Neuropsychopharmacology (2017) 42, S294–S475

www.neuropsychopharmacology.org

Poster Session II
Palm Springs, California, December 3-7, 2017

Sponsorship Statement: Publication of this supplement is sponsored by the ACNP.
Individual contributor disclosures may be found within the abstracts. Part 1: All Financial Involvement with a pharmaceutical or biotechnology company, a company providing clinical assessment, scientific, or medical products or companies doing business with or proposing to do business with ACNP over past 2 years (Calendar Years 2014–Present); Part 2: Income Sources & Equity of $10,000 per year or greater (Calendar Years 2014 - Present): List those financial relationships which are listed in part one and have a value greater than $10,000 per year, OR financial holdings that are listed in part one and have a value of $10,000 or greater as of the date of disclosure; Part 3: Financial Involvement with a pharmaceutical or biotechnology company, a company providing clinical assessment, scientific, or medical products or companies doing business with or proposing to do business with ACNP which constitutes more than 5% of personal income (Calendar Years 2014 - Present); Part 4: Grants from pharmaceutical or biotechnology company, a company providing clinical assessment, scientific, or medical products directly, or indirectly through a foundation, university, or any other organization (Calendar Years 2014 - Present); Part 5: My primary employer is a pharmaceutical/biotech/medical device company.
Asterisks in the author lists indicate presenter of the abstract at the annual meeting.

T1. The Senescent-Associated Secretory Phenotype: A Marker of Enhanced Molecular Senescence Changes in Late-Life Depression

Breno Diniz*, Charles Reynolds, Etienne Sibille, Meryl Butters

University of Texas Health Science Center at Houston, Houston, Texas, United States

Background: Late-Life Depression (LLD) is common and associated with higher risk of age-related negative health outcomes. The mechanisms of this association are not clear, but may involve the activation of biological pathways related to senescence. This study aims to investigate whether a systemic molecular pattern associated with aging (senescent-associated secretory phenotype [SASP]) is elevated in adults with late-life depression (LLD), compared with never-depressed elderly comparison participants.
Methods: We included 111 older adults (80 with LLD and 31 comparison participants) in this study. A panel of 22 SASP-related proteins was extracted from a previous multiplex protein panel performed in these participants. We conducted a principal component analysis to create the SASP index based on individual weights of each of protein.
Results: LLD showed a significantly higher SASP index compared with comparison participants, after controlling for potential confounders ($F(1,98) = 7.3$, $p = 0.008$). Correlation analyses revealed that the SASP index was positively correlated with age ($r = 0.2$, $p = 0.03$) and CIRS score ($r = 0.27$, $p = 0.005$), and negatively correlated with information processing speed ($r = -0.34$, $p = 0.001$), executive function ($r = -0.27$, $p = 0.004$) and global cognitive performance ($r = -0.28$, $p = 0.007$).
Conclusions: this is the first study to show that a set of proteins (i.e., SASP index) primarily associated with cellular aging is abnormally regulated and elevated in LLD. These results suggest that individuals with LLD display enhanced aging related molecular patterns that are associated with higher medical comorbidity and worse cognitive function. Finally, we provide a set of proteins that can serve as potential therapeutic targets and biomarkers to monitor the effects of therapeutic or preventative interventions in LLD.
Keywords: Late-Life Depression, Aging, Biomarkers
Disclosure: Nothing to Disclose.

T2. Postpartum Effects on the Dopamine System of Female Rats

Millie Rincón-Cortés*, Anthony Grace

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: The period after childbirth (i.e. postpartum period) is a time of elevated risk for the development of affective disorders (Stowe and Nemeroff, 1995). Indeed, the highest rates of anxiety and depression occur during the first few weeks, months or year postpartum compared with other times in a woman's life (Pawluski et. al 2017). In accordance, animal models of postpartum depression have also reported time-dependent effects on depressive-like behavior and anhedonia (Brummelte and Galea, 2010). In rodents, parity (i.e. the condition of having borne offspring) induces changes in DA-mediated behavioral responses, which may reflect some influence on DA neurotransmission (Hucke et. al, 2001). However, the neurobiological underpinnings of this increased female susceptibility to depression during the postpartum period remain poorly understood.
The dopamine (DA) system has traditionally been associated with anhedonia, the inability to derive pleasure from normally rewarding stimuli, and has been repeatedly implicated in the pathophysiology in depression. A causal link between a hypofunctioning DA system (i.e. decreased DA neuron activity) and stress-induced depression-related behaviors (i.e. anhedonia, despair, anxiety) has been demonstrated in animal models (Tye et. al, 2013; Chang and Grace, 2014), with females showing greater effects (Rincón-Cortés and Grace, 2017). Surprisingly, little is known about DA system function in females following reproductive experience, including parity (i.e. the condition of having borne offspring). To this end, we assessed anxiety-like behavior, social motivation and DA system function in virgin and during the early postpartum period in females.
Methods: Virgin and early postpartum (i.e. 1 day post-partum, 3 days postpartum) female rats were tested for anxiety-like behavior in the elevated plus maze (EPM) and

social motivation in the social approach test (SAT). Single-unit recordings of VTA DA neurons were conducted the day following behavioral testing and 3 parameters were measured: i) the number of spontaneously active DA cells (i.e. population activity), ii) basal firing rate and iii) firing pattern (i.e. the percentage of spikes firing in bursts).
Results: Postpartum female rats exhibited a reduction in the percentage of open arm entries ($p < 0.01$) and the percentage of time spent in the open arms ($p < 0.05$) of the EPM compared with virgin female rats at 1-day post-partum. Postpartum female rats tested at both time points (1-day, 3-days) exhibited reduced social motivation ($p < 0.001$), as indexed by less sniff time of a younger, same-sex animal in the SAT. Postpartum females exhibited an attenuation of DA population activity, as indexed by a reduction in the number of cells per electrode track in the VTA, compared with virgin rats ($p < 0.001$) but no differences in firing rate ($p = 0.42$) or the percentage of spikes occurring in bursts ($p = 0.53$).
Conclusions: Collectively, our findings suggest that parity can drive changes in affective behavior (i.e. increases anxiety-like behavior and reduces social motivation during the early postpartum period and that these behavioral changes are associated with an attenuation of DA activity within this period.
Keywords: Anxiety, Social Behavior, Postpartum Depression, Dopamine, Electrophysiology
Disclosure: Nothing to Disclose.

T3. Multifocal Transcranial Direct Current Stimulation Targeting the Medial Prefrontal Cortex Improves Exposure-Relevant Learning Among Obsessive-Compulsive Disorder Patients

Thomas Adams*, Benjamin Kelmendi, Jamilah George, Stephen Kichuk, Christopher Pittenger

Yale University School of Medicine, New Haven, Connecticut, United States

Background: Exposure and response prevention (ERP) is the psychosocial treatment of choice for obsessive-compulsive disorder (OCD), but it has several important limitations. ERP is thought to reduce anxious reactivity via fear extinction learning; an inhibitory process that is primarily mediated by a brain circuit that includes the medial prefrontal cortex (mPFC), hippocampus, and amygdala. After extinction occurs the original fearful meaning, which is primarily encoded in the amygdala, is inhibited, particularly by the mPFC. This raises the exciting possibility that modulation of extinction circuitry – particularly LTP-like plasticity processes in the mPFC – might augment response to therapeutic exposure. Here we present data from a nearly complete study that aims to use multifocal transcranial direct current stimulation (tDCS) targeting the mPFC to enhance therapeutic extinction learning during an ERP challenge among OCD patients.
Methods: Subjects diagnosed with OCD are randomly assigned to receive 20 minutes of active (n = 8) or sham (n = 8) multifocal tDCS targeting the mPFC (anode over Fpz, cathods over AF7, AF8, F3, F4, and Fz, 1.5mA, 30 sec. ramp in/out); computational modelling of electrical fields suggests the direct effects of this montage are concentrated in the mPFC, including the ventral mPFC. tDCS is followed by five 10-minute trials of individualized ERP to test the acquisition of therapeutic extinction learning. Subjects return 24 hours later to complete five additional exposure trials to test the recall of therapeutic learning. Subjective units of distress (SUDS) are assessed every minute before, during, and after the exposure task.
Results: tDCS had no significant effects on pre-exposure (but post-tDCS) SUDS levels and did not affect SUDS ratings during the first minute of the first exposure trials (ps > .05), suggesting it did not have a non-specific anxiolytic effect. However, data suggest that tDCS significantly enhanced acquisition of therapeutic extinction learning. Mixed ANOVA showed that, over the course of the five exposure trials on day 1 of the ERP challenge, subjects who received active-tDCS reported greater SUDS reductions than subjects who receive sham tDCS, $F(1.99, 27.79) = 3.90$, $p < .05$, $\eta 2p = .22$. Said otherwise, subjects in the active tDCS condition reported 67.94% reductions SUDS whereas subjects who received sham tDCS reported 18.34% reductions in SUDS, $F(1, 14) = 13.04$, $p < .01$, $d = 1.93$. Exploratory mixed ANOVAs suggest that active tDCS had a minimal effect on early phase extinction learning (average SUDS change across trials 1 to 2), $F(1, 6) = .65$, $p = .45$, but had a significant effect on late phase extinction learning (trials 4 to 5), $F(1, 6) = 11.34$, $p = .02$. Contrary to our prediction, tDCS did not appear to affect extinction recall. In fact, subjects who received active tDCS evinced marginally poorer recall compared to subjects who received sham tDCS, $F(1, 14) = 3.63$, $p = .08$, $\eta 2p = 21$.
Conclusions: Our findings suggest that multifocal tDCS targeting the mPFC prior to individualized exposure can improve acquisition of therapeutic extinction learning among OCD patients. Data suggest the effect is specific to the acquisition of therapeutic learning. Our ERP challenge lasts 55 minutes and basic research has shown that the after effects of tDCS on LTP-like plasticity last approximately 60 minutes. The direct effects of tDCS on the brain may have substantially diminished before the consolidation window opened. Future research should explore if stimulation during or after exposure improves consolidation and subsequent recall of therapeutic learning. Our preliminary analyses should be interpreted cautiously given the sample size. Nonetheless, the present findings are promising and justify further exploration of this adjunctive intervention for OCD and other anxious or stress-related conditions that are treated with therapeutic exposure.
Keywords: CBT, TDCS, Obsessive-Compulsive Disorder (OCD), Exposure Therapy
Disclosure: Nothing to Disclose.

T4. Anterior Insula Modulation by Attention Shifting in Social Anxiety Disorder

Elizabeth Duval*, Sonalee Joshi, Stefanie Russman Block, James Abelson, Israel Liberzon

University of Michigan Health System, Ann Arbor, Michigan, United States

Background: Social Anxiety Disorder (SAD) is associated with abnormal function in brain regions implicated in

emotional processing and attentional control. Multiple studies have investigated attention training as a way to address potential deficits in attention modulation of emotion, with some reports of decreased symptom severity, physiological reactivity, and attention bias toward social threat. However, many negative findings are also reported, and the specific mechanisms by which attention training meaningfully impacts SAD symptoms and associated neural function are yet to be identified. The purpose of this study was to investigate the neural mechanisms underlying attention modulation of emotion in SAD, and how these processes may be impacted by attention training paradigms.

Methods: SAD participants ($N = 29$) were randomly assigned to one of three attention training conditions: toward threat, away from threat, control. Healthy participants ($N = 12$) were trained away from threat. Four one-hour sessions of training were delivered using a modified dot-probe paradigm. Participants completed pre- and post-training symptom assessments and a shifted attention emotion modulation task during fMRI scanning.

Results: There were no training effects on SAD symptoms at the group level (ps $>$ .16). Prior to training, fMRI results revealed a significant difference between SAD and healthy subjects in anterior insula activation during attention modulation, $t(39) = 2.78$, $p = .008$, suggesting that SAD participants were less effective at engaging in attention modulation compared to healthy subjects. Across all attention training conditions, we observed a significant group by time interaction in the same region of the anterior insula, $F(1, 39) = 4.72$, $p = .036$, suggesting that SAD participants improved their attention modulation abilities over time relative to healthy participants. Interestingly, changes in anterior insula activation indicative of improvements in attention modulation were associated with improvements in symptoms across all participants ($r = .43$, $p = .005$) and in the SAD group ($r = .45$, $p = .021$). While no training effects reached significance, there was a trend-level time by training interaction in the anterior insula, $F(2, 26) = 3.2$, $p = .057$, suggesting that SAD participants trained both toward and away from threat improved their attention modulation abilities over time, compared to those in the control training condition.

