most profound 60 min post-injection; however, doses >0.05 mg/kg had larger hypothermic effects. Thus, similar to MA, a TAAR1-specific agonist has conditioned aversive and hypothermic effects in MALDR mice, suggesting that TAAR1 stimulation plays an important role in sensitivity to the aversive effects of MA. Aversive actions of MA mediated by TAAR1 may curb MA intake in MALDR mice.

1 Dept of Biology, Minot State University, Minot, ND USA. 2 Methamphetamine Abuse Research Center, Department of Behavioral Neuroscience, Oregon Health & Science University, Portland, OR. 3 Veterans Affairs Portland Health Care System, Portland, OR USA. Supported by: Institutional Development Award (IDeA) from the National Institute of General Medical Sciences of the NIH P20GM103442 (SS), the Department of Veterans Affairs (TJP), and NIH grants P50DA018165, U01DA041579 and R24AA020245 (TJP).

16:00 *Byron Jones*

46. Genetic differences in mice in neuroinflammatory effects of exposure to organophosphates

SPEAKER: *Byron Jones*

ABSTRACT. BC Jones1, JP O'Callaghan2, DB Miller2, W Zhao1

Gulf war illness (GWI) is a chronic disorder that afflicts up to one-third of the veterans of the Persian Gulf conflict. Why some individuals developed GWI while others have not, is the subject of our work. Soldiers were exposed to a number of chemicals, including organophosphates (OP) that inhibit cholinesterase. OPs also promote neuroinflammation. An animal model showed that high concentrations of corticosterone acted synergistically to increase the expression of pro-inflammatory cytokine genes. The animal model was the C57BL/6J (B6) male mouse. To address individual differences, we replicated the experiment, adding the DBA/2J (D2) strain and females. The design: Treatment groups and treatments. There were four conditions: Condition 1, Control. Injected i.p. with saline and six hours later killed by cervical dislocation and frontal cortex and hippocampus harvested. Condition 2, CORT. Corticosterone (200 mg/L) added to

drinking water for seven days prior to tissue harvest as in condition 1. Condition 3, DFP. Injected i.p. with 4 mg/kg diisopropylflurophosphate (DFP), an irreversible cholinesterase inhibitor and killed 6h later for tissue harvest. Condition 4, CORT+DFP. CORT in drinking water as in group 2 for seven days and on the eighth day, injected i.p. with 4 mg/kg i.p. DFP and tissues harvested six h later. The cytokines included (IL6), IL1-β, CCL2, LIF, OSM TNFα. Results. Overall, D2 mice were much less affected by CORT+DFP than the B6 mice and females of both strains less affected than males. Conclusion. These results support a likely genetic basis for individual differences in susceptibility to GWI.

1Department of Genetics, Genomics, and Informatics, University of Tennessee Health Center, Memphis, TN 2Center for Disease Control, National Institute of Environmental Health and Safety, Morgantown, WV Funding: USPHS Grant ES022614; DoD GWI 160086

16:00 *Catherine Czeisler*

48. PHOX2B regulates development of nested astrocytes that control breathing and sleep homeostasis

SPEAKER: *Catherine Czeisler*

ABSTRACT. Embryonic neural patterning genes regulate both patterning and formation of autonomic circuits. These autonomic circuits require appropriate neuronal and glial integration for efficient circuit function. However, the developmental interdependencies between neurons and glia of specific circuits has not been established. Here, we report the generation of an astrocyte population derived from PHOX2B patterned precursors in the mouse brainstem. These astrocytes are necessary for appropriate function of the PHOX2B-derived chemosensory centers, sleep homeostasis and proper synapse morphology. These findings indicate that PHOX2B may act by patterning precursor cells that later generate both astrocytes and neurons destined to form functional autonomic circuits.

1 The Ohio State University College of Medicine, Department of Pathology, 2Department of Physiology and Biophysics, Institute of Biomedical Science, University of Sao Paulo, 3 The Ohio State University College of Medicine, Department of Neuroscience, 4The Ohio State University College of Engineering, Department of Mechanical and Aerospace Engineering, 5The

Ohio State University Mathematical Biosciences Institute, 6 The Ohio State University Campus Microscopy and Imaging Facility, 7Department of Pharmacology, Institute of Biomedical Science, University of São Paulo. *= Co-corresponding Author. United States studies were supported by NIH/NHLBI R01HL132355. Brazil research was supported by the São Paulo Research Foundation (FAPESP; grants: 2014/22406-1 to ACT; 2015/23376-1 to TSM) and the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq; grant: 471263/2013-3 to ACT). CNPq fellowship (301219/2016-8 to ACT and 301904/2015-4 to TSM).

16:00 *Catharine Rankin*

50. High-throughput phenomic characterization of ASD-associated genes reveals a functional gene network underlying hypersensitivity and impaired habituation
SPEAKER: *Catharine Rankin*

ABSTRACT. A primary challenge facing Autism Spectrum Disorder (ASD) genetics is the large and growing number of genes and gene variants of unknown functional significance. Here, we used Caenorhabditis elegans to systematically functionally characterize ASD-associated genes in vivo. Using our custom machine vision system we characterized 26 quantitative phenotypes spanning morphology, locomotion, sensory, and habituation learning in 97 strains of C. elegans each carrying a mutation in an ortholog of an ASD-associated gene. This research has generated a large number of novel genotype to phenotype relationships that range from severe developmental delays and uncoordinated movement to subtle deficits in sensory and learning behaviours. Clustering based on multi-parametric phenomic profiles revealed a set of 12 genes that all result in a strikingly similar profile characterized by hypersensitivity and impaired habituation learning. Current epistasis experiments are aimed at determining whether the phenomic similarity among members of this cluster results from previously undiscovered functional interactions. One of the genes in this cluster is the sole C. elegans ortholog of neuroligins, nlg-1. Transgenic pan-neuronal expression of human NLGN3 in nlg-1 mutant C. elegans rescued their sensory and learning impairments; confirming functional conservation. The wealth of in vivo phenomic functional data generated in this work will inform more targeted studies in vertebrates and offers novel positive and negative pathway components as

therapeutic targets for ameliorating the effects of ASD.

1. Djavad Mowafaghian Centre for Brain Health, University of British Columbia, 2211 Wesbrook mall, Vancouver, B.C., V6T 2B5; 2. Dept. of Psychology, UBC, 2136 West Mall, Vancouver, B.C., V6T 1Z4 3. Department of Cell and Physiological Sciences, 2350 Health Science Mall, B.C. V6T 1Z3 4. Department of Psychiatry, 2255 Wesbrook Mall, B.C. V6T 2A1

16:00 *Chance Bainbridge*

52. Optimization of behavioral search strategies to cues of distinct physical natures
SPEAKER: *Chance Bainbridge*

ABSTRACT. To successfully navigate their surroundings, animals must decide how much attention they dedicate to the variety of environmental stimuli they routinely encounter. This decision is often reflected on the nervous system allocation of resources to different stimuli (e.g. humans rely on vision more than touch). In nature, animals must reconcile the physical nature, and the saliency of stimuli to optimize search strategies using finite behavioral repertoires. Most work in the field has focused on orientation strategies specific to highly salient stimuli (e.g. temperature or chemical gradients). It remains unclear how animals optimize their behavioral strategies for stimuli of different physical nature. Like many animals, the nematode C. elegans detects a large variety of stimuli and responds using a small well described repertoire of orientation strategies. Here we use C. elegans to investigate how it optimizes navigation to physically distinct stimuli of different salience. We recorded the worms orienting to temperature, chemical, or magnetic field stimuli and analyzed heading and orientation strategies. Worms appear to orient differentially in the presence of a temperature gradient by increasing velocity and clustered reversals (i.e. pirouettes) as a search strategy that was not observed in magnetic orientation or chemotaxis. Conversely, pirouettes appear suppressed in chemical gradients while gradual turning (weathervaning) is increased. These results suggest that C. elegans uses distinct orientation strategies depending on the physical nature of a stimulus.

16:00 *Abigail Benson*

54. Behavioral effects of RNAi-mediated silencing of gap junction proteins in Marbled Crayfish
SPEAKER: *Abigail Benson*

ABSTRACT. Gap junctions facilitate intercellular communication and rapid information processing in the nervous system, mediating swift and coordinated behavioral responses. In invertebrates, innexin are the structural components of gap junctions and provide direct entryways for electrical currents between neurons. While gap junction structure and function are well known, the specific contribution of innexins to neural circuit function and behavior remain poorly understood. We use RNA interference (RNAi) to target innexin expression in the marbled crayfish, Procambarus virginalis. Crayfish are well-suited for electrical coupling and behavioral studies due to their well-characterized tail-flip escape responses and their coordinated leg movements during locomotion. We hypothesize that innexins contribute substantially to both behaviors. Our analyses indicate that marbled crayfish share homology for seven innexin genes with other species, up to 84% within Cancer borealis, Homarus americanus and Homarus gammarus. To test specific expression in the ventral nerve cord (containing neurons that mediate both behaviors), we designed and tested innexin-specific primers. Agarose gel analyses showed that at least innexin-4 and innexin-2 are expressed. We were able to characterize 90% of the putative innexin-2 exons, and thus selected innexin-2 as the first candidate for investigating the behavioral consequences of innexin silencing. To induce RNAi, we created innexin-2 double-stranded RNA (~553bp). We are currently assessing innexin-2 contribution to locomotion and escape behaviors by subcutaneous injecting of dsRNA and measuring 1) innexin-2 suppression levels using real-time PCR, 2) effects of silencing on electrical communication in the escape circuit, and 3) both behaviors at 24, 48, and 72 hours post-injection.