Conclusions: These findings suggest that anterior insula activation differs in SAD compared to healthy subjects when shifting attention to modulate emotion. The degree of effective modulation in anterior insula is associated with a decrease in SAD symptoms over time. While attention training may improve attention modulation in the anterior insula, no training effects reached significance in the brain regions examined, nor did training impact SAD symptoms over time. Additional studies are needed to determine whether current attention training approaches are in fact therapeutically useful in SAD, and whether further dissection of their neural mechanisms can be used to produce more consistently effective treatments.

Keywords: Social Anxiety Disorder, Functional MRI (fMRI), Insula, Attention, Emotion Modulation

Disclosure: Nothing to Disclose.

T5. The FAAH C385A Variant Influences Baseline and Stress-Induced Changes Circulating in Endocannabinoids

Leah Mayo*, Anna Asratian, Johan Linde, Lovisa Holm, Niclas Stensson, Bijar Ghafouri, Markus Heilig

Linkoping University, Linköping, Sweden

Background: The endocannabinoid (eCB) system has been proposed to be a "stress buffer," playing an integral part in regulating the stress response. In animal models, stress rapidly decreases the eCB anandamide (AEA), which is believed to contribute to the manifestation of the stress response. However, human research in this area is sparse and often conflicting (Hill et al., 2009; Dlugos et al., 2012; Spagnolo et al., 2016); thus, it is uncertain how stress influences circulating eCBs and other N-acylethanolamines (NAEs) in humans.

Nonetheless, it is clear from preclinical animal research that eCBs and AEA in particular are implicated in the stress response. For instance, elevating AEA levels via inhibition of its main metabolic enzyme, fatty acid amide hydrolase (FAAH), protects against many of the negative effects of stress (Haller et al., 2009; Hill et al., 2009; 2010). While there are no FAAH inhibitors currently approved for human use, insights into the consequences of elevated AEA levels in humans have come from the functional FAAH C385A variant. This single nucleotide polymorphism results in elevated levels of AEA (Sipe et al., 2010) likely via enhanced FAAH sensitivity to proteolytic degradation. It has been reported that carriers of this variant demonstrate decrease threat responding (Hariri et al., 2009; Gunduz-Cinar et al., 2013) and stress responsivity (Spagnolo et al., 2016).

Here, we aimed to determine whether FAAH C385A variation influenced baseline levels of eCBs and other NAEs, as well as stress-induced changes in these molecules. In addition, we asked whether variation at this locus influenced other indices of stress responsivity, such as cortisol response, subjective stress, and physiological measures.

Methods: Using a prospective genotyping approach, we recruited 25 of each FAAH C385A genotype (C/C, A/C, A/A) and exposed them to a 2-day paradigm consisting of a stress task and control task completed on separate days. Prior to the tasks, participants filled out questionnaires regarding affect (the Positive and Negative Affect Scale; PANAS) and trait anxiety (STAI-T). Participants were then fitted with sensors for physiological recordings and an indwelling venous catheter to collect blood samples for eCB and cortisol analysis.

The stress procedure (the Maastricht Acute Stress Task, MAST; Smeets et al., 2012) was a 10min task that consisted of alternating trials of hand immersion in ice cold water and mental arithmetic performed aloud in the presence of an experimenter who provided negative verbal feedback. The control task consisted of hand immersion in room temperature water and simple counting. Questionnaires regarding affect and subjective stress response were also completed. Blood sample were collected prior to (-10, 0min) and after (+10, +20, +30min) stress/control procedures.

Serum was extracted from the blood samples and subjected to LC-MS/MS for analysis of eCBs and NAEs.
Results: Preliminary results suggest that the FAAH C385A variant (A-allele) is associated with elevated baseline levels of AEA, PEA, and OEA, but not 2-AG. Stress appears to reduce circulating AEA, though this was true only for C/C and A/C individuals and not A/A participants. There were no differences between genotype groups in levels of 2-AG or other NAEs following stress.
Stress increased negative affect (measured via the PANAS) and subjective stress, but neither factor was influenced by genotype. Subjective stress did differ between sexes, with females reporting that the task was more "stressful", but this was consistent across genotypes. Personality measures (e.g. trait measures of anxiety and positive and negative affect) were generally consistent across genotype groups.
Conclusions: Elevated levels of AEA conferred via the FAAH C385A A-allele appear to protect against stress-induced decreases in AEA. While baseline and stress-induced changes varied across genotype groups, there was no difference in subjective stress or negative affect. Future work will explore the potential benefits of elevated AEA levels in regard to stress responsivity.
Keywords: Anandamide, FAAH, Acute Stress
Disclosure: Nothing to Disclose.

T6. A Wearable Morning Light Therapy for Post-Traumatic Stress Disorder

Alyson Zalta*, Zerbrina Valdespino-Hayden, Mark Pollack, Helen Burgess

Rush University Medical Center, Chicago, Illinois, United States

Background: Although several front-line psychotherapies and pharmacotherapies exist for posttraumatic stress disorder (PTSD), evidence suggests that a significant number of individuals with PTSD fail to receive treatment let alone a therapeutic dose. Uptake of these treatments is poor for a variety of reasons including stigma, avoidance, unpleasant side effects, and accessibility. Thus, novel treatments are needed that are safe, readily available, convenient, and appealing to patients. Light treatment has great potential as a novel non-invasive, low risk treatment for PTSD. Evidence suggests that later circadian timing may exacerbate PTSD-related sleep disturbance. Additionally, research suggests that some circadian photoreceptors project directly to the amygdala, which is implicated in PTSD. We are testing a novel wearable light treatment device, the Re-timer®. We also created a placebo device by dimming the light intensity with neutral density filters. The goal of this study is to determine the feasibility, acceptability and efficacy of a wearable light treatment in a sample of individuals experiencing probable PTSD.
Methods: In this ongoing study, individuals with probable PTSD (Criterion A trauma + PTSD Checklist for DSM-5 [PCL-5] $>$ 33) are randomized to self-administer a 4 week, daily 1-hour morning light treatment or placebo. Light treatment begins at each participant's average wake time. Treatment adherence is objectively assessed with an accelerometer and photosensor placed on the inside of the Re-timer®. Weekly visits are conducted to assess outcome measures (PCL-5; Patient Health Questionnaire – 9 [PHQ--9]; Insomnia Severity Index [ISI]; Pittsburgh Sleep Quality Index [PSQI]) and provide feedback on treatment adherence.
Results: We currently have preliminary results for 12 participants (7 active, 5 placebo) who have completed all study visits to date including 8 women and 4 men with an average age of 48.2 (SD = 10.6). Based on the accelerometer and photosensor data, participants completed part or all of the morning light treatment on an average of 19.7 of 28 days of treatment (SD = 7.3 days, range = 9 to 27 days) and received an average of 33.8 minutes per day (SD = 6.0 minutes) on days they engaged in treatment. Treatment adherence did not significantly differ based on treatment group and overall, those in the placebo group had higher adherence based on number of days and minutes per day. Moreover, post-treatment evaluation showed that the placebo was credible. Overall, participants who received the active treatment demonstrated larger improvements on all outcome measures from pre- to post-treatment compared to participants who received placebo. Effect sizes differed across outcomes with a large difference between groups for changes in PTSD symptoms (d = .87) and depression symptoms (d = .81), and a small to moderate differences between groups for changes in insomnia (d = .42) and sleep quality (d = .21). Exploratory analyses showed that changes between the groups in PTSD symptoms were largely driven by changes in the cognitive / mood (d = 1.05) and hyperarousal (d = .78) symptoms rather than re-experiencing (d = .51) and avoidance (d = .05) symptoms. Notably, the average PCL-5 change in the morning light treatment group was greater than 10 points (mean change = 12.6 points), indicating a clinically meaningful change in PTSD symptoms from pre- to post-treatment.
Conclusions: Our preliminary evidence suggests that a wearable morning light therapy may be efficacious for individuals with PTSD. In particular, active light therapy appears to improve cognitive/mood and hyperarousal symptoms for those with probable PTSD. Having a placebo-control condition lends confidence that therapeutic effects are due to the effect of light and not simply adjustments in sleep timing. With recruitment ongoing, we will present data from a larger sample of randomized participants at the conference.
Keywords: PTSD, Light Therapy, Clinical Trial
Disclosure: Part 1: Annum Health, Consultant, Spouse, ORCAS, Consultant, Spouse, Ginger.io, Advisory Board, Spouse, Kemoko, Inc., Advisory Board, Spouse.

T7. Unbiased Identification of Mouse Behavioral Phenotypes

Carl Hammarsten, Alexandra Bord, Mary Ellen Shelton, Marie Walicki, Tayler Sheahan, Judith Golden, Robert Gereau, Jordan McCall*

Washington University in St. Louis, St. Louis, Missouri, United States

Background: Preclinical animal studies are critical to advancing basic neuroscience and clinical medicine. However, most animal behavior assays rely on preconceived notions regarding the important observed features of the

dataset, rather than unbiased assessment of the fundamental structure of the data. Fortunately, recent work in mathematics has been aggressively pursuing new means to blindly identify the important features in a dataset. In particular, the method of "persistent homology" offers a means of identifying unseen, but critical, relationships within highly complex datasets. Here we use persistent homology to blindly identify salient features in mouse behavior data following exposure to acute stress.
Methods: As a proof-of-principle experiment, we examined the locomotor behavior of more than one hundred mice following exposure to stress. Briefly, we collected a large survey of animal behavior in the open field test (OFT) following a 30-minute restraint stress or a no-stress control group. To assess anxiogenic-like responses in OFT, mice were placed in a dimly lit (~30 lux) open-air, four-walled chamber. Physical location within the center of the chamber during a 20-minute trial is recorded and multiple dimensions of the behavior are extracted (e.g. XY coordinates of the body, head, and tail; head direction; body length; instantaneous velocity; etc.). Persistent homological analysis was then performed on the extracted behaviors from each mouse. To test whether stress-related persistent homological factors can be disrupted by pharmacological manipulation, new mice were placed in the OFT following acute stress exposure and treatment with either vehicle or the non-selective beta-adrenergic receptor antagonist, Propranolol (10 mg/kg).
Results: This approach generated a 'diagnostic framework' to assess the behavioral phenotype of individual animals. In a double-blind fashion, the persistent homological analyses successfully identified two groups of mice. These two mathematically-identified groups were segregated by stress exposure. Using the persistent homological features of these two groups, we were then able to test new animals against this 'training set' to blindly determine whether new animals had been exposed to stress. By testing new animals against this 'training set' we can blindly predict whether new animals had been stressed and whether commonly used anxiolytics could reverse these unguided classifications. This approach yields greater than 90% accuracy using only the XY position coordinates in an open field.
Conclusions: This mathematical approach is capable of identifying critical features of animal behavioral data that may not otherwise be recognized by human observers. Such an unguided 'diagnostic framework' may be useful in modeling psychiatric disorders and potential therapeutic approaches. We are currently determining whether the same approach can be used to separately identify pain states in animals. Such a distinction may be useful in considering and modeling psychiatric sequelae following chronic pain.
Keywords: Acute Stress, Anxiety, Behavioral Analysis
Disclosure: Nothing to Disclose.

T8. Investigating Discordance Across Physiological and Clinical Measures Within Memory Reconsolidation Targeting Fear of Flying

Jessica Maples-Keller*, Matthew Price, Tanja Jovanovic, Seth Norrholm, Barbara Rothbaum

Emory University School of Medicine, Atlanta, Georgia, United States

Background: When a specific memory trace is retrieved from long-term memory, it exists in a labile state and stored information becomes amenable to change (Monfils, Cowansage, Klann, & LeDoux, 2009). When this memory is stored again, it is known as reconsolidation (Nader et al., 2000). Both animal models (Mondils et al., 2009) and a human preclinical study (Schiller et al., 2009) have provided support that extinction training within the reconsolidation window results in decreased fear responding. Attempts to replicate these reconsolidation paradigms in healthy human samples using within-subjects design have demonstrated mixed results. Fear and anxiety can be measured in different ways, all of which provide important and potentially clinically relevant information, and assessment of different aspects of anxiety often demonstrate discordance.
Methods: We investigated a reconsolidation paradigm in a sample of patients with clinically significant fear of flying ($N = 89$) receiving a full course of virtual reality exposure therapy in order to investigate if using a cue to reactive the memory of the feared stimulus 10 minutes before extinction training leads to decreased anxiety. Participants were randomly assigned the reactivation group or control group and completed follow-up assessments at 1, 3, 6, and 12 months. The present investigation will focus on the outcomes and concordance across a multi-modal assessment including heart rate, skin conductance level, self-report clinical measures, and clinician-rated interview.
Results: Significant differences were identified with regard to heart rate and skin conductance, but not with regard to clinical self-report and interview measures. The physiological variables were not significant correlated with each other (p values ranging from .12 to .81).
Conclusions: Memory reactivation prior to extinction training in clinical samples may enhance its effects, at least on physiological responses, and it is notable that different outcome variables demonstrate discordance. This discordance across the physiological measures and with clinical measures may be related to previous mixed results in healthy human samples. Because heart rate is controlled by sympathetic and parasympathetic nervous system outputs, while skin conductance is purely a sympathetic measure, the two would not necessarily reflect the same neural activation.
Keywords: Memory Reconsolidation, Fear of Flying, Psychophysiology, Exposure Therapy
Disclosure: Nothing to Disclose.