16:00 *Keith Babbs*

56. Genetic differences in the behavioral organization of binge eating, conditioned food reward, and compulsive-like eating in C57BL/6J and DBA/2J mice and in an F2 cross

SPEAKER: *Keith Babbs*

ABSTRACT. Binge eating (BE) is a heritable symptom of eating disorders associated with anxiety, depression, malnutrition, and obesity. Determining its genetic basis could facilitate discovery of more efficacious therapeutics. We

used an intermittent, limited access paradigm to examine inbred mouse strain differences in BE of sweetened palatable food (PF), conditioned food reward, and compulsive-like eating between C57BL/6J (B6J) and DBA/2J (D2J) inbred mouse strains. We identified a robust escalation in PF consumption in D2J and a conditioned place preference for the PF-paired side. D2J also showed compulsive-like eating in the light/dark conflict test, revealing a unique acquire-retreat hoarding-like behavior of PF to the dark side of the apparatus. To gain insight into the genetic basis of BE in D2J versus B6J, we phenotyped and genotyped a cohort of 133 B6JxD2J-F2 mice at three historic, B6J/D2J-derived quantitative trait loci (QTLs), including a chr.4 QTL associated with sweet taste (156 Mb), a chr.6 QTL associated with bitter taste (133 Mb), and a chr.11 QTL (50 Mb) associated with behavioral sensitivity to drugs of abuse, including methamphetamine, cocaine, and ethanol. Both the chr 6 and chr 11 D2J alleles were associated with increased PF consumption and slope of escalation, thus accounting for a portion of phenotypic variance. In contrast, the sweet taste locus did not affect PF consumption. Hnrnph1 haploinsufficient mice (chr.11: 50 Mb) showed an increase in PF consumption, supporting Hnrnph1 as a causal gene. QTL mapping in BXD recombinant inbred lines will identify other genes contributing to the various components of BE.

1 Laboratory of Addiction Genetics, Department of Pharmacology and Experimental Therapeutics, Boston University School of Medicine,

2 Boston University Undergraduate Research Opportunity Program (UROP),

3 NIGMS Training Program in Biomolecular Pharmacology, Boston University School of Medicine,

4 Boston University Transformative Training Program in Addiction Science (TTPAS), Burroughs Wellcome Fund,

5 Departments of Genetics, Genomics and Informatics and Anatomy and Neurobiology, University of Tennessee Health Science Center.

16:00 *John Mootz*

58. Trace amine-associated receptor 1 functionality can dictate some methamphetamine-related behaviors
SPEAKER: *John Mootz*

ABSTRACT. To explore the genetic origins of methamphetamine (MA) use disorder, selective breeding was used to create lines of mice for high (MAHDR) and low (MALDR) MA drinking from the F2 cross of the DBA/2J (D2) and C57BL/6J (B6) inbred strains. Sensitivity to MA-induced conditioned taste aversion (CTA) is genetically correlated with amount of MA consumed in the selected lines, indicating some common genetic regulation. A quantitative trait locus was found that accounts for >50% of the genetic variance in MA intake in the lines. The trace amine-associated receptor 1 (Taar1) falls within this region, and MA is a direct agonist at the receptor (TAAR1). Five replicate sets of lines have been produced, and all replicates of the MAHDR lines are homozygous for a mutant Taar1 allele (Taar1m1J), derived from the D2 strain, which expresses a nonfunctional receptor. The MALDR lines are heterozygous or homozygous for the alternative Taar1 allele, derived from the B6 strain, which expresses a functional receptor. Existing data are consistent with the hypothesis that Taar1m1J homozygosity drives the higher MA intake in MAHDR mice. To confirm this, CRISPR-Cas9 allele swap was performed, replacing Taar1m1J in MAHDR mice with B6 Taar1. In two-bottle choice and CTA procedures, MAHDR allele-swapped mice consumed less MA, relative to typical MAHDR mice, and exhibited levels of these phenotypes similar to those of MALDR mice. This proves that TAAR1 is responsible for differences in MA intake and CTA between the MADR lines, and substantiates interest in the role of Taar1 in MA abuse.

1Oregon Health & Science University, 2Veterans Affairs Portland Health Care System, Portland, OR, USA. Funding Support: Department of Veterans Affairs I01BX002106, NIH NIDA P50DA018165 and U01DA041579

16:00 *Richard Brown*

60. Frailty as a measure of healthspan and lifespan in transgenic mice

SPEAKER: *Richard Brown*

ABSTRACT. Mice of different strains and transgenic mouse models of AD have marked differences in life expectancy depending on the genotype and sex of the model (Rae & Brown 2015 Neurosci Biobehav Rev. 57: 238-51). A measure of aging which is independent of chronological age is necessary to compare age-related changes in mice with different lifespans. Frailty could provide a measure of healthspan

facilitate comparisons of aging between mice of different genotypes and sexes. This study used a validated mouse frailty index (FI) assessment tool based on deficit accumulation (Whitehead et al., 2014 J Gerontol Biol Sci Med Sci 69: 621–32) to explore genotype and sex differences in lifespan and healthspan of 3xTg-AD mice. Results showed that male 3xTg-AD mice aged 300-600 days had a higher FI score (Mean FI=0.21) than either male wild-type (Mean FI=0.15) or female 3xTg-AD mice (Mean FI=0.10), and the elevated frailty scores were accompanied by parallel increases in mortality. Frailty increased exponentially with age in all groups, and higher rates of deficit accumulation elevated mortality risk in all groups. When mice were stratified by FI score, frailty predicted mortality, at least in females. The mouse clinical FI provides a valuable tool that can be used to evaluate healthspan in mice of different strains and in transgenic mice.

16:00 *Booher Winona*

62. AAV Mediated MiR-138 Injections into the Medial Habenula of Mice Reduces Nicotine Consumption

SPEAKER: *Booher Winona*

ABSTRACT. In 2015, 36.5 million U.S. adults regularly smoked cigarettes, however 68% wished they could quit (CDC). Although there are several nicotine replacement products and prescription non-nicotine medications to facilitate the quitting process, the success rate is still less than 10%. Pre-vious human genetics studies in our lab led to an interest in a microRNA (miRNA; miR-138), which is potentially involved in expression of the CHRNB4 gene (Gallego et al., 2013). CHRNB4 is highly expressed in the medial habenula (MHb) and is important for nicotine intake/preference behaviors. To assess whether miR-138 is behaviorally relevant in vivo, an adeno-associated virus (AAV) containing either miR-138 or a scrambled control miRNA was injected into the MHb of male C57BL/6J mice. The mice were then singly housed and given a two-bottle choice procedure where one bottle contained water and one bottle contained escalating concentrations of nicotine (25 - 200 ug/mL). Compared to the mice injected with the scrambled control miRNA, those receiving the miR-138 injection showed a reduction in voluntary nicotine intake/preference. We hypothesize that this reduction of nicotine consumption is caused by reduced expression of Chrnb4 in the MHb, which will be tested using epibatidine-binding experiments following AAV injections. Finally, because of the known sex

differences in smoking behaviors, we have begun the same behavioral testing procedures in females. This study will provide improved understanding of the role miR-138 contributes to nicotine addiction and may help in developing potential treatments for nicotine dependence.

1Institute for Behavioral Genetics, University of Colorado, Boulder, CO, USA 2Department of Integrative Physiology, University of Colorado, Boulder, CO, USA

16:00 *Johanna Kowalko*

64. Uncovering the role of the oculocutaneous albinism 2 gene in the evolution of behavior in the Mexican cavefish Astyanax mexicanus

SPEAKER: *Johanna Kowalko*

ABSTRACT. There is an immense amount of diversity observable in the living world today. Understanding the genetic basis of trait evolution is critical to identifying the mechanisms that generated this diversity and the evolutionary processes that led to the establishment of new traits. Astyanax mexicanus, the blind Mexican cavefish, exists in two interfertile forms, a surface-dwelling form and multiple independently evolved cave-dwelling forms. As cavefish have evolved a number of behavioral traits and A. mexicanus are amenable to genetic manipulation, this system provides a unique opportunity to functionally test candidate genes for their role in evolution of behaviors. We have leveraged CRISPR/Cas9 genome editing techniques to mutate the oculocutaneous albinism 2 (oca2) gene in surface fish. Mutations in this gene are hypothesized to underlie both the evolution of albinism and, through changes in catecholamine levels, evolution of behaviors. Thus, through examining CRISPR-generated mutant fish, we can gain unprecedented insight into the consequences of naturally occurring mutations in the evolution of morphology and behavior.

16:00 *David G Mets* and *Michael S. Brainard*

66. Genetic variation interacts with experience to determine inter-individual differences in learned song

SPEAKER: *David G Mets*

ABSTRACT. Learning reflects the influence of experience on genetically determined circuitry, but little is known about how experience and genetics interact to determine complex learned phenotypes. Here, we used vocal learning in songbirds to study how experience and genetics

contribute to inter-individual differences in learned song. Previous work has established that such differences in song within a species depend on learning, but in principle some of these differences could also depend on genetic variation. We focused on song tempo, a learned and quantifiable feature that is controlled by central neural circuitry. To identify genetic contributions to tempo we computer-tutored juvenile Bengalese finches (Lonchura striata domestica) from different genetic backgrounds with synthetic songs in which tempo was systematically varied. Computer-tutored birds exhibited unexpectedly strong heritability for song tempo, and comparatively weak influence of experience. We then tested whether heritability was fixed and independent of experience by providing a second group of birds with enriched instruction via live social tutoring. Live-tutoring resulted in not only a significant increase in the influence of experience on tempo, but also a dramatic decrease in the influence of genetics, indicating that enriched instruction could overcome genetic biases evident under computer tutoring. Our results reveal strong heritable genetic contributions to inter-individual variation in song tempo, but that the degree of heritability depends profoundly on the quality of instruction. They suggest that for more complex learned phenotypes, where it can be difficult to identify and control relevant experiential variables, heritability may similarly be contingent on the specifics of experience.

Department of Physiology, University of California, San Francisco, CA 94158; Center for Integrative Neuroscience, University of California, San Francisco, CA 94158; Howard Hughes Medical Institute, University of California, San Francisco, CA 94158.

18:00-21:00 Session : Dinner on your own

Dinner on your own

2018 IBANGS MEETING: THE 20TH ANNUAL GENES, BRAIN & BEHAVIOR MEETING

PROGRAM FOR SUNDAY, MAY 20TH

Days: previous day next day all days

View: session overview talk overview

08:30-09:30 Session Reg: Registration

Meeting Registration

LOCATION: Mayo Civic Center Grand Lobby West

09:00-10:00 Session 9: Distinguished Scientist Award

Distinguished Scientist Awardee

CHAIR: *Rob Williams*

LOCATION: Mayo Civic Center 102/103

09:00 *Rainer Spanagel*

Neurochemical mechanisms of addicted behaviour

SPEAKER: *Rainer Spanagel*

ABSTRACT. Rainer Spanagel Institute of Psychopharmacology, Central Institute of Mental Health, Medical Faculty Mannheim, University of Heidelberg, Mannheim, Germany rainer.spanagel@zi-mannheim.de The characterization of disease-related neurocircuitries is fundamental for understanding mental disorders and for drug development. Neuroscientists are currently making a major effort to map those neurocircuitries, mainly by means of tracing studies and optogenetic methodology. Here I will present an alternative approach based on the first global neurochemical connectome and a novel database on neurochemical fingerprints produced by psychoactive drugs. By using advanced data mining, supervised machine learning, and network analysis we have generated a multiscale, multilayer neurochemical connectome of the rat brain (www.chemnetdb.org). The connectome consists of 125 scale-consistent cerebral nuclei and 2,931 multiscale, multi-chemical connections. In addition we have used a further big data approach to extract and integrate data from pharmacological microdialysis experiments. We systematically extracted data from 3,383 original in vivo microdialysis experiments (110,674 rats) which resulted in a large database (Systematic Pharmacological Database or Syphad (www.syphad.org)). We

have then developed an in silico learning system that integrates Syphad and ChemNetDB data for predicting neurochemical fingerprints of various drug consumption patterns. In this lecture I will present data on neurochemical fingerprints of repeated intoxicating cycles of alcohol exposure. Our simulation of neurochemistry following chronic alcohol exposure predicts switches in neurochemical activity, especially a hyper-dopaminergic state in the nucleus accumbens and a hyper-glutamatergic state in the infralimbic cortex. These predictions can indeed be validated in post-mortem brain tissue from deceased alcoholics and in the alcohol-dependent rat brain. Furthermore, we show how mGluR2 agonist can restore these neurochemical switches and can thereby reduce alcohol craving and relapse.

10:00-10:30 Session Break: AM Break

AM Break

LOCATION: Mayo Civic Center 104/105

10:30-12:30 Session 10: Selected Talks II

Selected Talks 2

CHAIR: *Gang Chen*

LOCATION: Mayo Civic Center 102/103

10:30 *David Mets*

Genetic variation interacts with experience to determine inter-individual differences in learned song

SPEAKER: *David Mets*

ABSTRACT. David G. Mets1 and Michael S. Brainard1 Learning reflects the influence of experience on genetically determined circuitry, but little is known about how experience and genetics interact to determine complex learned phenotypes. Here, we used vocal learning in songbirds to study how experience and genetics contribute to inter-individual differences in learned song. Previous work has established that such differences in song within a species depend on learning, but in principle some of these differences could also depend on genetic variation. We focused on song tempo, a learned and quantifiable feature that is controlled by central neural circuitry. To identify genetic contributions to tempo we computer-tutored juvenile Bengalese finches (Lonchura striata domestica) from different genetic backgrounds with synthetic songs in which tempo was systematically varied. Computer-tutored birds exhibited unexpectedly strong heritability for song tempo, and comparatively weak influence of

experience. We then tested whether heritability was fixed and independent of experience by providing a second group of birds with enriched instruction via live social tutoring. Live-tutoring resulted in not only a significant increase in the influence of experience on tempo, but also a dramatic decrease in the influence of genetics, indicating that enriched instruction could overcome genetic biases evident under computer tutoring. Our results reveal strong heritable genetic contributions to inter-individual variation in song tempo, but that the degree of heritability depends profoundly on the quality of instruction. They suggest that for more complex learned phenotypes, where it can be difficult to identify and control relevant experiential variables, heritability may similarly be contingent on the specifics of experience. 1 Department of Physiology, University of California, San Francisco, CA 94158; Center for Integrative Neuroscience, University of California, San Francisco, CA 94158; Howard Hughes Medical Institute, University of California, San Francisco, CA 94158.

10:45 *Anna Kukekova*

Transcriptome analysis of the hypothalamic-pituitary-adrenal axis in tame and aggressive foxes (Vulpes vulpes)

SPEAKER: *Anna Kukekova*

ABSTRACT. AV Kukekova1, JP Hekman1, JL Johnson1, W Edwards2, AV Vladimirova3, RG Gulevich3, AV Kharlamova3, LT Raetzman2, LN Trut3 Domesticated species are characterized by reduced fearfulness, increased social tolerance, and increased resistance to stress. These behaviors are closely linked to reduced reactivity of the hypothalamic-pituitary-adrenal (HPA) axis, the hormonal cascade associated with the stress response in mammals. Specifically, reductions in circulating levels of adrenocorticotrophic hormone (ACTH), released by the anterior pituitary, and glucocorticoids, released by the adrenals, have been demonstrated in several domesticated species. Here we used the tame and aggressive strains of the red fox to explore mechanisms associated with the HPA axis reactivity. RNA extracted from the anterior pituitaries of six tame and six aggressive foxes and from the right adrenal glands of 11 tame and 11 aggressive foxes was sequenced on an Illumina HiSeq2500. The gene expression and network analyses looking at the differences in anterior pituitaries of tame and aggressive foxes indicated the importance of genes related to the regulation of exocytosis, specifically mediated by cAMP, the organization of pseudopodia, and cell

motility. In adrenals, differential gene expression analysis suggested differences in ectodermal cell differentiation and interleukin-8 production, while weighted gene co-expression network analysis found differences in cholesterol biosynthesis and cell migration, suggesting that the biosynthesis of cholesterol (a steroid hormone precursor), rather than the biosynthesis of adrenal steroid hormones may differ between the two strains. These finding suggest that the tame and aggressive foxes may have differences in HPA reactivity due to differences in regulation of the hormone release. 1Department of Animal Sciences, College of Agriculture, Consumer, and Environmental Sciences, University of Illinois at Urbana-Champaign, IL 61801, USA 2Department of Molecular and Integrative Physiology, School of Molecular and Cellular Biology, College of Liberal Arts and Sciences, University of Illinois at Urbana-Champaign, IL 61801, USA 3Institute of Cytology and Genetics of the Russian Academy of Sciences, Novosibirsk, 630090, Russia

11:00 *Halie Rando*

Genomic targets of selection for behavior in the red fox

SPEAKER: *Halie Rando*

ABSTRACT. Halie M. Rando1,2, Jennifer L. Johnson2, Guojie Zhang3,4,5, Lyudmila N. Trut6, Anna V. Kukekova2 1Illinois Informatics Institute, 2Department of Animal Science, College of Agricultural, Consumer and Environmental Sciences, University of Illinois at Urbana-Champaign, Urbana, IL USA; 3China National Genebank, BGI-Shenzhen, Shenzhen, Guangdong, China; 4Section for Ecology and Evolution, Department of Biology, University of Copenhagen, Copenhagen, Denmark; 5State Key Laboratory of Genetic Resources and Evolution, Kunming Institute of Zoology, Chinese Academy of Sciences, Kunming, China; 6Institute of Cytology and Genetics of the Russian Academy of Sciences, Novosibirsk, Russia; Funding Support: NIH R01 GM120782 and National Defense Science & Engineering Graduate Fellowship (NDSEG) Program