T9. The Effect of Frustration on Response Control in Children With ADHD With and Without Comorbidity Compared to Typically-Developing Controls

Karen Seymour*, Keri Rosch, Mary Martinelli, Stewart Mostofsky

Johns Hopkins University School of Medicine, Baltimore, Maryland, United States

Background: Emotion dysregulation (ED) is a key impairment for many individuals with attention-deficit/hyperactivity disorder (ADHD). One particularly impairing form of ED in children with ADHD is irritability which is defined as a "mood state characterized by a low threshold for frustration" in which frustration refers to "an affective response to

blocked goal attainment". Compared to typically-developing children (TD), children with ADHD show greater levels (i.e., intensity) of frustration, are more likely to quit a frustrating task, exhibit greater focus on the negative aspects of the task and display less constructive patterns of emotional coping when frustrated.

However, to date no studies have examined the effect of frustration on core neuropsychological deficits in children with ADHD. Response control (i.e., attentional control and response inhibition) is a primary neuropsychological deficit in children with ADHD, and prior research consistently shows that compared to TDs, children with ADHD show deficits in response control, particularly intrasubject variability (ISV) and commission error rate. In the context of emotional stimuli, the inability to inhibit a response (i.e., poor response control) would likely result in dysregulated emotion including excessive emotional responses, mood lability and atypical allocation of attention to emotional stimuli. In order to inform prevention and intervention targeting ED in children with ADHD, it is critical to understand how emotional states such as frustration interact with neurocognitive deficits given the frequency with which children with ADHD encounter frustrating situations in academic, home, and social situations.

Therefore, the goal of this project was to address these limitations by examining the effect of frustration on response control in children with ADHD only vs. children with ADHD+comorbidity vs. TD controls.

Methods: Participants included 77 children, ages 8-12 years-old, in one of three groups: children diagnosed with DSM-5 ADHD only ($n = 22$); children diagnosed with ADHD + either comorbid Oppositional Defiant Disorder, an anxiety disorder or a depressive disorder (i.e., ADHD+comorbidity, n = 26) or TD controls ($n = 29$).

Participants completed the Frustration Go/NoGo (GNG) task, a novel behavioral paradigm developed by Drs. Seymour and Mostofsky to examine the effect of frustration on response control. This task is similar to other GNG tasks in that participants are asked to respond to Go stimuli and withhold a response to NoGo stimuli. There are 3 blocks to this task: Block 1(non-frustration), Block 2 (frustration), Block 3 (non-frustration). To elicit frustration, in Block 2, 50% of the Go trials are rigged so that despite participant effort they will not be successful on these trials. Dependent variables for this task include: ISV of Go Reaction Time (Tau) and commission error rate. Participants will complete the PANAS-C at four points during the task to assess frustration (baseline, after run 1, after run 2, after run 3).

Results: A manipulation check was conducted to establish that this task elicited frustration in participants. For all participants, results showed that frustration increased from baseline to Block 1 and from Block 1 to Block 2 (p's $< .001$). Frustration decreased from Block 2 to Block 3 ($p < .001$). While there was a main effect of Block on frustration ratings, the Block X Group interaction was non-significant. However, there was a trend for group differences when examining changes in frustration between Blocks 2 and 3. Specifically, the ADHD+comorbidity group reported a greater reduction in frustration (once frustration was removed) compared to children with ADHD only $F(2,74) = 2.77$, $p = .069$.

Repeated measures GLM analyses revealed a main effect of frustration on commission error rate, $F(2,73) = 4.05$, $p < .05$, such that for all groups commissions errors increased from Block 1 to Block 2, but deceased from Block 2 to Block 3. The Block X Group interaction was not significant, $F(4,146) = .534$, $p = .711$.

For Tau, there was also a main effect of frustration across the blocks, $F(2,73) = 48.60$, $p < .001$, such that as frustration increased from Block 1 to Block 2 so did Tau, but it decreased in the absence of frustration from Block 2 to Block 3. Moreover, there was a trend for a Block X Group interaction, $F(4, 146) = 2.30$, $p = .062$. Post hoc analyses showed that in Block 1, there were significant group differences between all groups in Tau (ADHD+comorbidity > ADHD only > TD). In Block 2, there were significant group differences such that ADHD+comorbidity > than ADHD only and TD, but there were no differences between the ADHD only and TD groups. Finally, in Block 3, there were significant group differences such that the ADHD +comorbidity > TD and ADHD only > TD, but there were no differences between ADHD groups.

Conclusions: Results support the validity of this novel task at assessing the effects of frustration on response control. Results also suggest that especially for children with ADHD +comorbidity frustration may adversely affect response control particularly attentional lapses. Future studies are needed to elucidate the neurobiological underpinning of frustration in children with ADHD both with and without comorbidity.

Keywords: Attention Deficit Hyperactivity Disorder, Emotional Regulation, Frustration Tolerance, Children and Adolescents, Irritability

Disclosure: Nothing to Disclose.

T10. Neural Response to Positive Stimuli is Differentially Associated With Child Behavioral Reward Seeking in High and Low Risk Young Children

Judith Morgan*, Brittany Woods, Erika Forbes

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: Children of depressed parents are at increased risk for developing depression and other psychiatric illnesses relative to their peers. Although elevated rates of psychiatric illness are well-established, early developmental mechanisms underlying transmission of risk are less understood. Given that depression is a disorder of altered positive affect, physiological differences in how high risk young children respond to positive stimuli may be especially important. Altered neural reward response to positive stimuli (e.g., blunted striatal response) has been demonstrated in high risk adolescents and has been shown to predict increases in depressive symptoms over time, suggesting that these reward disruptions are important to the pathophysiology of depression. However, little work has been done with early school age children, a period of rapid socio-emotional growth. Given that the early school age period is associated with increasing efforts to regulate emotion and attention, neural differences in how high-risk children allocate attention to positive stimuli may be emerging. Prior neurobehavioral work has found a link between neural reward responding and positive affect expression in adolescents; altered

responding in neural reward regions in high risk young children may be related to way in which they pursue positive experiences behaviorally. Particularly, function within neural reward regions may be especially important for high risk young children to promote healthy pursuit of positive experiences. Understanding how young children at risk for depression may differ in neural processing of positive stimuli may be important for preventive intervention.

Methods: The current study evaluated 49 6- to 8-year old (M = 6.8 years, SD = .77; 55% female) typically developing children with no history of psychiatric illness. Children were categorized as high risk if they had a biological mother with two or more lifetime episodes of Major Depressive Disorder or Dysthymia on the SCID (i.e., recurrent and/or chronic depression) ($n = 20$). Children were categorized as low risk if mothers had no lifetime history of any Axis I disorder ($n = 29$). Children completed a computerized progressive ratio (PR) schedule, in which they must work at increasing levels of effort to inflate a virtual balloon. Children earned a token for each balloon that they inflated and could exchange tokens for prizes later that day. This task measures how much effort a child is willing to exert to achieve a reward and serves as a behavioral measure of motivation and reward seeking. Children also completed a 3.5-minute fMRI paradigm in a 3T Siemens TIM Trio in which they viewed blocks of unfamiliar adults displaying happy, sad, and neutral expressions. This task measures implicit emotion processing and response to social reward (happy faces). The contrast of interest for this analysis was happy faces > neutral faces.

Results: There were no group differences in child age, gender, or behavioral reward seeking (ps = .40-.99). Regression models in SPM8 included child age, child gender, child risk status, number of tokens received in PR schedule, and the multiplicative interaction of risk status x tokens. Whole brain analyses were used to probe significant clusters at a cluster forming threshold of $p < .001$ and corrected for multiple comparisons using family wise error of $p < .05$. There was a significant interactive effect of risk by tokens on response in the dorsal striatum (394 voxels, t = 4.72, [-32, -10, 36], pFWE = .004). Simple slopes analyses suggested that the positive association between behavioral reward seeking and neural response to social reward in the dorsal striatum was significant only for high risk children. Further, there was also a main effect of risk status on neural response to happy faces, such that children at high risk for depression showed lower response in the dorsal lateral prefrontal cortex (dlPFC, 447 voxels, t = 5.49, [-40, 22, 34], pFWE = .002). This main effect was qualified by a significant interaction with number of tokens earned on the PR schedule, such that high-risk children with low behavioral reward seeking showed the lowest response in the dlPFC.

Conclusions: The current study demonstrated that neural response in the dorsal striatum, a region implicated in reward processing, is positively associated with behavioral reward seeking, but only for young children at familial risk for depression. We did not find that high and low risk children differed in their neural response to happy faces in the striatum, as has been found in studies with high risk adolescents. Instead, our findings suggest that stronger neural function in this key reward region may be required to promote pursuit of reward in difficult circumstances for young children at high familial risk for depression. However, for young children without this familial risk, response in this circuitry is not related to their effort to seek out rewards. Further, our findings demonstrated a group difference in neural response to happy faces in the dlPFC, a region implicated in attention and response to unpredictable happy faces, perhaps suggesting that unfamiliar happy faces are less meaningful to high risk youth. This appeared to be especially so if the high risk young children were behaviorally less motivated to make effort to receive reward. Our findings provide important information about the importance of strong neural responding in reward circuitry for behavioral reward seeking in young children at risk for depression.

Keywords: Reward, Children, Social Behavior
Disclosure: Nothing to Disclose.

T11. Using Transcriptomic Data to Characterize Impact of Early Life Stress Across Reward Circuitry and Predict Response to Antidepressant Treatment

Catherine Pena*, Yong-Hwee Eddie Loh, Aarthi Ramakrishnan, Immanuel Purushothaman, Hope Kronman, Rosemary Bagot, Hannah Cates, Bhakti Patel, Li Shen, Eric Nestler

Icahn School of Medicine at Mount Sinai, New York, New York, United States

Background: Abuse, neglect, and other forms of early life stress (ELS) significantly increase risk for depression and drug abuse. Depressed patients with a history of ELS also have earlier onset and more frequent episodes of depression, and are less responsive to traditional antidepressant medication. We sought to understand how ELS alters transcriptional development in key reward-related brain regions, which may ultimately impact response to drug treatments.

Methods: We recently established a "two hit" stress paradigm in male and female C57BL/6 mice in which stress in a postnatal sensitive window increases susceptibility to depression-like behavior after additional stress in adulthood. Mice in the current study were standard-reared or exposed to ELS from P10-17, and half of each group faced chronic social defeat stress (males) or sub-threshold variable stress (females) in adulthood. RNA-seq was performed in adult male and female ventral tegmental area (VTA), nucleus accumbens (NAc), and prefrontal cortex (PFC). RNA-seq was analyzed by standard and custom pipelines. Patterns of transcriptional change were then compared to published RNA-seq datasets from the same brain regions of male mice characterized as stress-susceptible and responsive or non-responsive to imipramine treatment.

Results: Across brain regions and sexes, we discovered a similar "depression-like" transcriptional pattern among ELS mice prior to behavioral changes as among adult-stress mice exhibiting depression-like behavior. Among male and female mice, the largest main effect of ELS on transcription was discovered in the NAc. The largest interaction between ELS and adult stress on transcription was observed in the male and female VTA, and male PFC. Using a threshold-free rank-rank hypergeometric overlap analysis, we show that the transcriptional signatures of ELS more closely resemble transcriptional signatures observed in treatment-resistant

mice than treatment-responsive mice, particularly within VTA and PFC.
Conclusions: Different regions within reward circuitry encode distinct and long-lasting responses to ELS, and are predicted to differently contribute to active vs latent behavioral consequences of ELS. Our analyses support the hypothesis that ELS alters transcriptional development of key reward regions, which may contribute to diminished efficacy of traditional antidepressant treatments among ELS-exposed subjects.
Keywords: Early Life Stress, Depression, RNA-seq, Treatment Resistant Depression
Disclosure: Nothing to Disclose.