Sociality is fundamental to human and animal behavior, and atypical social behavior is implicated in many human psychiatric and neurodevelopmental disorders. Though mapping behavioral traits in humans is very challenging, the selective breeding of red foxes (Vulpes vulpes) presents a novel model for the genetic regulation of social behavior. Fifty generations of

breeding has produced hypersociable “tame” foxes and defensively “aggressive” foxes in which 9 behavioral quantitative trait loci (QTL) were identified, but low marker resolution prevented the identification of candidate genes. With the new red fox genome assembly, these regions can now be fine mapped. The genomes of 30 foxes (10 each from the tame, aggressive, and conventional farm-bred strains) were each sequenced at 2.5x and pooled by population for SNP calling. SNPs were analyzed within sliding windows for within-population selective sweeps and between-population divergence (FST). Additional foxes comprising the least-related individuals from the tame (N=20) and aggressive (N=22) strains were then sequenced at 15x each, and putative regions under selection were identified. In the pooled analysis, either Hp and/or FST identified 103 genomic regions as likely to be under selection. Thirty of these regions overlapped 5 previously identified fox QTL. Loci within QTL intervals identified by both the pooled and individual analyses are strong candidates for social behavioral regulation. The deep sequencing data also presents the first opportunity to characterize haplotypes and even polymorphisms differentiating the tame and aggressive strains to identify novel candidates for the genetic regulation of mammalian social behavior.

11:15 *Svitlana Bach*

CRISPR/dCas9 manipulation of Bdnf transcription in rat primary hippocampal neurons

SPEAKER: *Svitlana Bach*

ABSTRACT. SV Bach, D Hosein, D Williams, NV Gallus, FA Sultan, KD Bunner, KE Savell & JJ Day

Brain-derived neurotrophic factor (Bdnf) plays a critical role in brain development, neuronal differentiation, dendritic growth and synaptic plasticity. Rodent Bdnf gene consists of nine 5’ non-coding exons (I-IXa) and one 3’ coding exon (IX). Each non-coding exon has its own promoter region where transcription of different variants is initiated. To investigate specific roles of the activity-regulated Bdnf variants I and IV we used a CRIPSR/dCas9 – VPR system, in which a strong transcriptional activator, VPR, is targeted to Bdnf I and IV promoter regions with the help of specific guide RNAs (gRNAs). Using this system in primary rat hippocampal neurons we are able to selectively upregulate Bdnf variants I and IV

from their endogenous gene loci while leaving other variants mostly unaffected. To assess functional significance of selective Bdnf variant upregulation, we used Multichannel Electrode Arrays (MEAs) to perform single-unit recordings from neurons treated with CRISPR constructs. Upregulation of select Bdnf variants causes an increase in the spike frequency as well as the number of spontaneously active neurons. To assess subcellular localization of Bdnf mRNAs, single-molecule RNA fluorescent in situ hybridization (FISH) was used to visualize individual Bdnf I, IV and IX transcripts, which occupy diverse cellular compartments upon neuronal depolarization with potassium chloride. With this work we demonstrate the unprecedented precision of the endogenous Bdnf transcript variant upregulation using CRIPSR/dCas9 tools, the functional significance of Bdnf transcript variant upregulation for neuronal physiology, and the subcellular localization of Bdnf mRNAs with unparalleled resolution.

Department of Neurobiology, University of Alabama at Birmingham, Birmingham, AL 35294, USA

11:30 *Marissa Ehringer*

Whole genome sequencing of replicate inbred strains of mice selected for high and low open-field activity

SPEAKER: *Marissa Ehringer*

ABSTRACT. MA Ehringer1, MS Powers1, LM Evans1, JC DeFries1 1Institute for Behavioral Genetics, University of Colorado Boulder; Boulder, Colorado, USA.

Anxiety disorders lead to debilitation in individuals and families, leading to high social and economic costs. Genetic factors have been shown to contribute to underlying risk for anxiety, but little is known about the specific genes and gene networks involved. Open-field activity is commonly used as a mouse model for anxiety-related behaviors, and bidirectional selection for high and low open-field activity was initiated in 1965 (DeFries et al, 1978). This process generated replicate inbred strains that differ ~40-fold in activity. We recently completed whole-genome sequencing of one male and one female from each of the four strains (H1, H2, L1, L2) using an average 10X coverage depth to pursue three separate questions. First, we examine strain distribution patterns in the four strains for all variants that differed between the BALB/cJ and C57BL/6J parental strains. Next, we assess

genome-wide allele sharing both between and within the High and Low strains (viz., H1 vs. L1, H2 vs. L2, H1 vs. H2, and L1 vs. L2). Finally, genomic regions previously identified with high confidence from QTL studies will be carefully investigated for differences shared by both High compared to Low strains. This will be the first report of whole genome sequencing for these unique strains, which will provide a valuable resource for future studies that may integrate data generated from other genomic technologies to reveal underlying molecular genetic differences contributing the extreme phenotypic differences.

Funding Support: T32 AA007464; University of Colorado Institutional funds

11:45 *Antonios Diab*

The adaptor protein Nck1 regulates of anxiety-like behaviours

SPEAKER: *Antonios Diab*

ABSTRACT. Advances in genomics and proteomics have implicated a number of genes involved in actin polymerization in psychiatric and neurological disorders associated with irregular synaptic function. Here we examine the function of Nck1, an intracellular scaffolding protein known to play a role in actin dynamics, in the mouse brain and behavior. Our study found that mice lacking Nck1 display increased levels of anxiety-like behaviors, while other sensorimotor responses, including vision, olfactory responses and locomotion are indistinguishable from controls. The anxiety-like phenotype was rescued by treatment with diazepam, an anxiolytic that functions as a positive allosteric modulator of the GABAA receptor, implicating a deregulation of inhibitory control. Given that the amygdala is closely coupled with anxiety-like behaviours, we focused our further analysis on this brain region. We show that in the amygdala Nck1 is expressed in neurons but not microglia. Further analysis revealed that mice lacking Nck1 had fewer synapses and changes in synaptic morphologies in the spiny principal neurons of the basolateral amygdala. Taken together our study proposes that Nck1 is likely important in regulating neuronal excitability. Finally, our data suggests that Nck1 is necessary for normal synapse development in the amygdala and that the loss of Nck1 leads to the development of behaviors linked to anxiety.

1Department of Pharmacology, 2Department of Surgery, Dalhousie University, Halifax, Nova Scotia, Canada. Funding Support: CIHR (MOP 341174)

12:00 *Catharine Rankin*

Functional Variomics Group: Precise structure-function analysis of ASD associated gene variants in PTEN using targeted CRISPR gene replacement in Caenorhabditis elegans

SPEAKER: *Catharine Rankin*

ABSTRACT. A primary challenge facing Autism Spectrum Disorder genetics is the large and growing number of genes and gene variants of unknown functional significance. To determine the functional effects of ASD-associated variants we developed a novel strategy based on CRISPR-Cas9 genome engineering in the high-throughput genetic model organism Caenorhabditis elegans to generate single-copy integrated knock-in lines expressing the exact human gene variants identified in ASD. We have begun by focusing on the high-confidence ASD-associated gene PTEN. In C. elegans, the sole ortholog of PTEN, daf-18, regulates attractive navigation behaviour up a concentration gradient of NaCl (this behaviour is called NaCl chemotaxis). Using a novel automated machine vision chemotaxis paradigm we have shown that either directly replacing daf-18 with human WT PTEN using CRISPR or nervous system specific expression of human WT PTEN is able to substitute for loss of daf-18 and rescue this behavioural deficit. Surprisingly, all ASD-associated missense mutations in PTEN assessed resulted in partial or complete loss-of-function and failed to rescue this sensory deficit. Collaborative complementary in vivo functional assays in yeast, and fly as well as in vitro assays in HEK293 cells and rat neural culture directly corroborate our finding that ASD-associated PTEN variants are loss-of-function. The wealth of in vivo empirical data from this research will improve algorithms that estimate the pathogenicity of missense mutations, improve diagnostic accuracy, and further precision medicine efforts to treat ASD.

Troy A. McDiarmid1, Kathryn Post3, Riki Dingwall3, Payel Ganguly3, Matthew Edwards3, Ben Callaghan4, Manuel Belmadani4, Fabian Meili1, Warren Meyers1, Barry Young3, Sanja Rogic4, Chris Loewen3, Douglas Allan1,3, Timothy O'Connor1,3, Shernaz Bamji1,3, Paul Pavlidis4, Kurt Haas1,3, Catharine H. Rankin1,2

1. Djavad Mowafaghian Centre for Brain Health, University of British Columbia, 2211 Wesbrook mall, Vancouver, B.C., V6T 2B5; 2. Dept. of Psychology, University of British Columbia, 2136 West Mall, Vancouver, B.C., V6T 1Z4 3. Department of Cell and Physiological Sciences,

2350 Health Science Mall, B.C. V6T 1Z3 4. Department of Psychiatry, 2255 Wesbrook Mall, B.C. V6T 2A1

12:30 *Annie Park*

Alcohol-Intoxicated Flies Become Aggressive
SPEAKER: *Annie Park*

ABSTRACT. Alcohol-induced violence causes an immense social and economic burden worldwide. Despite the pervasiveness of this phenomenon, it is an understudied behavior and it's neurogenetic underpinnings are unknown. In this study, we describe a novel fly alcohol behavior; male Drosophila melanogaster become more aggressive after being exposed to a single low dose of alcohol. After this same low dose of alcohol female flies become more receptive to courtship, but do not exhibit alcohol-induced aggression. We predicted that alcohol was cross-potentiating the endogenous cVa (11-cis-vaccenyl acetate) sex-pheromone pathway, which resulted in potentiation of aggression and receptivity to courtship. The cVa sensitive olfactory receptor neurons (ORNs) express both FruM and Or67d. Here we show that both FruM and Or67d are necessary for alcohol-induced aggression. When flies are exposed to high levels of alcohol they have lower levels of FruM and have specifically downregulated FruM production in the Or67d expressing ORNs that project to the DA1 glomerulus. Flies previously treated with this higher dose of alcohol show reduced aggression. Taken together, these results suggest that ethanol regulates FruM to produce changes in aggression in a dose dependent manner.

12:30-13:30 Session L3: Lunch

Lunch

13:30-14:30 Session 11: Business Meeting

Business Meeting

LOCATION: Mayo Civic Center 102/103

14:30-15:00 Session Break: PM Break

PM Break

LOCATION: Mayo Civic Center 104/105

15:00-17:00 Session 12: Workshop Integrative Genomics

Description

Robert W. Williams1, Elissa J. Chesler2 University of Tennessee Health Science Center1; The Jackson Laboratory2 Systems genetics has been an effective

strategy for gene discovery and for finding the relations among behavioral traits and their neurobiological mechanisms in genetic reference populations. Integrative genomics combines data from diverse experimental paradigms to find the common and distinct biological mechanisms of biological and behavioral traits within and across species. As these methods have evolved, so too have the software systems that support interactive user initiated analyses. Flexible tools have made rapid interrogation of integrative genetic and genomic data resources possible.Participants in this workshop will have an opportunity for hands on demos using thelatest versions of Gene Network, Mouse Phenome Database and GeneWeaver tools for integrative genetic and genomic analyses. These resources allow users to explore the relations among genetics, genomics, behavior and other disease related phenotypes inan interactive manner. Participants will be guided through the use of these platforms using simple tutorial demonstrations. Following the introductory tutorial demonstrations,

CHAIRS: *Elissa Chesler* and *Rob Williams*
LOCATION: Mayo Civic Center 102/103

18:00-19:00 Session : Dinner on your own

Dinner on your own

2018 IBANGS MEETING: THE 20TH ANNUAL GENES, BRAIN & BEHAVIOR MEETING

PROGRAM FOR MONDAY, MAY 21ST

Days: previous day all days

View: session overview talk overview

08:00-09:00 Session Registration: Registration

Meeting Registration

LOCATION: Mayo Civic Center Grand Lobby West

08:30-10:30 Session 13: Symposium 4

Complex models, complex disorders

CHAIR: *Sarah Schoenrock*

LOCATION: Mayo Civic Center 102/103

08:30 *Abraham Palmer*

Using N/NIH heterogeneous stock (HS) rats to study the genetic baiss of behavior

SPEAKER: *Abraham Palmer*

ABSTRACT. Drug abuse continues to be an extremely serious public health problem. While human genome-wide association studies (GWAS) have begun to elucidate genes that influence drug abuse and various traits that are relevant to drug abuse, translation of those results into biological insights remains limited. With the support of a NIDA P50 grant, we are taking a complementary approach in which we study the genetic basis of numerous drug abuse-relevant behavioral traits using genetically heterogeneous rats. We ue selected rats rather than mice beacause our studies focus on complex operant and other procedures that are difficult to perform in mice. We are utilizing a unique rat population: the N/NIH heterogeneous stock (HS), which is derived from 8 deeply sequenced inbred rat strains and has been maintained as an outbred population for more than 80 generations. Because of the extensive number of accumulated generations (and thus chromosomal recombinations) HS rats allow us to implicate very small chromosomal regions in specific behavioral tasks. Drs. Terry Robinson and Shelly Flagel from The University of Michigan focus on the propensity of animals to attribute motivational value (“incentive salience”) to discrete reward cues. Dr. Hao Chen from University of Tennessee Health Sciences Center examines intravenous nicotine self-administration behavior in adolescent rats. Drs. Jerry Richards and Paul Meyer from University at Buffalo, Research Institute on Addictions are studing sensation seeking, impulsivity and cocaine-induced cue preferences. In addition to the projects directly supported by this grant, our center provides core resources and support for multiple additional projects, including two U01 grants led by Olivier George (The Scripps Research Institute) that focus on cocaine and oxycodone self-administration. In order to identify specific genes for specific behaviors, we are integrating data about behavioral, gene expression and known protein coding variants using a systems genetics approach. 1 Department of Psychiatry, University of California San Diego, La Jolla, CA, USA. Funding Support: National Instutute of Drug Abuse P50DA037844.

09:00 *Sarah Schoenrock*

Utilizing genetically diverse Collaborative Cross strains to investigate addiction-related behaviors
SPEAKER: *Sarah Schoenrock*

ABSTRACT. Substance use disorders (SUDs) are highly prevalent and result in significant burden on affected individuals, loved ones and society but few effective treatments exist. Efforts to develop effective therapies have been hampered by gaps in our knowledge about the underlying biological mechanisms that increase risk for SUDs, including genetic causes. Rodent models have proven useful for assessing the genetics of behaviors that predict propensity to use drugs, initial drug response and behaviors that emerge after repeated drug exposure and withdrawal.

Our laboratory is using a relatively new eight-way recombinant inbred mouse population, the Collaborative Cross (CC), which offers increased genetic and phenotypic diversity over traditional inbred mouse strains. We identified two CC strains that are phenotypic outliers novelty-induced locomotion - a trait previously shown to predict addiction-related behaviors. We have extensively characterized the two strains and found divergent behavioral profiles for cocaine-related behaviors including initial locomotor sensitivity to cocaine. We performed genetic mapping in a F2 intercross population derived from the CC strain with low cocaine locomotor activation and identified three significant quantitative trait loci (QTL). We are currently prioritizing candidate genes using a variety of bioinformatic tools and designing strategies to further narrow regions of interest.

Our work highlights the utility of the CC population for studying complex behaviors including those that model SUDs. We believe that this unique mouse population will be useful for identifying genes associated with increased vulnerability and/or resistance to the development of SUDs.

1Department of Genetics, 2Bowles Center for Alcohol Studies, 3Lineberger Comprehensive Cancer Center, 4Department of Psychiatry, 5Division of Pharmacotherapy and Experimental Therapeutics, Eshelman School of Pharmacy, University of North Carolina, Chapel Hill, NC, 6The Jackson Laboratory, Bar Harbor, Maine Research funding provided by grant R01MH100241 from the National Institute of Mental Health and P50 DA039841 from the National Institute on Drug Abuse, National Institutes of Health

09:30 *Zeynep Yilmaz*
Genome-Wide Association Studies of Anorexia Nervosa: A Research Update from the Anorexia Nervosa Genetics Initiative and the Eating Disorders Working Group of the Psychiatric Genomics
SPEAKER: *Zeynep Yilmaz*

ABSTRACT. This work is presented on behalf of the Anorexia Nervosa Genetics Initiative and the Eating Disorders Working Group of the Psychiatric Genomics.

1 Center of Excellence for Eating Disorders, Department of Psychiatry, University of North Carolina, Chapel Hill, North Carolina, USA

Anorexia nervosa (AN) is heritable (twin-based estimates ~50-60%) and has the highest mortality rate of any psychiatric disorder. Although the familial component of AN has been well documented, biological mechanisms contributing to AN risk are poorly understood. The objective of this study was to examine the genetic risk factors in the etiology of AN. Using 15,807 AN cases and 50,411 controls from the Anorexia Nervosa Genetics Initiative

(ANGI) and the Eating Disorders Working Group of the Psychiatric Genomics Consortium (PGC-ED), we performed a genome-wide association study (GWAS) of 7,696,751 common variants (minor allele frequency > 1%). In addition to identifying six genome-wide significant regions associated with AN risk, we observed high positive genetic correlations between AN and obsessive-compulsive disorder, major depressive disorder, and anxiety, meaning that AN shares a similar genetic architecture with these psychiatric disorders. Additionally, there were strong negative genetic correlations between AN and fasting insulin levels, body fat percentage, and BMI as well as a strong positive genetic correlation between AN and high-density lipoprotein (HDL) cholesterol, suggesting a protective role for AN risk against these unfavorable metabolic traits. Our results replicate and build on the previous PGC-ED GWAS findings (Duncan et al., 2017, Am J Psychiatry) and encourage re-conceptualization of AN as both a psychiatric and metabolic disorder.