T12. Optogenetic Examination of Prefrontal-Amygdala Synaptic Development

Maithe Arruda-Carvalho*, Wan-Chen Wu, Kirstie Cummings, Roger Clem

University of Toronto, Scarborough, Toronto, Canada

Background: Early life experiences are crucial at defining cognitive and mental health function throughout life. Childhood and adolescence are the predominant age of onset for the majority of mental disorders, periods in which key brain areas involved in emotional processing, such as medial prefrontal cortex (mPFC) and amygdala are maturing. Anatomical and morphological changes occur in such areas during early life; nevertheless, how these changes affect circuit function and its consequences to the onset of mental illness is currently unknown.
Methods: We used optogenetics and ex-vivo electrophysiology in mice to investigate how synaptic transmission between PFC and amygdala changes across several developmental stages. We infused AAV virus expressing the excitatory optogene channelrhodopsin (ChR2) into the PFC of C57BL6/J mice at postnatal day (P) 10, 15, 21, 30, 45 and 60, corresponding to infant, early and late juvenile, adolescent, late adolescent and adult developmental stages, respectively. Seven days after infection, brains were processed for patch clamp electrophysiology to record from basal amygdala principal neurons while selectively activating PFC terminals by pulsing blue light.
Results: We found that PFC projections arrive in basolateral amygdala (BLA) at around P15, coinciding with a massive increase in BLA spontaneous synaptic drive. Following synapse formation and continuing through adolescence, mPFC-BLA circuits present pre- and postsynaptic strengthening of excitatory synapses as well as a transient enhancement in feedforward inhibition.
Conclusions: These data define for the first time a postnatal period of synaptic refinement within the PFC-Amygdala pathway, centered between P15 and P30. In sensory development, changes in excitatory transmission and maturation of inhibition are key mechanisms in sensitive period timing. The profile and timing of our data are indicative of a developmental sensitive period in the PFC-Amygdala pathway taking place between the juvenile and adolescent stages, during which synaptic transmission may be especially sensitive to adverse experience. Whilst it is now clear that most mental illnesses start at an early age, the vast majority of research on how emotional learning circuits might contribute to mental disorders such as PTSD and depression focuses on adult animals. Understanding how brain circuits implicated in mental illness mature will be critical for gaining insight into the etiology of those disorders, a necessary step for designing more targeted and efficient therapies.
Keywords: Prefrontal-Amygdala-Connectivity, Optogenetics, Development, Electrophysiology, Mice
Disclosure: Nothing to Disclose.

T13. Plasticity of Cognitive Control in Adolescents With Adverse Childhood Experiences

Jyoti Mishra*, Rajesh Sagar, Kiran Modi, Sana Alam, Adam Gazzaley

University of California, San Diego, La Jolla, California, United States

Background: Adverse childhood experiences (ACEs) range from neglect i.e. inadequate access to food, clothing, shelter, or nurturing care, to forms of trauma such as emotional, physical or sexual abuse. During their lifetime, individuals with ACEs are at risk for several mental health problems including depression, anxiety, attention deficit disorder, as well as physical illnesses such as poor cardiovascular health, all predictive of early mortality. As such, the scale of ACEs in our country (1 in 4 individuals) poses a huge healthcare and socio-economic burden, and it is an imperative to better understand and serve the mental health needs of this vulnerable population. In this project, we investigated whether digital interventions can impart improved cognitive control abilities in adolescents with ACEs.
Methods: In this international project, we enrolled 45 adolescents (10-18 years of age range) with a history of ACEs, recruited from a Child Welfare Services Center in New Delhi, India. All participants were living in stable foster-care for a minimum of 2 years i.e. all ACEs were from prior to this period, were school-going and understood English. Study participants underwent neuro-cognitive assessments, including resting state functional MRI measures, cognitive tests of sustained attention and interference processing, and behavioral assessments of attention deficit and hyperactivity. Adolescents were then cluster randomized to three study arms: (i) breath-focused self-regulation training akin to mindfulness practice (internal-focus arm), (ii) cognitive training with auditory and visual attention exercises (external-focus arm), and (iii) no intervention arm. The intervention period lasted 6 weeks with ~30 min of intervention engagement over 30 days. Post-intervention, we assessed neuro-cognitive, i.e. resting state functional connectivity, cognitive functioning and behavioral outcomes in all children. Academic performance measures were also obtained from teachers at post-intervention.
Results: We found that only the 'self-focus' intervention that inculcated breath-based self-regulation, was able to significantly strengthen cognitive control functions post vs. pre-intervention, while the 'external-focus' and no intervention arms did not show significant gains. Results for 'self-focus' training included (i) enhanced resting state functional connectivity of the cingulo-opercular network, which is the cognitive control network implicated in sustained task focus;

(ii) enhanced cognitive functioning in both sustained attention and interference processing tasks; (iii) reduced hyperactivity as rated by the child's caregiver; and (iv) a trend for higher academic performance abilities.

Conclusions: In a pilot study, we demonstrated that a mobile, digital self-regulation intervention can significantly improve cognitive control in adolescents with ACEs. Mobile interventions are cost-effective, scalable and accessible in low-resource settings where ACEs are commonplace. Our encouraging pilot results suggest that a larger randomized controlled study of the digital self-regulation program in adolescents with ACEs is warranted.

Keywords: Adverse Childhood Experiences, Cognitive Control, Self-Regulation, Translational Research, Cingulo-Opercular Network

Disclosure: Nothing to Disclose.

T14. Computational Phenotyping Reveals a Double Dissociation in the Neural Mechanisms of Irritability and Anxiety in Youth

Katharina Kircanski*, Lauren White, Wan-Ling Tseng, Jillian Wiggins, Heather Frank, Stefanie Sequeira, Susan Zhang, Kenneth Towbin, Argyris Stringaris, Daniel Pine, Ellen Leibenluft, Melissa Brotman

National Institute of Mental Health, Bethesda, Maryland, United States

Background: Comorbidity is ubiquitous in psychiatry and raises fundamental questions about phenotype-specific versus shared neurobiology (McTeague et al., 2017; Shanmugan et al., 2016; Stoddard et al., 2017). As psychiatry moves toward transdiagnostic dimensionally-assessed phenotypes, how to parse neural mechanisms of distinct but correlated dimensions remains an open question. Here, we leveraged computational methods to differentiate mechanisms of co-occurring symptom dimensions in treatment-seeking youth with relatively severe psychopathology.

Pediatric irritability and anxiety are prevalent and impairing, and they frequently co-occur, complicating treatment (Brotman et al., 2017; Stoddard et al., 2014). Parsing their unique and common neural mechanisms could reveal precise targets for early-life intervention. Both irritability and anxiety are characterized by high-arousal negative affect and biased attention orienting toward social threats, such as angry faces (Bar-Haim et al., 2007; Hommer et al., 2014). However, whereas irritability is associated with approach behavior to threat, anxiety is associated with avoidant behavior (Brotman et al., 2017). Thus, threat orienting is an ideal domain in which to examine unique and common neural mechanisms.

Methods: fMRI data were acquired from 197 youth ages 8 to 18 years (mean age = 13.1; 46% female). Providing wide variation in dimensional levels of irritability and anxiety, the sample spanned several diagnoses (disruptive mood dysregulation disorder [$n = 54$], anxiety disorder [$n = 50$], attention-deficit/hyperactivity disorder [$n = 37$], healthy volunteers [$n = 56$]). Participants were assessed on levels of irritability using the Affective Reactivity Index parent- and youth-report (Stringaris et al., 2012) and anxiety using the Screen for Child Anxiety Related Emotional Disorders parent- and youth-report (Birmaher et al., 1999).

We employed bifactor analysis to quantify the unique and shared variances of irritability and anxiety. Bifactor analysis estimates latent factors from observed data to reflect underlying constructs (Reise, 2012). Consistent with prior approaches (Kircanski et al., 2017), the best-fit model included unique factors of parent-reported irritability, youth-reported irritability, and anxiety (parent- and youth-reported), and a common factor termed negative affectivity (CFI = .959; NNFI = .950; RMSEA = .066).

A canonical fMRI dot-probe task assessed attention orienting to angry (i.e., threat) versus neutral faces (White et al., 2016). Analyses in AFNI examined the impact of the empirically-derived phenotypes on neural activation and amygdala-based functional connectivity during threat orienting (threat-incongruent vs. threat-congruent trials) and general threat viewing (threat [congruent/incongruent] vs. neutral trials) using ANCOVAs ($p < .005$; whole-brain multiple-testing correction to $\alpha = .05$; $k > 69$).

Results: While behavioral performance did not vary by any phenotype (all ps > .37), neural findings revealed a double dissociation between irritability and anxiety. When the task required attending away from the threat (threat-incongruent vs. threat-congruent trials), higher levels of parent-reported irritability were associated with increased activity in multiple regions mediating attentional and motor responses to negatively-valenced stimuli: left amygdala (ROI; $p = .022$), right insula, left ventrolateral prefrontal cortex, and bilateral dorsolateral prefrontal cortex, inferior parietal lobule, and caudate (whole-brain corrected; all ps < .001).

Anxiety was not associated with neural activity in any region. However, for functional connectivity, higher levels of anxiety were associated with decreased connectivity of the amygdala with hubs of cortico-limbic networks: bilateral cingulate and thalamus, left precentral gyrus, and right postcentral gyrus (whole-brain corrected; all ps < .001). Irritability was not associated with functional connectivity.

Last, when viewing threat vs. neutral trials, higher levels of negative affectivity were associated with increased activity in the right dorsomedial nucleus of the thalamus (whole-brain corrected; $p < .001$), which may reflect a general increase in motivation-driven processing of threat shared by irritability and anxiety.

Conclusions: A computational approach to parsing co-occurring symptom dimensions revealed a double dissociation. During threat orienting, only irritability was associated with increased neural activity, whereas only anxiety was associated with decreased neural connectivity. Consistent with a threat imminence framework (Fanselow, 1994), the increased neural activity specific to irritability may reflect heightened arousal mediating maladaptive approach behavior toward non-imminent threats. In contrast, decreased cortico-limbic connectivity in anxiety may reflect subtle but aberrant higher-order processing that mediates maladaptive avoidant behavior. Thus, while pediatric irritability and anxiety often co-occur, phenotype-specific brain mechanisms underlie dysfunction in threat orienting.

Distinct brain mechanisms may drive different treatment approaches for irritability and anxiety. These findings inform precision psychiatry and a path toward targeted interventions despite the challenges of phenotype co-occurrence. The identification of discrete early-expressed biomarkers of

psychiatric disease may inform more effective, targeted treatments in youth.
Keywords: Functional MRI (fMRI), Children and Adolescents, Irritability, Anxiety, Computational Psychiatry
Disclosure: Nothing to Disclose.

T15. Discrimination-Related Increases in Amygdala Functional Connectivity are Associated With Memory Impairment

Uraina Clark*, Desiree Byrd, Rachal Hegde

Icahn School of Medicine at Mount Sinai, New York, New York, United States

Background: Social discrimination, a type of chronic stressor, is associated with poorer physical, cognitive, and mental health outcomes, yet we have little understanding of how discrimination affects neural functions in marginalized populations. By contrast, the effects of psychosocial stress on brain function are well documented, with evidence of significant effects on amygdala function. Accordingly, we conducted an examination of the relation between self-reported discrimination exposure and amygdala activity in a diverse sample of adults. In line with findings from studies of chronic stress and posttraumatic stress disorder, we hypothesized that greater discrimination exposure would correlate with greater intrinsic amygdala activity (at rest) and stronger resting-state functional connectivity (rsFC) between the amygdala and brain regions within the salience network. Because greater salience network connectivity has been linked to cognitive dysfunction and dementia risk, we further examined the relation of discrimination-related changes in amygdala activity and rsFC to measures of cognitive function.
Methods: We included 74 adults (aged 25–70 years; 43% female; 72% African American; 23% Hispanic; 32% homosexual/bisexual). The Everyday Discrimination Scale, a 9-item self-report questionnaire, assessed the frequency of unfair treatment during common everyday situations. Spontaneous amygdala activity and functional connectivity were assessed during resting-state functional MRI. A brief battery of standardized neuropsychological tests was administered outside of the MRI scanner to assess three domains of cognitive function: executive functions, psychomotor speed, and verbal memory abilities.
Results: Greater self-reported discrimination exposure was associated with higher levels of spontaneous amygdala activity ($\beta = .33$ [$t = 2.06$, $p = .043$]), as well as stronger functional connectivity between the amygdala and several regions of the salience network (anterior insula, putamen, caudate, anterior cingulate, medial frontal gyrus), with the most robust effects observed in the thalamus (voxel-wise $p < .005$; cluster size > 90 voxels; $p < .05$, family-wise error corrected). The observed effects were independent of several demographic (e.g., age, race, ethnicity, sex, sexual orientation) and psychological (i.e., current stress, depression, anxiety, posttraumatic stress disorder-related symptoms) factors. Regarding associations with cognitive functions, we observed a negative association between amygdala-thalamic rsFC and verbal memory function ($\beta = .-31$ [$t = -2.50$, $p = .015$]), which was maintained even after controlling for demographic and psychological factors ($\beta = .-31$, [$t = -2.25$, $p = .028$]).
Conclusions: Our findings provide the first evidence that social discrimination is independently associated with elevations in amygdala activity and functional connectivity, thus revealing clear parallels between the neural substrates of discrimination and stressors of other origins. Moreover, our data also suggest that discrimination-related changes occurring within this network may contribute to memory impairments. Such results have important implications for our understanding of the impact of discrimination on neural functions among marginalized individuals. Considering the growing body of evidence demonstrating that, in the US, several marginalized populations (e.g., African Americans) experience higher rates of dementia, these results should spur future investigations of the links between discrimination exposure and cognitive dysfunction, with a focus on amygdala-based networks as a potential etiological factor.
Keywords: Amygdala, Salience Network, Social Discrimination, Acute and Chronic Stress, Memory
Disclosure: Nothing to Disclose.

T16. The Biomarker Potential of Quantitative Electroencephalographic Measures of NREM Slow Waves and Sleep Spindles to Track Alzheimer's Disease Pathological Burden in Healthy Older Adults

Bryce Mander*, Joseph Winer, Shawn Marks, Brandon Lu, Jared Saletin, Sonia Ancoli-Israel, William Jagust, Matthew Walker

University of California, Irvine, Orange, California, United States

Background: Accurate early biomarkers of Alzheimer's disease (AD) pathology are urgently needed to determine individual risk and offer effective pre-disease or early disease stage treatment interventions. The in vivo gold standard, utilizing positron emission tomography (PET) or cerebrospinal fluid sampling of Aβ or tau proteins is impractical at a population level, expensive, and invasive. However, recent work has linked Aβ and tau burden in healthy older adults to specific quantitative electroencephalographic (EEG) measures during non-rapid-eye-movement (NREM) sleep. For example, measures of slow wave activity (SWA) particularly in the < 1 Hz frequency range, are correlated with the degree of Aβ burden. Building on this recent evidence, here we examine whether < 1Hz NREM SWA accurately discriminates between cognitively healthy individuals with and without Aβ pathology, and whether sleep spindle measures successfully track tau burden. We also examine whether there is a topographic specificity to such relationships, and whether this is consistent with or in contrast to differences observed in normal aging.
Methods: 42 cognitively normal older adults (75.5 ± 3.9 years) received [11C]PIB PET scans to assess Aβ pathology, with a subset ($n = 27$, 75.5 ± 3.9 years) also receiving [18F] AV1451 tau PET. Sleep was recorded using polysomnography (PSG) including 19-channel EEG. Structural magnetic resonance images were co-registered to PET images. Mean AV1451 SUVR was calculated in hippocampus, and a global cortical PIB DVR measure was calculated (PIB Index), both

using cerebellar gray matter as a reference region. Sleep analyses focused a priori on (1) proportion of NREM SWA <1Hz, previously associated with Aβ, and (2) mean frequency of fast-frequency sleep spindles, previously associated with hippocampus structure. For Aβ, analyses were conducted both by utilizing PIB Index as a continuous regressor, and, for all diagnostic calculations, by stratifying subjects' PIB status as PIB+ or PIB- using an established PIB DVR cut-off of 1.08. SWA was examined as a continuous regressor, and was stratified for diagnostic calculations based on a median split across subjects. For all parametric analyses, PIB Index was log transformed. AV1451 SUVR was only examined as a continuous measure, as no standard cut-offs for positivity have been validated.

Results: For Aβ, similar to our previous report, proportion of NREM SWA <1Hz was negatively associated with PIB index, particularly over frontal EEG derivations (FP1, FP2, F7, F3, FZ, F4, and F8; $P<0.05$ false discovery rate (FDR) corrected). Demonstrating specificity for NREM sleep, proportion of spectral power during REM sleep <1Hz was not significantly associated with PIB Index at any derivation (all $r2<0.07$, all $P>0.12$ uncorrected). Stratifying by PIB status, <1Hz NREM SWA was significantly lower in Aβ+ relative Aβ- adults across most EEG derivations, with the peak difference over frontal EEG derivations (FP1, FP2, F7, F3, FZ, F4, F8, CZ, and C4 all $P<0.01$ FDR corrected). Proportion of REM spectral power <1Hz (all $P>0.31$ uncorrected) did not differ significantly by PIB status. Binary logistic regression accounting for age and sex revealed that proportion of NREM SWA <1Hz also significantly predicted PIB status over most EEG derivations, with the peak significance detected over frontal electrodes (FP1, FP2, F7, F3, FZ, F4, F8, CZ, C4 all $P<0.025$ FDR corrected). Similar binary logistic regression models failed to detect any significant associations between proportion of REM spectral power <1Hz and PIB status (all $P>0.30$ uncorrected). Proportion of NREM SWA <1Hz was most accurate at categorizing PIB status over frontal EEG derivations [peak at F4: 81.0% categorization accuracy (34/42), 88.9% sensitivity, 75% specificity, 61.5% positive predictive value, 93.8% negative predictive value, 3.56 positive likelihood ratio, 0.15 negative likelihood ratio, 24 (99.9% C.I.:1.3—442.7) diagnostic odds ratio, and κ inter-rater reliability = 0.62]. Proportion of REM spectral power <1Hz did not accurately discriminate PIB status [all categorization accuracy <0.67, all diagnostic odds ratio <4 (all 99.9% C.I.s include 1), and κ inter-rater reliability <0.3]. Hippocampal tau burden was also not associated with proportion of NREM SWA <1Hz (e.g. at F4: $r=-0.09$, $P=0.65$). Instead, tau hippocampal burden was associated with lower mean fast sleep spindle frequency over parietal electrode derivations (P3, PZ, and P4: $r=-0.42$, $P=0.031$), while Aβ burden was not ($r=-0.12$, $P=0.55$).

Conclusions: These findings support the utility of quantitative estimates of NREM SWA features as sensitive biomarkers of PIB status in healthy older adults without cognitive impairments, particularly over frontal electrode derivations. The tau findings, though preliminary, further indicate that quantitative estimates of NREM SWA are specific to Aβ, and not driven by hippocampal tau burden, at least in healthy older adults. Moreover, tau burden also appears to have a distinct effect on the expression of NREM sleep spindles that is independent of the degree of Aβ burden. Therefore, quantitative estimates of distinct NREM sleep oscillations may provide useful surrogate biomarkers of distinct AD neuropathologies, and thus may indicate AD risk in healthy older adults in a manner that is safe, relatively inexpensive, non-invasive, and scalable to a population level.

Keywords: Slow Waves, Sleep Spindles, Alzheimer's Disease, PET Imaging, Quantitative EEG

Disclosure: Nothing to Disclose.

T17. A Dimensional Measure of Prematurity is Associated With Structural Brain Network Abnormalities in Children, Adolescents, and Young Adults

Antonia Kaczkurkin*, Rula Nassar, Cedric Xia, Aristeidis Sotiras, Marieta Pehlivanova, Tyler M. Moore, Angel Garcia de la Garza, David R. Roalf, Adon Rosen, Scott Lorch, Kosha Ruparel, Russell T. Shinohara, Christos Davatzikos, Ruben C. Gur, Raquel E. Gur, Theodore D. Satterthwaite

University of Pennsylvania, Philadelphia, Pennsylvania, United States

Background: Premature gestational age at birth is associated with neurocognitive abnormalities in childhood, yet it remains unknown whether the extent of prematurity is systematically related to delayed cognitive outcomes and neurostructural deficits during adolescence. Prior case-control studies typically compare those born very or extremely preterm to those born full term. A significant gap remains in our understanding of the potential neurobiological vulnerability of infants born moderately or late preterm, which comprise a substantial majority of preterm births. The purpose of the current study was to determine if a dimensional measure of prematurity impacts cognitive performance and brain structure during adolescence.

Methods: In a retrospective cohort study, participants were children, adolescents, and young adults ages 8-22 who represented a sub-sample of the Philadelphia Neurodevelopmental Cohort, a community-based sample of youth. The final sample ($n=278$, mean age = 13.26 years, 143 females) included 72 preterm youth born before 37 weeks gestation and 206 youth who were born term (37 weeks or later). Cognitive performance was assessed using the University of Pennsylvania Computerized Neurocognitive Battery, and summarized using a previously-validated factor analysis. High-resolution structural brain imaging was performed with a T1-weighted sequence acquired at 3T. Whole brain volumes were quantified on a voxelwise basis and then summarized as structural covariance networks using non-negative matrix factorization (NMF), an advanced multivariate analysis technique. Statistical analyses utilized generalized additive models with penalized splines to evaluate linear and nonlinear associations between gestational age and both cognition and brain structure. We also tested whether the association between cognitive performance and gestational age was mediated by deficits in structural networks using a mediation analysis with bootstrapped confidence intervals. Throughout, multiple testing was accounted for by controlling the False Discovery Rate.

Results: Lower gestational age was associated with diminished overall cognitive performance ($p = 0.03$), specifically

with deficits in executive function ($p = 0.02$). Furthermore, lower gestational age was associated with smaller volumes in 11 of 26 structural brain networks involving the orbitofrontal, temporal, parietal, and occipital cortices, as well as subcortical regions including the hippocampus. Lastly, the relationship between lower gestational age and poorer executive function was mediated by structural deficits in 9 of these 11 networks.

Conclusions: These results suggest that executive functioning and structural deficits are associated with a dimensional measure of prematurity. Together, these findings emphasize the durable impact of all levels of prematurity on cognition and brain structure, which persists into adolescence and young adulthood. The contribution of other potential factors requires further investigation. Give that the majority of premature births are not extreme, the current data suggests that interventions aimed at reducing the incidence of even relatively mild forms of prematurity may have a substantial benefit to public health.

Keywords: Human Neuroimaging, Prematurity, Executive Function, Brain Volume

Disclosure: Nothing to Disclose.

T18. Dopamine Mediated Effects of Amphetamine on Food and Water Intake

Miriam Bocarsly*, Veronica Alvarez

National Institute on Alcohol Abuse and Alcoholism/ National Institutes of Health, Rockville, Maryland, United States

Background: Psychostimulants, such as amphetamine, which are known to increase striatal dopamine (DA), have been shown to decrease food intake (hypophagia) while simultaneously increasing water intake (polydipsia). These effects have been exploited in the marketing of dietary supplements for weight loss. However, little is known about the specific receptor-level mechanisms by which these drugs are acting to facilitate changes in consummatory behavior. Dopamine D2 receptors (D2Rs) repeatedly emerge as a critical substrate for both the etiology and consequence of chronic DA-related neurological conditions, such as drug abuse and obesity. D2Rs are expressed in a number of neurochemically distinct populations of cells within the striatal circuitry, and due to this ubiquitous expression and the poor selectivity of pharmacological agents, the actions of D2Rs localized on different neuronal populations cannot be studied using conventional methods. In the current study, we used transgenic mouse models in which we have selectively targeted D2R deletion in medium spiny neurons (MSN) in the striatum or dopaminergic terminals emanating from the midbrain. These deletions allow for better understanding of the specific contributions of these receptors to the actions of amphetamine on consummatory behaviors.

Methods: First, we set-up and validated a detection method for food and liquid intake with the appropriate sensitivity and temporal resolution needed to study the effects of stimulants on consummatory behavior. In Exp. 1 this system was used to profile the effects of amphetamine (2 mg/kg; i.p.) on food and water intake in wild-type mice. While amphetamine profoundly affects dopamine functioning, it is known to have effects beyond the D2Rs. To determine if hypophagia and polydipsia maybe D2R-mediated behaviors (Exp. 2), systemic quinlorane (0.03 mg/kg; i.p.), a D2R agonist, was administered to wild-type mice. Finally, in Exp. 3, we administered amphetamine to transgenic mice lacking D2Rs on medium spiny neurons (MSN) in the striatum or dopaminergic terminals emanating from the midbrain, and profiled locomotion, food and water intakes. All animal studies were approved by the Institutional Animal Care and Use Committee and were conducted in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals.

Results: In Exp.1 wild-type mice receiving a peripheral dose of amphetamine showed an acute decrease in standard chow intake and an increase in water intake, both which correspond with an increase in locomotor behavior. Administration of a D2R agonist (Exp. 2) led to a decrease in chow intake and increased water intake, while simultaneously having little to no effect on locomotion. These data indicate that hypophagia and polydipsia are likely modulated by the dopamine D2 receptor. Finally (Exp. 3) we found that targeted deletion of D2Rs on MSNs impairs amphetamine-induced hypophagia, but has no effect on polydipsia. Conversely, mice specifically lacking D2Rs on dopaminergic terminals emanating from the midbrain show impaired amphetamine-induced polydipsia, but no effect on hypophagia. Both transgenic mouse lines demonstrated intact locomotor responses to amphetamine. These data suggest that D2Rs on MSNs are required in amphetamine-induced hypophagia, while D2Rs on midbrain dopamine neurons are required in amphetamine-induced polydipsia.