Main Funding Support: Klarman Family Foundation, National Institute of Mental Health, Wellcome Trust Case Control Consortium 3, Lundbeck Foundation Initiative for Integrative Psychiatric Research (iPSYCH), QSkin Sun and Health Study, Riksät (Swedish National Quality Register for Eating Disorders), National Institute for Health Research (NIHR) Biomedical Research Centre at South London, Maudsley NHS Foundation Trust and King's College London.

10:00 *Alaa Koleilat*

Development of the First Pharmacotherapy for the Treatment of Usher Syndrome Type I due to Variants in MYO7A

SPEAKER: *Alaa Koleilat*

ABSTRACT. Alaa Koleilat1 Trace Christenson2, Aaron M. Lambert3, Jeffrey L. Bellah 4, Mark A. Masino3, Stephen C. Ekker1, Lisa A. Schimmenti5,6 1Department of Biochemistry and Molecular Biology, Mayo Clinic 2Mayo Clinic Microscopy and Cell Analysis Core, Mayo Clinic 3Department of Neuroscience, University of Minnesota, Twin Cities 4Department of Genetics and Development, Columbia University 5Department of Otorhinolaryngology, Mayo Clinic 6Department of Clinical Genomics, Mayo Clinic

The mariner mutant, a zebrafish model of Usher Syndrome Type 1 (USH1), is caused by homozygous premature stop codon variants in myo7aa and exhibits deafness, abnormal swimming and balance defects. We identified that this model has fewer ribbon vesicles and a smaller ribbon synapse area compared to wildtype. Currently, there are no available pharmacological treatments to improve hearing for patients with USH1. The aim of this study is to rescue the abnormal phenotypes exhibited by the mariner mutant through the use of L-type voltage gated calcium channel agonists. We hypothesize that these compounds will increase intracellular calcium and mediate the release of neurotransmitter filled vesicles at the basolateral end of the hair cell, increasing the probability that hair cell deflection will be coupled to ribbon vesicle release. Wildtype and mariner mutant swimming behavior was measured by the global change in body orientation as a function of time in response to an electric stimulus. Hearing assessments were conducted by administering various tones and recording the presence or absence of a startle response. Lastly, ribbon synapse structures were examined by electron microscopy. Our data supports that treatment with L-type voltage gated calcium channel agonists shifts mariner mutant swimming behavior towards wildtype swimming, improves startle to sound and causes a physiological change at the ribbon synapse. This data represents a significant step towards discovering compounds to treat hearing loss caused by human pathogenic variants in MYO7A.

10:30-11:00 Session Break: AM Break

AM Break

LOCATION: Mayo Civic Center 104/105

11:00-12:30 Session 14: Selected Talks III

Selected Talks 3

CHAIR: *Pamela McLean*

LOCATION: Mayo Civic Center 102/103

11:00 *John Henley*

Glucocorticoid regulation of ependymal glia and regenerative potential after vertebrate spinal cord injury

SPEAKER: *John Henley*

ABSTRACT. CM Nelson1,2, H Lee3, RG Krug3, A Kamilova1, NN Madigan4, KJ Clark3, VA Lennon2,4,5, AJ Windebank4, JR Henley1,6 Following injury, the mammalian spinal cord forms a glial scar and fails to regenerate. In contrast, spinal cord tissue of vertebrate fish regenerates and swimming movements recover. The mechanisms underlying functional regeneration are not fully understood. Here we report that the glucocorticoid pathway regulates functional neural regeneration by directly affecting ependymal glia. Cord transection in larval zebrafish (Danio rerio) causes paralysis and neural cell death, with subsequent ependymal glial proliferation, extension of bipolar glia across the lesion, and neurogenesis. Functional connectivity is restored by axons extending from spared and nascent neurons along trans-lesional glial bridges. Studies in the transgenic SR4G reporter zebrafish reveal downregulation of both the glucocorticoid receptor Nr3c1 and glucocorticoid signaling activity in ependymal glia follow injury. Functional recovery is impaired by dexamethasone (Dex) treatment, which attenuates injury-induced ependymal glial proliferation, bridging, and neural tissue regeneration, and is independent of haematopoietic-derived immune cells. Loss-of-function mutagenesis of nr3c1 reverses functional impairment by Dex. By contrast, in the adult rat, NR3C1 levels and signaling activity in ependymal glia are upregulated following spinal cord transection. The unanticipated negative regulation of neural regeneration by glucocorticoid signaling via a direct effect on ependymal glia calls into question the putative benefit of corticosteroid therapy in early management of spinal cord injury. Indeed, therapeutic down-regulation of CNS glucocorticoid receptors might improve patient outcomes. Departments of 1Neurological Surgery, 2Laboratory Medicine and Pathology, 3Biochemistry and Molecular Biology, 4Neurology, 5Immunology, and 6Physiology and Biomedical Engineering, Mayo Clinic, College of Medicine, Rochester, MN, USA 55905. Present address, AK: Vanderbilt Brain Institute, Vanderbilt University School of Medicine, Nashville, TN, USA 37232. Funding Support: NIH-NINDS (USA) R01 NS67311.

11:15 *David Ashbrook*

Sequencing the BXD family, a cohort for experimental systems genetics and precision medicine

SPEAKER: *David Ashbrook*

ABSTRACT. David G. Ashbrook1, Danny Arends2, Megan K. Mulligan1, Evan G. Williams3, Cathleen Lutz4, Alicia Valenzuela4, Casey Bohl1, Jesse Ingels1, Melinda McCarty1, Arthur Centeno1, Reinmar Hager5, Johan Auwerx6, Saunak Sen7, Lu Lu1, Kelley Harris8, Abraham Palmer9, Yu-yu Ren9, Jonathan K Pritchard10,

Andrew G. Clark11, Robert W. Williams1 1. Department of Genetics, Genomics and Informatics, University of Tennessee Health Science Center, Memphis, TN, USA 2. Lebenswissenschaftliche Fakultät, Albrecht Daniel Thaer-Institut, Humboldt-Universität zu Berlin, Invalidenstraße 42, Berlin, Germany 3. Department of Biology, Institute of Molecular Systems Biology, ETH Zurich, Zurich, Switzerland 4. Mouse Repository and the Rare and Orphan Disease Center, The Jackson Laboratory, Bar Harbor, ME USA 5. Division of Evolution & Genomic Sciences, Faculty of Biology, Medicine and Health, The University of Manchester, Oxford Road, Manchester, UK 6. Laboratory of Integrative and Systems Physiology, École Polytechnique Fédérale de Lausanne, Lausanne, Switzerland 7. Department of Preventive Medicine, University of Tennessee Health Science Center, Memphis, TN, USA 8. Department of Genome Sciences, School of Medicine, University of Washington, Seattle, WA, USA 9. Institute for Genomic Medicine, Department of Psychiatry, University of California San Diego, La Jolla, CA, USA 10. Department of Genetics, Stanford University, Stanford, CA, USA, 11. Department of Molecular Biology & Genetics, Cornell University, Ithaca, NY.

Abstract The BXD mouse genetic reference population is the most deeply phenotyped mammalian model system, with ~6000 phenotypes in GeneNetwork.org, the repository for BXD family data. GeneNetwork allows examination of complex interactions between gene variants, phenotypes from different biological levels, and environmental factors. The family consists of 152 inbred strains, each of which is a unique mosaic of alleles from the C57Bl/6J and DBA2/J, and segregate for ~4.5 million known sequence variants. Using the current genotype data, it is possible to achieve mapping precision of under ±2.0 Mb over most of the genome. We have carried out 40X sequencing of all BXD strains using a Chromium linked-read barcoding strategy. This deep sequencing of ~40 kb DNA fragments has several uses including: identification of structural variants not reliably detected using short read shotgun sequencing; identification of variants unique to each ‘epoch’ of BXD, derived in the last four decades; identification of rare spontaneous mutations; and production of the first ‘infinite marker maps’, allowing even higher precision mapping of phenotypes. We have confirmed ~4.5 million variants between the DBA/2J and C57BL/6J parents. We have aligned sequences for >50 samples and identified haplotype blocks with greater precision than was possible with microarray-based genotyping. Candidates have been identified for a phenome-wide association study. This family is an excellent resource for testing networks of causal and mechanistic relations among clinical phenotypes and millions of molecular and organismal traits, including metabolic syndrome, infection, addiction, neurodegeneration, and longevity. Full sequencing of all lines will only increase its usefulness.