Conclusions: The current study indicates that different subsets of D2Rs are likely important in controlling amphetamine-induced food versus water intake. Future directions will further characterize and validate the roles of various subpopulations of D2Rs in modulating the effects of stimulants on food and water intake. While the use of traditional stimulants, such as amphetamine, are no longer medically condoned for weight loss, better understanding of the neuronal circuitry underlying stimulant-induced hypophagia and polydipsia will inform the development of safe pharmaceuticals targeting obesity.

Keywords: Obesity, Dopamine 2 Receptor, Psychostimulant, Amphetamine

Disclosure: Nothing to Disclose.

T19. Alpha-7 Nicotinic Acetylcholine Receptors in the Hippocampus Bidirectionally Regulate Aggressive Behavior in Mice

Alan Lewis*, Steven Pittenger, Yann Mineur, Kara Furman, Colin Bond, Dawson Stout, Philip Smith, Ralph DiLeone, Marina Picciotto

Yale University, New Haven, Connecticut, United States

Background: The alpha-7 nicotinic acetylcholine receptor (nAChR) is an excitatory ligand-gated ion channel expressed in multiple cells types of the hippocampus. The gene coding for alpha-7, CHRNA7, resides in an unstable genomic region prone to deletions and duplications. Deletion of CHRNA7 is observed in the 15q13.3 microdeletion syndrome, which

results in a heterogeneous neuropsychiatric phenotype that frequently includes persistent impulsive aggression. We previously reported that the nAChR agonist nicotine reduced aggressive behavior in mice, an effect that required activation of alpha-7 nAChRs and was recapitulated by GTS-21/DMXB-A, an alpha-7 nAChR partial agonist. These findings across multiple species support the association of alpha-7 nAChR signaling and regulation of aggression. Here we sought to identify the brain regions and potential neurocircuitry important for alpha-7 nAChR regulation of aggressive behavior in mice.

Methods: F1 crosses of C57BL/6 and BALB/c mice heterozygous for Chrna7 and their wild type littermates were tested in a resident-intruder paradigm. Immediate early gene staining using Arc and cFos was performed to quantify neuronal activation in the hippocampus and other relevant brain regions during aggressive interactions as well as after treatment with GTS-21. Adeno-associated virus expressing short hairpin RNA (shRNA) targeting Chrna7 or a scrambled control was infused bilaterally into the dentate gyrus (DG) of the hippocampal formation and mice subsequently tested in resident-intruder tests. Finally, the calcium indicator GCaMP6s was infused into the DG and fiber photometry performed during social interactions in mice to determine DG activity in real-time.

Results: Mice heterozygous for Chrna7 attacked faster and more often than wild type littermates. Arc staining was markedly upregulated in the dentate gyrus by aggressive interaction, which was reduced by pretreatment with GTS-21 (17 mg/kg). Knockdown of Chrna7 in the DG by shRNA shortened time to attack and blunted the anti-aggressive effects of nicotine (0.5 mg/kg) and GTS-21 (20 mg/kg). Finally, fiber photometry demonstrated DG activity was correlated with interaction bouts in both male-male and male-female social interactions.

Conclusions: Our findings further implicate the alpha-7 nAChR in the regulation of aggressive behavior and demonstrate hippocampal alpha-7 signaling is both necessary and sufficient to limit the onset of aggression. Furthermore, we identify the DG as a region dynamically reactive to social interaction. Taken together, these findings suggest that regulation of excitation and inhibition in the DG may be a key mechanism to regulate aggressive behavior.

Keywords: Aggression, Hippocampus, Alpha-7 Nicotinic Acetylcholine Receptor, Dentate Gyrus, Mouse Model

Disclosure: Nothing to Disclose.

T20. Adolescent Ethanol Exposure Increases Adult Anxiety-Like Behavior: Involvement of Intrinsic Excitability of Medium-Sized Spiny Neurons in the Nucleus Accumbens

Lili Shan Shan, Jordan Carpenter, Katarzyna Rybczyk, Terrence Deak, Yao-Ying Ma*

State University of New York at Binghamton, Binghamton, New York, United States

Background: Alcohol use typically begins early in adolescence, during which neurodevelopment can be interfered, increasing the likelihood of adult mental disorders such as anxiety. However, the cellular mechanisms underlying the consequences of adolescent alcohol exposure remain poorly understood. This study was designed to investigate the prolonged effects of chronic intermittent ethanol (CIE) during adolescence or adulthood, compared with chronic intermittent water (CIW), on anxiety levels using the light-dark transition (LDT) test and determining the intrinsic excitability of medium-sized spiny neurons (MSNs) in the striatum.

Methods: Rats were administered ethanol (4 g/kg/day, 3 days on and 2 days off for one cycle, repeated by 4 cycles, CIE), or water (CIW) via oral gavage at the same volume and temporal pattern during either their adolescent or adult stage. 3 weeks after the last administration, rats underwent one of the following procedures: 1) whole-cell patch clamp recordings in slices to measure the basic membrane properties, cellular excitability and response to SK channel modulators on MSNs in the dorsolateral striatum (DLS) or the ventromedial striatum (primarily, the nucleus accumbens shell, NAcS), biocytin was also perfused to the cell for morphological evaluations; 2) DLS and NAcS were dissected and SK protein level was assessed by Western blot immunostaining; 3) LDT test was performed to evaluate anxiety and general locomotion; some rats, cannulated bilaterally in the NAcS 2 weeks after CIW/CIE procedure, were tested 10 min after microinjection of SK channel modulators.

Results: The cross-over latency from the light to dark side during LDT test in adolescent CIE rats was significantly reduced relative to adolescent CIW control rats, indicating increased anxiety in rats with an adolescent CIE history. Cross-over latency in adult CIE and adult CIW groups showed no difference from that in the adolescent CIW group. In parallel, the excitability of MSNs in the NAcS, but not DLS, from rats with an adolescent CIE history was significantly higher than that of rats with an adolescent CIW history, which was at a similar level as adult CIE or adult CIW group. Detailed analysis of specific components of the action potential, demonstrated that the amplitude of the medium afterhyperpolarizations (mAHP), assumed to be mediated by small conductance (SK) calcium-activated potassium channels, but not the fast AHP, assumed to be mediated by large conductance (BK) calcium-activated potassium channels, was decreased specifically in the adolescent CIE group. The down-regulation of SK channel function in this adolescent CIE group was accompanied by decreased expression of SK3 protein. Furthermore, the increased anxiety level and the increased NAcS excitability in rats with an adolescent CIE history were significantly reversed by in vivo NAc microinjections and slice bath application of SK channel activator 1-ethyl-2-benzimidazolinone (1-EBIO), respectively; whereas in vivo or slice application of SK channel blocker apamin in rats with an adolescent CIW history was able to mimic the effects of adolescent CIW treatment, i.e., these CIW rats showed a significantly higher level of anxiety and increased NAcS excitability.

Conclusions: Adolescent ethanol exposure increases adult anxiety-like behavior by down regulation of SK channel function and protein expression, which leads to an increase of intrinsic excitability of MSNs in the NAcS. SK channels in the NAcS may serve as a target to prevent or treat adolescent alcohol binge exposure-induced mental disorders, such as anxiety in adulthood.

Keywords: Nucleus Accumbens Shell, SK Calcium-Activated Potassium Channels, Adolescence, Alcohol, Anxiety
Disclosure: Nothing to Disclose.

T21. Individual Differences in Personality and Neural Function Among Unmedicated Depressed Outpatients

Jay Fournier*, Henry Chase, Neil Jones, Logan Cummings, Simona Graur, Mary Phillips

University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania, United States

Background: Personality functioning represents one of the only predictors of differential response to psychotherapy versus medication treatments for depression to have replicated, but the mechanisms underlying these effects are unknown. A core challenge in identifying neurobiological signatures associated with measures of personality dysfunction, such as neuroticism, in the context of depression is the high correlation between markers of personality and measures of depression and anxiety symptoms. In prior work, we demonstrated that it is possible to disentangle specific facets of neuroticism from acute clinical symptoms, and we showed that one facet in particular, excessive self-consciousness, was uniquely associated with deficits in interpersonal functioning among depressed patients. In the current set of studies, we replicated and extended these findings by examining the degree to which excessive self-consciousness interferes with neural mechanisms associated with the effortful regulation of negative emotions and with the evaluation of beliefs about oneself and others.
Methods: Study 1: A sample of $N = 320$ adults (18-40 years old) completed the neuroticism scale of the NEO-PI-R as well as measures of acute depression (Hamilton Rating Scale for Depression) and anxiety (Hamilton Rating Scale for Anxiety) symptoms. Participants spanned the full range of depression severity, from those with no symptoms to those experiencing an acute depressive episode. We used a confirmatory factor analytic technique (confirmatory bifactor modeling) to replicate our prior findings and to separate the facets of neuroticism from symptomatic distress.
Study 2: During fMRI, a subset of unmedicated depressed adults ($N = 36$) from Study 1 completed an emotion regulation task in which they viewed negative interpersonal scenes and either attended to or reappraised the content of the scenes in order to alter their emotional reactions. We examined the degree to which levels of excessive self-consciousness were associated with BOLD response during reappraisal compared to naturalistic viewing.
Study 3: During fMRI, a subset of the adults in Study 1 ($N = 24$, depressed and healthy controls) performed a novel self/other belief task in which they evaluated whether positive and negative beliefs were true of themselves and famous others. Belief statements were drawn from the Automatic Thoughts Questionnaire, a self-report measure that captures positive and negative self-referential thinking, has strong psychometric properties, and is sensitive to change with treatments for depression. Statements were matched for average word frequency and sentence, word, and phoneme length.
Standard imaging preprocessing was performed using SPM12. Whole-brain results were FWE cluster corrected at $p < 0.05$, with a cluster-forming threshold of $p < 0.005$.
Results: We replicated our previous findings and demonstrated that the bifactor model fit the data well, RMSEA = 0.05, CFI = 0.96, and TLI = 0.96 (Study 1). Critically, we also replicated our findings that the associations between symptom severity and neuroticism were captured by the general factor in this model ($|r|s = 0.77$-0.79), whereas the specific facets of neuroticism, including excessive self-consciousness, were relatively independent of symptoms ($|r|s = 0.01$-0.14).
Whole brain analyses revealed that during regulation of negative emotional scenes (Study 2), depressed individuals with higher levels of excessive self-consciousness displayed increased BOLD response in the three large clusters frequently associated with the default mode network: a cluster in the posterior cingulate and precuneus ($K = 299$, FWE-corrected $p < 0.05$) and clusters in the left ($K = 308$, FWE-corrected $p < 0.05$) and right ($K = 167$, FWE-corrected $p < 0.05$) angular gyri.
During evaluation of self- and other-related beliefs, we observed expected task effects in the medial prefrontal cortex ($K = 988$, FWE-corrected $p < 0.05$) and the posterior cingulate and precuneus ($K = 335$, FWE-corrected $p < 0.05$). In whole brain analyses, individuals higher in excessive self-consciousness displayed increased activity across both self- and other-related conditions in a cluster in the posterior cingulate, extending into occipital cortex, ($K = 261$, FWE-correct $p < 0.05$). Likewise, in a medial prefrontal region of interest, individuals higher in excessive self-consciousness displayed increased activity across both self and other conditions ($F(1,14) = 9.48$, $p = 0.008$).
Conclusions: We replicated our prior findings and demonstrated in a separate sample that it is possible to separate components of neuroticism from measures of symptom severity. We extended these findings and demonstrated that the excessive self-consciousness facet of neuroticism is associated with specific alternations in neural function during emotion regulation and self/other processing. Specifically, we observed that excessive self-consciousness is associated with increased activity in regions typically associated with the default mode network during both tasks. These patterns suggest that depressed individuals high in excessive self-consciousness may have a reduced ability to consciously alter emotional experience and to shift effectively from self-referential processes when completing emotionally-relevant tasks. Furthermore, the findings point to possible treatment targets in the default mode network which could be engaged by targeted, neuromodulatory interventions.
Keywords: Depression, Personality, fMRI, Emotional Regulation, Self-Other Processing
Disclosure: Nothing to Disclose.

T22. GPR30 is Associated With Major Depressive Disorder

Teniel Ramikie*, Guia Guffanti, Jennifer Stevens, Kerry Ressler

McLean Hospital, Harvard Medical School, Belmont, Massachusetts, United States

Background: Mood disorders are one of the highest causes of disability worldwide. Strikingly, the prevalence of the mood

disorders, major depressive disorder (MDD), is higher in females as compared to males, with females developing this disorder at approximately 2-fold higher rates. This sex-specific susceptibility increases, in females, during puberty and declines following menopause—periods characterized by high and low fluctuations in gonadal hormone levels, respectively. Consistent with these observations are mounting evidence demonstrating that high and low levels of gonadal hormones, across the menstrual cycle, are differentially associated with susceptibility to depressive symptoms. Overall, this evidence suggest that gonadal hormones shape major depressive disorder vulnerability in males and females. While the reasons for this vulnerability remains to be fully understood, estradiol, a neuroactive steroid and the predominant gonadal hormone in females, has emerged as a strong candidate in the influence of gonadal hormones on mood regulatory systems. In the brain, estradiol exerts its biological functions through the actions of its cognate receptors which activate distinct secondary messenger pathways. Of these receptors, the G-protein coupled receptor 30 (GPR30), a relatively novel estrogen receptor, has emerged as a potential regulator of mood-related systems but the mechanisms by which GPR30 achieves this function remains largely unknown. To this end, this study seeks to examine this question by investigating the relationship between GPR30 and MDD using human data and animal models.

Methods: Genetic epidemiological analyses were carried out using subjects from the Grady Trauma Project (GTP), a cohort of highly traumatized urban civilian subjects, previously described in Ressler et al. 2011. Male and naturally-cycling female C57/BL6J mice, 9-12 weeks of age, were used for all rodent experiments. Estrous cycle examinations were performed using a combination of vaginal inspection (VI) and vaginal cytology (VC). VI and VC analyses were carried out for 2 consecutive cycles before the start of experiments. Immunohistochemical analysis were carried out as previously described (McCullough et al., 2016) and all analyses were performed using ImageJ and GraphPad Prism 7.

Results: To determine whether polymorphisms within GPR30 genetic locus are associated with a measure of MDD, we performed a tag-SNP analyses across the GPR30 receptor gene, in our GTP cohort, to determine the association of twelve SNPs with Beck depression inventory (BDI) total score, a psychometric test that measures depression severity. In our combined analyses ($n = 4{,}095$), two SNPs were found to be significantly associated with BDI total score ($P < 0.05$) following experimental-wide corrections. Following stratification by sex, only two SNPs were found to be significantly associated with BDI total score in our female ($n = 3069$, $P < 0.05$), but not male ($n = 1026$), GTP cohorts. Next, we performed similar analyses with total PTSD symptoms and found that this was not associated ($P > 0.05$) with our SNPs of interest, suggesting that these SNPs may be relatively specific to MDD. Given these gender differences, we next examined whether these two SNPs were associated with intermediate phenotypes in a female cohort with moderate depression symptoms. Within this cohort ($n = 72$), one of these two BDI-associated SNPS were associated with greater habituation of amygdala fMRI responses to fearful face stimuli in risk allele carriers as compared to controls ($F(1,56) = 3.73$, $P < 0.05$). Our subsequent immunohistochemical analyses revealed that GPR30 protein is indeed expressed within the rodent amygdala of both males ($n = 11$) and female ($n = 10$-12/estrous stage) and it exhibits sex ($P < 0.005$) and estrous-cycle dependent expression changes ($r2 = 0.1631$, $P < 0.005$) —signifying that GPR30 may differentially function in each sex and across the estrous cycle.

Conclusions: Genetic and endophenotypic analyses shows a strong relationship between GPR30 and a measure of MDD and that this relationship appears to occur in a sex-dependent manner. Furthermore, our rodent analyses revealed that GPR30 is expressed within the amygdala, the brain region implicated by our endophenotype. Furthermore, our additional analyses reveal that GPR30 expression is regulated by the estrous cycle and in a sex-dependent manner. Collectively, these data demonstrate a potential role of GPR30 in the sex-specific regulation of MDD pathophysiology.

Keywords: Major Depression Disorder, Sex Steroids, Amygdala

Disclosure: Nothing to Disclose.

T23. A Rare Variant in ANK3 in a Patient With Bipolar Disorder Leads to Altered Cortical Circuitry

Andrew Nelson, Rene Caballero-Floran, Kathryn Walder, Vann Bennett, Melvin McInnis, Chao Wang, Paul Jenkins*

University of Michigan Medical School, Ann Arbor, Michigan, United States

Background: Bipolar disorder is a common mental illness characterized by pathological swings in mood ranging from mania to depression. Available therapeutics are insufficient for effectively treating the underlying cause of bipolar disorder, thus there is an unmet need to identify the cellular mechanisms that contribute to bipolar disorder to identify new therapeutic targets. Several large genome-wide association studies have identified ANK3 as one of the most consistent and significant genes associated with bipolar disorder. The giant, 480kDa splice variant ankyrin-G (product of the ANK3 gene) is a critical adaptor protein essential for the proper formation of axon initial segments and nodes of Ranvier. Recently, giant ankyrin-G was discovered to be critical for stabilizing GABAergic inhibitory synapses, which underlie the proper synchronization and function of neuronal networks, and abnormalities in GABAergic interneuron circuitry have been linked to bipolar disorder.

Methods: Using cultured neurons, we have shown that giant ankyrin-G interacts directly with GABA receptor associated protein (GABARAP) to inhibit GABAA receptor endocytosis and stabilize GABAergic synapses. To test this mechanism in vivo, we generated a mouse model with a W1989R mutation in Ank3, which completely abolishes ankyrin-G association with GABARAP, as well as GABAA receptor clustering in vitro.

Results: Coronal brain sections from Ank3 W1989R mice showed the loss of GABAergic basket and chandelier cell synapses on cortical pyramidal neurons in layer II/III of the somatosensory cortex. In addition, whole-cell patch clamp recordings of miniature inhibitory postsynaptic currents

(mIPSCs) revealed a reduction in both the frequency and amplitude of inhibitory GABAA receptor-dependent currents. The axon initial segments and nodes of Ranvier are spared in the W1989R mouse, making this a useful model to study the specific role of ankyrin-G and GABAergic circuitry to understand how dysfunction in these circuits may contribute to neuropsychiatric disease. In addition to changes in inhibitory changes, we also see dramatic reductions in dendritic spines in cultured neurons from the Ank3 W1989R mouse, suggesting potential compensation for loss of inhibitory tone. Importantly, we have recently identified the first family trio carrying the ANK3 W1989R variant in our patient cohort in the Heinz C. Prechter Bipolar Research Program at the University of Michigan the proband (II: 1, age 45) is an employed Caucasian male with type I BD characterized by recurrent mania and depression with an age of onset of 17 years and successfully treated with lithium. Mother (I: 2, age 73) was diagnosed with BD with age of onset in her mid-30's and is currently characterized by chronic depression and intermittent irritability consistent with hypomania and mixed affective features, and is being treated in the community with antidepressant and anti-anxiety medication. Father (I; 1, age 72) is a retired veteran with a history of PTSD in the years following three tours of duty in Vietnam, and a history of alcohol abuse. He had no history of depression or treatment of any psychiatric disorder until one year ago whereupon he experienced depressive symptoms (untreated) in the context of his wife's multiple mood episodes that required hospitalization. Both the proband and the mother are heterozygous for the ANK3 W1989R mutation.

Conclusions: Overall, our studies have found that a rare ANK3 variant from a patient trio with bipolar disorder causes major alterations in cortical circuitry in a mouse model of this ANK3 W1989R mutation. Future studies will examine neurons derived from induced pluripotent stem cells (iPCSs) from the W1989R ANK3 patients and neurotypical controls to evaluate GABAergic signaling and the ability of potential therapeutics to rescue abnormal electrophysiology. Ultimately, establishing the mechanisms by which giant ankyrin-G regulates GABAergic circuitry may reveal novel therapeutic targets for the treatment of neuropsychiatric disorders.

Keywords: ANK3, Bipolar Disorder, GABAergic Circuitry, Cortical Excitation-Inhibition Balance

Disclosure: Nothing to Disclose.

T24. Resting State Functional Connectivity of the Ventrolateral Prefrontal Cortex in Offspring of Parents With Bipolar Disorder: Assessing Markers of Risk and Resilience

Danella Hafeman*, Henry Chase, Lisa Bonar, Kelly Monk, Mary Beth Hickey, Alicia McCaffrey, Tina Goldstein, Benjamin Goldstein, David Axelson, Boris Birmaher, Mary Phillips

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: Offspring of parents with bipolar disorder (BD) are at risk for BD themselves, as well as other psychopathology. Understanding neural correlates of risk and resilience in these at-risk youth might help us to identify individuals who are at "ultra"-high risk of the disorder, as well as better understand the neural underpinnings of the disorder. Both structural and functional MRI studies have identified the ventrolateral prefrontal cortex (VLPFC; Brodmann Area 47) as a region associated with both family risk and BD onset. We assessed how resting state functional connectivity (rsFC) between the right and left VLPFC and other brain regions differed between offspring of parents with bipolar disorder (OBP) vs. offspring of parents with other psychopathology (OCP). Furthermore, we tested whether rsFC in regions showing group differences also correlated with risk of new-onset BD, using a recently published risk calculator for 5-year risk of new-onset bipolar disorder in OBP (Hafeman et al., JAMA Psychiatry 2017). In this way, we aimed to assess whether rsFC with VLPFC might serve as a marker for either risk or resilience in these youth.

Methods: After excluding those with excess movement (>3mm) and/or mean frame-wise displacement (a measure of frame-to-frame movement; $>.5$mm), we used rsFC data from 27 OBP and 23 OCP (ages 8-17 years old) from the Bipolar Offspring Study (BIOS) longitudinal cohort. A standard preprocessing pipeline was implemented in Nipype, which included realignment, coregistration, fieldmap correction, normalization (using DARTEL), despiking, and smoothing; SPM8 was used for the first-level analyses, using right and left VLPFC seeds. After adjusting for age, gender, mean frame-wise displacement, and psychiatric diagnosis, we tested whether there were any regions where rsFC with the right or left VLPFC was associated with a family history (OBP vs. OCP); and, secondarily, whether any voxels in these regions correlated with risk score in OBP (using a recently published risk calculator). We first created a mask of regions associated with OBP vs. OCP (voxel-wise $p<.001$, $k>100$); next, we tested for correlation with risk score (in the OBP) within this mask (voxel-wise $p<.001$).

Results: Compared to OCP, OBP showed higher rsFC between the right VLPFC and both the right thalamus and left inferior parietal lobe (IPL) (voxel-wise $p<.001$; $k>100$). The peak voxel in the left IPL survived whole-brain correction (peak voxel = -42 -28 24; FWE-corrected $p=.038$); the peak voxel in the right thalamus approached significance (peak voxel = 20 -24 2; FWE-corrected $p=.095$). Within the mask of regions showing group differences (OBP vs. OCP; uncorrected $p<.001$), there was a small cluster in the right thalamus (pulvinar region) where rsFC to right VLPFC was also positively correlated with risk score within the OBP (20 -26 6; $k=3$, uncorrected $p<.001$). For the left VLPFC seed, we did not find any regions that showed differential rsFC for the OBP vs. OCP comparison, or a correlation with risk score.

Conclusions: We found differential rsFC in offspring of parents with BD (compared to offspring of individuals with other psychiatric disorders), in particular, between right VLPFC and left IPL. In addition, we found a small cluster in the right thalamus that showed more connectivity to the right VLPFC in the OBP vs. OCP, and also was positively correlated with risk of new-onset bipolar disorder within the OBP. While the latter finding is exploratory and does not survive whole brain correction, a region showing this pattern

of results might represent a marker of risk in these youth. The VLPFC, thalamus, and IPL are part of a network key to executive function, which has been found to be abnormal in individuals with and at-risk for BD. The thalamus has been shown in some studies (but not all) to have both volumetric and functional differences between individuals with BD and healthy controls, and abnormalities have been found to be correlated with depressive symptoms. Limitations of this analysis include a relatively small sample size, particularly for the risk score analysis, and risk scores that were right skewed; only six individuals had a predicted risk of $>10\%$. We also do not know the extent to which our findings are driven by particular diagnoses and symptoms, as the sample size is too small to conduct further sub-analyses. Future work will assess correlations between rsFC and risk score in a larger sample, with a more complete distribution of risk patterns.

Keywords: Bipolar Disorder, Resting-State fMRI, Familial Risk

Disclosure: Nothing to Disclose.