11:30 *Nicole Nelson*

A mechanism of public misunderstanding of animal behavior genetics research

SPEAKER: *Nicole Nelson*

ABSTRACT. Nicole C. Nelson

Media coverage of animal behavior genetics research often misrepresents the complex etiology of behavioral disorders, typically portraying these disorders as arising from the effects of single genes rather than multiple genetic and environmental causes. This paper explores one mechanism through which these misrepresentations develop: the separation of knowledge about environmental contributors to behavior from knowledge about genetic contributors during the publication process. Using

ethnographic research from animal behavior genetics laboratories, I describe how researchers informally acquire knowledge about how environmental factors (noise, smells, other stressors) alter mouse behavior, and how the designation of this knowledge as “methodological” or “tacit” limits its circulation outside of the laboratory. The limited circulation of environmental knowledge, as compared to the much wider circulation of genetic findings, contributes to a distorted public perception of disease etiology. As a counterfactual, I present media coverage of published instances of environmental knowledge (Crabbe, Wahlsten and Dudek’s well-known 1999 paper on mouse behavior and interactions with the laboratory environment), demonstrating that alternative media narratives about behavioral disorders are possible. This research has important implications for the way that animal behavior geneticists communicate their findings publically, suggesting that more effort to circulate environmental knowledge is needed to counterbalance genetic knowledge.

11:45 *Max Melin*

Localizing Origins of Essential Tremor Phenotype in a Novel Mouse Model

SPEAKER: *Max Melin*

ABSTRACT. Melin MD1, Zhou M1, Sudhof TC1

Essential tremor (ET) is the most common movement disorder, affecting more than 200,000 US citizens each year. However, our understanding of this disease is still very limited, therapies are either invasive or have minimal impact, and we have very few predictive and accurate animal models to study. It has been thought that the primary cause of essential tremor lies in the inferior olivary nucleus, but more recent studies suggest that the cerebellum plays an important role in the tremor phenotype. Previous work in the Sudhof Lab has shown that the knockout of the Synaptotagmin 2 (Syt2) calcium sensor in parvalbumin-positive neurons produces and Essential Tremor-like behavior in mice. Using this novel genetic mouse model we explored which brain region underlies the etiology of essential tremor via spatial and cell type specific knockout of Syt2. Results from this study indicate that the cerebellum plays an active role in generating tremor in this mouse model. Furthermore, we showed that aberrant functioning of the deep cerebellar (fastigial, dentate, and interposed) nuclei contribute to the ET phenotype. The findings from this study indicate that dysfunction of the cerebellum may indeed be responsible for the manifestation of ET, and it validates this Syt2-PV KO mouse as a more accurate model of ET than previous models. Thus this study offers a novel method of studying ET, as well as a window into the possible brain regions and cell type that could be targeted for more effective treatment.

1Department of Molecular and Cellular Physiology, Stanford University. Stanford, California. Funding: Stanford BioX Undergraduate Research Major Grant.

12:00 *Alexandra Goetjen*

GABRA2 genetic variants and chromosome conformation in induced pluripotent stem cell-derived neural cultures

SPEAKER: *Alexandra Goetjen*

ABSTRACT. Goetjen AM[1, 2], Clinton KL[2], Lieberman R[2, 3], & Covault J[1-3]

[1]University of Connecticut Graduate School Biomedical Sciences PhD Program: Genetics and Developmental Biology; [2]UConn Health Department of Psychiatry; [3]University of Connecticut Graduate School Biomedical Sciences PhD Program: Neuroscience

Approximately 8.5% of American adults are afflicted by either moderate or severe alcohol use disorder (AUD), defined as excessive alcohol use within the last twelve months that impedes the safety of oneself and others, while being unable to reduce one's drinking. The Collaborative Study on the Genetics of Alcoholism used linkage analysis to suggest, in European Americans, a significant association between alcohol dependence and a 140kb haplotype block in GABRA2. Synonymous SNP rs279858 tags this haplotype block, and has a minor allele frequency of 0.45. In addition to AUD, rs279858 has been associated with a number of neuropsychiatric phenotypes including comorbid illicit drug use and childhood conduct disorder. Neuro-endophenotypes such as increased activation of the insular cortex and nucleus accumbens in reward anticipation and differential activation of the ventral tegmental area and medial frontal cortex in response to alcohol cues are also associated with this haplotype block. The chr4p12 locus codes for γ1, α2, α4, and β1 GABA receptor subunits; iPSC lines carrying the minor allele at rs279858 have reduced expression not only of GABRA2, but the other three genes within this cluster. Virtual chromatin conformation capture (4C) data supports this hypothesis of cis regulation of GABA gene expression. Identification and characterization of allele-specific variants involved in mediating long-range intrachromosomal interactions in this locus is a step forward in the process of elucidating genetic risk variants for AUD and subsequently developing more-specific therapies for those at increased genetic risk.

12:15 *Glen Howel Acosta*

Genetic modifiers of motor decline symptoms of Alzheimer's disease in a novel transgenic murine panel

SPEAKER: *Glen Howel Acosta*

ABSTRACT. Developing therapeutics to address cognitive symptoms in Alzheimer's disease (AD) remains a priority. However, non-cognitive symptoms significantly impact quality of life for patients and caregivers. Our recent work suggests that genetic mechanisms underlying cognitive decline are distinct from non-cognitive symptoms (e.g. decline in muscle strength). Understanding underlying causes of non-cognitive symptoms in AD is important, as these will likely require therapeutic attention. Here we use a mouse genetic reference panel that better models AD in humans to identify genetic mechanisms regulating grip strength--a measure of frailty that is associated with aging and increased risk of AD in humans. Decline in grip strength was measured from 6 to 14m in a genetically diverse panel of AD transgenic mice, and their age-matched nontransgenic controls (Ntg). Both AD and Ntg mice showed age-related decline in grip strength; however, the magnitude of decline was dependent on background strain (n=25 strains). Quantitative trait loci (QTL) mapping identified a locus on mouse chromosome 12 (Chr12: 57.8-65.0Mb) associated with variation in grip strength in the AD population that differed from Ntg controls (Chr6: 17.5-28.2Mb). These data suggest genetic modifiers of motor function may differ in the AD versus normal aging, which was supported by weak correlation between grip strength in AD vs. Ntg strains (r 2=0.14, p=0.08). Combined with ongoing work, we demonstrate that genetic mechanisms modulating cognitive and motor-related decline in AD are distinct. We are using existing gene expression

data and variant prediction to identify positional candidates for validation and potential therapeutic targets. 1. The Jackson Laboratory, Bar Harbor, Maine, USA 2. University of Tennessee Health Science Center, Memphis, TN. Funding Support: NIA 1 R01 AG057914-01 to C.C.K., NIA 1 R01 AG054180-01A1 to C.C.K

12:30-13:30 Session L4: Lunch

Lunch

13:30-15:30 Session 15: Symposium 5

Neural Basis of Impulsivity in Addiction and Psychiatric Disorder

CHAIR: *Doo-Sup Choi*

LOCATION: Mayo Civic Center 102/103

13:30 *Ja-Hyun Baik*

Role of dopamine D2 receptors in impulsive behavior
SPEAKER: *Ja-Hyun Baik*

ABSTRACT. Ja-Hyun Baik Molecular Neurobiology Laboratory, Dept. of Life Sciences, College of Life Sciences and Biotechnology, Korea University, Seoul 02841, South Korea

Impulsive behavior—the tendency to act in premature, risky, or inappropriate ways, without consideration of the consequences—is often associated with psychiatric conditions such as drug addiction, as well as eating and personality disorders. Because of the complex aspects of impulsivity, neural correlates of impulsivity have not been well characterized yet despite its clinical and social importance. Increasing evidence from both human and animal studies suggests the importance of dopaminergic regulation in the pathophysiology of impulsive behavior. We now show that the absence of the D2 dopamine receptor (D2R) increases impulsive behavior in mice, whereas restoration of D2R expression specifically in the central amygdala (CeA) of D2R knockout mice normalizes their enhanced impulsivity. I will present recent findings obtained in our laboratory in the analysis of the role of D2R for the control of impulsive behavior using genetic, anatomical and optogenetic manipulations. Our identification of the key contribution of D2R-expressing neurons in the brain circuit to the control of impulsive behavior may provide a basis for the development of new approaches to the management of neuropsychiatric disorders associated with impulsivity.

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14:00 *Luc Maroteaux*

Serotonin 2B receptors in mesoaccumbens dopamine pathway in impulsivity and cocaine responses
SPEAKER: *Luc Maroteaux*

ABSTRACT. Luc Maroteaux1, Stéphane Doly1, Emily Quentin1, Arnauld Belmer1

A functional stop codon in the human 5-HT2B-receptor gene (HTR2B) releases impulsive behavior. Similarly, Htr2B knockout (Htr2B−/−) mice displayed more impulsive choice, sought novelty, and were more active after receiving a D1 dopamine receptor agonist, fully consistent with a role of serotonin (5-HT) in the

rewarding effects of psychostimulant. Long-term inactivation (chronic pharmacological blockade or genetic ablation) of the 5-HT2B receptor is required for novelty- and cocaine-induced hyperlocomotion, while acute pharmacologic inhibition had no effect. Interestingly, in cocaine-injected Htr2B-/- mice, reduced release of DA blunts activation of medium spiny neurons of the NAcc shell, without modification of reactivity of the dorsal striatum. We also observed that 100% of the retrogradly labeled DA neurons from NAcc to VTA do express 5-HT2B receptors that modulate their activity by controlling firing rate. Local recombination of Htr2B gene in DA neurons recapitulates novelty- and cocaine-induced hyperlocomotion seen in Htr2B-/- mice or chronic antagonist-treated mice. A delay appears thus necessary to allow neuroadaptations responsible for these apparent paradoxical responses, the increased cocaine response being only an indirect consequence of the reduced DA tone observed in NAcc and downstream adaptation. Our findings in mice provide evidence for a role for the 5-HT2B receptor in mesolimbic pathways, with a lack of 5-HT2B receptor representing a source of permanent 5-HT-dependent DAergic system hypofunction in NAcc. This functional deficit may represent a susceptibility factor for both impulsive and addictive behavior; two behaviors that have been tightly interwoven, and may explain both the impulsivity and cocaine vulnerability of Htr2B−/− mice.

1INSERM, U839, Institut du Fer à Moulin, 17 rue du du Fer à Moulin 75005 Paris, France. Funding Support: Fondation pour la Recherche sur le Cerveau, the Fondation de France, the Fondation pour la Recherche Médicale "Equipe FRM DEQ2014039529", the French Ministry of Research (Agence Nationale pour la Recherche ANR-12-BSV1-0015-01 and the Investissements d'Avenir programme ANR-11-IDEX-0004-02).

14:30 *Kathryn Cunningham*

Clues into Cues: Serotonergic Circuits Engaged in Impulsivity and Cue Reactivity

SPEAKER: *Kathryn Cunningham*

ABSTRACT. KA Cunningham,1 FG Moeller,2 Ed Boone3, Qin Wang,3 NC Anastasio1

1Center for Addiction Research and Department of Pharmacology and Toxicology, University of Texas Medical Branch, Galveston, Texas, USA; 2Institute of Drug and Alcohol Studies, and Department of Psychiatry, Virginia Commonwealth University, Richmond, Virginia, USA; 3Institute of Drug and Alcohol Studies, and Department of Statistics, Virginia Commonwealth University, Richmond, Virginia, USA

Cocaine use disorder (CUD) remains a disturbing chronic health problem as well as an individual and societal burden in the United States. A 61% increase in adults initiating cocaine use (2013-2015) coupled with the highest cocaine overdoses reported since 2006 reveal alarming trends (www.samhsa.gov/data/sites/default/files/report_2736/ShortReport-2736.html). A hallmark of CUD is “continued drug use despite adverse consequences,” which aligns with the definition of impulsivity, a predisposition toward rapid unplanned reactions to stimuli without regard to negative consequences. Impulsivity is interlocked with cue reactivity which is defined as the attentional orientation toward drug-associated stimuli that predict reward in humans and rodents. We report translational studies employing gene-mediated viral vector targeting strategies in rodents and analyses of structural and effective connectivity in fMRI studies in humans that interrogate the neurocircuitry engaged in impulsive action and the response bias toward cocaine-associated cues. Our data support the contention that medial prefrontal cortex (mPFC) □

nucleus accumbens shell (NAcSh) circuitry in rodents and the anterior cingulate cortex in humans are brain circuits involved in the overlap of impulsive action and cocaine cue reactivity. Furthermore, we have uncovered that impulsive action and cue reactivity are mechanistically-linked to disrupted serotonin (5-HT) signaling through a 5-HT2A receptor (5-HT2AR) and 5-HT2CR interaction within this circuitry. Employing integrative Bayesian Analyses of Neuroimaging-Genetics methods, we recently demonstrated that single nucleotide polymorphisms (SNPs) of the HTR2A and HTR2C are interrelated with cocaine use, impulsivity, and altered brain structural and functional connectivity on fMRI in humans. This interaction may be relevant to our discovery that CUD subjects carrying the same HTR2C SNP exhibit the highest cue reactivity. Given the importance of cue reactivity and impulsivity in the risk for relapse in CUD, the outcomes of these studies contribute a greater appreciation of the serotonergic neurocircuitry underlying these constructs and suggest new concepts in the treatment of CUD.

15:00 *Doo-Sup Choi*

Striatal Adenosine A2A Receptor Regulates Impulsivity, Goal-Directed Alcohol Seeking Behaviors

SPEAKER: *Doo-Sup Choi*

ABSTRACT. Hong SI1, Starski P1, Choi S1, Kang S1, Choi DS1.

Adenosine A2A receptor (A2AR) is abundantly expressed in the striatopallidal neurons in the striatum and has been implicated in increased alcohol drinking through enhancement of goal-directed behaviors and impulsivity. Using mice that were exposed to differential reward of low rate (DRL) schedules during Pavlovian conditioning, second-order schedule discrimination, and the 5-choice serial reaction time task (5-CSRTT), we demonstrate that deficits of A2AR function promote impulsive responses. In a binge-drinking paradigm, mice were exposed to vaporized ethanol for 4 h in every 4th day displayed increased premature responses during the challenge tests, suggesting that binge alcohol consumption increase impulsivity through dampening A2AR. Next, we investigated whether the A2AR in the dorsomedial striatum (DMS) contributes to goal-directed behavior in mice exposed to vaporized ethanol for 16 h per day during 4 days. Our results show that ethanol-exposed mice elicited anxiety-like behavior in elevated plus maze test without affecting working memory in y-maze test. During acquisition of instrumental behavior training in C57BL/6J mice, CGS21680 (0.3 mg/kg, i.p.), A2AR agonist, and optogenetic stimulation of A2AR-expressing neurons in the DMS abolished goal-directed behavior in response to 10% ethanol, whereas ZM241385 (20 mg/kg, i.p.), A2AR antagonist, did not impede goal-directed behavior under valued condition. Interestingly, A2AR activation prevented voluntary ethanol consumption in operant chamber even after 24 h following CGS21680 (0.3 mg/kg, i.p.) injection. Taken together, our results demonstrate that the DMS A2AR regulates impulsivity, goal-directed and alcohol seeking behaviors.

1Department of Molecular Pharmacology and Experimental Therapeutics, Mayo Clinic College of Medicine, Rochester, Minnesota, 55905, USA. Funding Support: the Samuel C. Johnson for Genomics of Addiction Program at Mayo Clinic, the Ulm Foundation, and National Institute on Alcohol Abuse and Alcoholism (AA018779).

15:30-16:00 Session Break: PM Break

PM Break

LOCATION: Mayo Civic Center 104/105

16:00-17:00 Session 16: Keynote Address

Keynote Address

CHAIR: *Karl Clark*
LOCATION: Mayo Civic Center 102/103

16:00 *Edward Burton*

Phenotype-based drug discovery for neurodegenerative diseases using automated neurobehavioral profiling in genetic zebrafish models
SPEAKER: *Edward Burton*

ABSTRACT. Progressive supranuclear palsy (PSP) is a common and incurable neurodegenerative disease associated with prominent motor, oculomotor and cognitive-behavioral deficits, and a poor prognosis for functional independence and survival. The pathology of PSP is characterized by neuronal loss and accumulation of abnormal aggregates of the 4-repeat isoform of the microtubule-associated protein tau (4R-tau), which is strongly implicated in pathogenesis by convergent lines of genetic evidence. Phenotype-based drug discovery has previously identified many first-in-class drugs and is particularly applicable when therapeutic targets are unclear. Our current work is directed towards developing effective treatments for PSP and related tauopathies, by exploiting the potential of zebrafish genetics and high-throughput automated neurobehavioral phenotyping to identify chemical modifiers of pathogenesis, unbiased by preconceptions about mechanism of action. We developed a zebrafish tauopathy model that replicates many features of PSP including: impaired survival; neurobehavioral deficits; oculomotor abnormalities; somatodendritic accumulation of hyperphosphorylated 4R-tau; and neurodegeneration. Using automated neurobehavioral profiling to detect phenotypic rescue, we recently completed a pilot chemical modifier screen, identifying several small molecules that rescue the neurobehavioral phenotype of the zebrafish tau model. Validation of these ‘hit’ compounds and verification of their targets is ongoing, but preliminary studies suggest unexpected modes of action, thereby justifying a phenotype-driven approach. We conclude that automated behavioral phenotyping to detect chemical rescue in genetic disease models in vivo may have general application in drug discovery for neurological disease. The zebrafish tauopathy model will also be useful for evaluating the functional effects of putative genetic PSP modifiers identified in GWAS studies.

18:00-18:15 Session 17: Trolley Bus Loading- Outside Civic Center

18:15-19:00 Session 18: Travel and Reception

Travel and Reception

19:00-23:00 Session : Banquet: Four Daughters Vineyard & Winery

Banquet: Four Daughters Vineyard & Winery

CHAIR: *Stephanie Ferguson*
LOCATION: Four Daughters Winery

20:30-23:00 Session : Return Transportation

Return Transportation. Trolleys will leave about every 30 minutes from 8:30 until the close of the event at 11PM.