T25. Assessing the Role of SKA2 in Stress-Associated Psychiatric Disorders

Jakob Hartmann*, Harry Pantazopoulos, Claudia Klengel, Kenneth McCullough, Rhomberg Thomas, Jordan Walton, Torsten Klengel, Sabina Berretta, Kerry Ressler

Harvard Medical School, Belmont, Massachusetts, United States

Background: Mood and anxiety disorders represent a major disease and social burden worldwide, but the underlying molecular mechanisms are still poorly understood. In recent years, evidence has emerged for the crucial role of genes involved in the regulation of the hypothalamic-pituitary-adrenal (HPA) axis, especially in the context of stress-related psychopathologies such as anxiety and depression. The glucocorticoid receptor (GR) is the main mediator of the negative feedback loop of the HPA axis in response to stress. The Ska2 gene, encoding the spindle and kinetochore associated complex subunit 2, has previously been identified as GR interaction partner. Interestingly, single nucleotide polymorphisms and epigenetic status within the Ska2 gene, as well as gene expression alterations, have been associated with posttraumatic stress disorder and suicide risk in several studies in the past. Yet, little is known about the underlying molecular mechanisms and the role of Ska2 in the brain. Therefore, we set out to further investigate the role of Ska2 in the CNS and validate it as a potential candidate gene in the context of stress-associated psychiatric disorders.

Methods: We performed Immunohistochemistry (IHC) to study the expression pattern of Ska2 in human postmortem amygdala samples and in the mouse brain. In addition, we conducted western blot analysis to investigate Ska2 protein expression in basolateral amygdala (BLA) samples of individuals with bipolar depression and matched controls. Stress-induced changes in Ska2 mRNA expression in mice were investigated via qPCR following fear conditioning.

Results: IHC analyses in postmortem human amygdala samples revealed a prominent expression of Ska2 protein in Vglut1-positive, glutamatergic pyramidal, neurons. Furthermore, we detected significantly increased Ska2 protein levels in the BLA of individuals with bipolar depression compared to matched controls (ANCOVA, $F = 5.83$, $p < 0.025$, $n = 14$ per group). Detailed mapping and co-labeling studies in mice also revealed a distinct pattern of Ska2 expression in neurons of the BLA, as well as in the hippocampus (HC), mediodorsal thalamus, the paraventricular nucleus of the hypothalamus and throughout the cortex. Most of the Ska2-positive neurons also expressed the GR. Consequently, we assessed whether stress is able to modulate Ska2 gene expression. Using qPCR, we found dynamic changes of Ska2 mRNA expression four hours after stress (fear conditioning, 5 tone/foot shock pairings). Stressed mice showed significantly decreased Ska2 mRNA levels in the HC ($T(18) = 2.446$, $p < 0.05$, $n = 8$ (ctrl), $n = 12$ (stress)) and increased levels in whole amygdala punches ($T(15) = 2.693$, $p < 0.05$, $n = 5$ (ctrl), $n = 12$ (stress)) compared to baseline controls (home cage group).

Conclusions: Together these findings suggest that Ska2 is expressed in GR-containing neurons throughout regions involved in emotion processing and that the Ska2 gene is dynamically regulated in these brain areas following stress exposure. Collectively, our results point to an important, and thus far unappreciated, role of Ska2 in stress-related psychiatric disorders, which is relevant to our understanding of the molecular mechanisms underlying such diseases. We further aim to manipulate Ska2 gene expression in the mouse brain using viral vectors in order to investigate its causal role in regulating stress- and anxiety-related behaviors.

Keywords: Bipolar Disorder, PTSD, Suicide, HPA Axis, Glucocorticoid Receptor

Disclosure: Nothing to Disclose.

T26. Aging- and Stress-Related Epigenetic Disinhibition of FKBP5 Contributes to NF-KB-Driven Inflammation and Cardiovascular Risk

Antonios Zannas*, Meiwen Jia, Jens Baumert, Kathrin Hafner, Maik Ködel, Andreas Haehle, Stella Iurato, Tania Carrillo-Roa, Rebecca Emeny, Jari Lathi, Katri Räikkönen, Johan Eriksson, Amanda Drake, Melanie Waldenberger, Simone Wahl, Sonja Kunze, Susanne Lucae, Bekh Bradley, Christian Gieger, Felix Hausch, Alicia Smith, Kerry Ressler, Bertram Müller-Myhsok, Karl-Heinz Ladwig, Theo Rein, Nils Gassen, Elisabeth Binder

Max Planck Institute of Psychiatry, Munich, Germany

Background: Aging and stress-related phenotypes are both associated with heightened inflammation and disease risk, including cardiovascular disease, but the underlying molecular mechanisms are unknown. Our objective was to examine the role of the stress-responsive immunophilin FKBP5 in these relations.

Methods: Peripheral blood DNA methylation was measured across the FKBP5 gene in four independent cohorts: the Grady Trauma Project (GTP, $n = 393$); the Cooperative

Health Research in the Region of Augsburg (KORA; $n = 1,727$); the Max Planck Institute of Psychiatry (MPIP; $n = 538$); and the Helsinki Birth cohort study ($n = 160$). Genome-wide gene expression was assessed in peripheral blood in the GTP ($n = 355$). Functional protein interaction, reporter gene, and ELISA experiments were performed in cellular models of immune function.

Results: FKBP5 DNA methylation consistently decreased with age at selected CpGs, and this age-related demethylation was accelerated by childhood trauma ($p = 7.4$ x 10-3) and depressive symptoms (interaction $p = 3$ x 10-2) and was associated with increased FKBP5 mRNA ($p = 1.6$ x 10-2) and stronger cortisol-FKBP5 relationship (interaction $p = 1.4$ x 10-3). In peripheral blood, FKBP5 upregulation correlated with a proinflammatory profile and extensive changes in NF-κB-related gene networks. In accordance, FKBP5 overexpression in immune cells promoted chemokine secretion and strengthened the interactions of regulatory kinases of the NF-κB pathway. Notably, the same age-and stress-related CpGs associated with FKBP5 upregulation were also less methylated in subjects with a history of myocardial infarction in both the KORA ($p = 4.4$ x 10-2) and MPIP ($p = 3.1$ x 10-2).

Conclusions: These findings identify FKBP5 as a mediator of stress-augmented peripheral inflammation with aging and potential contributor to cardiovascular risk.

Keywords: Aging, Epigenetics, Inflammation, Stress

Disclosure: Nothing to Disclose.

T27. A 6-Week Open-Label Study of Treatment of Insomnia With Zaleplon in HIV Positive Patients With Comorbid Depression

Anup Sharma*, Arjun Sengupta, Lynne Taylor, Knashawn H. Morales, Tyler M. Moore, Ruben C. Gur, Michael E. Thase, Aalim Weljie, Matthew S. Kayser

University of Pennsylvania, Philadelphia, Pennsylvania, United States

Background: An estimated >50% of HIV positive individuals report insomnia. Insomnia and other sleep disturbances negatively impact outcomes in this population by contributing to metabolic dysfunction, reducing quality of life, and impairing psychosocial functioning. Additionally, in HIV depressed populations, insomnia may hinder remission from clinical depression. To date, there have been no pharmacotherapeutic studies treating insomnia in this population, emphasizing the need to determine whether known hypnotic medications work as effectively in HIV positive patients, including those with comorbid depression. Non-benzodiazepine hypnotics represent an attractive pharmacotherapy for treatment of insomnia in a depressed population, as there are reduced side effects and abuse potential compared to classical benzodiazepines. Here, the authors conduct a 6-week open-label study evaluating zaleplon for insomnia in 20 HIV positive patients with comorbid depression.

Methods: HIV patients with comorbid clinical depression (defined by DSM-IV-TR criteria) were treated with zaleplon 5-10mg daily for insomnia. The primary efficacy end point was change in insomnia severity index (ISI) from baseline to 6 weeks. The key secondary efficacy end point was change in Epworth sleepiness scale (ESS) from baseline to 6 weeks. Additional secondary outcomes included changes in mood symptoms as measured by the Quick Inventory of Depressive Symptomatology (QIDS) and the 24-item Behavior and Symptom Identification Scale (BASIS-24) depression subscale scores, changes in cognitive symptoms as measured by the Penn Computerized Neurocognitive Battery (CNB), and changes in metabolomic measures at baseline and 6-week visits. Treatment effects and change associations were estimated using linear mixed effects models and spearman correlations.

Results: There were no treatment emergent adverse events. Analyses in $n = 20$ subjects with HIV and co-morbid depression treated with zaleplon showed a significant reduction in ISI total score from baseline (20.75) to week 6 (11.47, $P < 0.0001$). Treatment with zaleplon also significantly improved secondary outcome measures including mean ESS scores (baseline: 11.10, week 6: 6.97, $P = 0.0013$), mean QIDS scores (baseline 11.87, week 6: 8.54, $P = 0.0066$), and mean BASIS-24 depression subscale scores (baseline: 1.80, week 6: 1.41, $P = 0.0202$). Improvements with the ISI were associated with improvement in CNB efficiency scores, and were significant ($P < 0.05$) for emotion recognition and verbal reasoning. Improvements with the ISI were also associated with significant changes ($P < 0.05$) in branched chain amino acid metabolites from baseline to week 6.

Conclusions: A 6 week open-label treatment with zaleplon in HIV+ depressed subjects significantly improved sleep outcomes including insomnia severity and daytime sleepiness. The intervention also significantly improved key self-report measures of depression from baseline to endpoint. Improvements in the primary sleep outcome were associated with improvements in cognitive function as measured by the Penn CNB. Concomitant changes in the primary sleep outcome were associated with significant changes in key branched chain amino acid metabolites that may serve as useful biomarkers of treatment response.

Keywords: HIV, Depression, Insomnia, Biomarker

Disclosure: Nothing to Disclose.

T28. An Epigenetic Biomarker for Depression and Trait of Childhood Trauma With Sex-Specific Effects

Carla Nasca*, Betty Bigio, Francis Lee, Marin Kautz, Sarah Young, Ashly Cochran, James Beasley, David Millington, James Kocsis, James Murrough, Bruce McEwen, Natalie Rasgon

Rockefeller University, New York, New York, United States

Background: Development of diagnostics and more effective treatments for major depressive disorders (MDD) can be guided by mechanistic insights from animal studies. These are urgent medical and public health needs given that MDD is now the leading cause of illness and disability worldwide with a large economic burden. There is a broad literature from our and other groups supporting rapid antidepressant effects of the epigenetic modulator of glutamatergic function with insulin-sensitizing properties, acetyl-L-carnitine (LAC), across animal models. A common trait of such animal

models with depressive and metabolic-like dysfunction was an endogenous reduction in LAC levels in the plasma and in mood regulatory brain regions, such as the hippocampus and prefrontal cortex. Therefore, based upon several converging preclinical evidence, we started by investigating the association between LAC and MDD in humans with a targeted-oriented and mechanistic-driven approach.
Methods: Plasma distribution of LAC and the internal control free-carnitine were determined in 45 healthy controls (HC) and 71 patients suffering with MDD using ultra-performance liquid chromatography-tandem mass spectrometry (UPLC-MS/MS), and stable isotope dilution electrospray-tandem mass spectrometry (ESI-MS/MS). Study participants were recruited at two independent sites, the Weill Cornell Medicine and the Mount Sinai Icahn School of Medicine. The psychiatric examination included the Structured Clinical Interview for DSM-IV (SCID, MINI), the two psychiatric scales, HDRS-17 and MADRS. Study participants completed the Childhood Trauma Questionnaire (CTQ) to assess the effects of childhood traumatic experiences, i.e., physical, sexual and emotional abuse and physical and emotional neglect. All patients with MDD were in an acute episode during study participation. Two-tailed t-tests, chi-square, Pearson correlations and multiple regression were used as appropriate to specific analyses.
Results: HC and patients with MDD did not differ on any demographic and clinical characteristics, except that, as expected, patients with MDD had significantly higher depression severity scores as assessed with the two psychiatric scales, HAM-17 and MADRS (Ham-17: HC = 0.6+/-1.1, MDD = 20.2+/-3.3, $p < 0.0001$; MADRS: HC = 1.4 +/-2.4, MDD = 32.9+/-4.7, $p < 0.0001$).
Supported by validation and replication data, our findings show that LAC (and not free-carnitine) is significantly lower in patients with MDD compared to HC ($p < 0.0001$, power 0.99, effect size = 0.8). Within the group of patients with MDD, LAC was lower in patients who exhibited greater severity and earlier age of onset of MDD, independently of psychotropic drug treatment. These correlations remained significant controlling for number of past episodes, length of current episode, sex, and age. Consistent with lower LAC levels in a more severe clinical phenotype of MDD (i.e., greater severity, earlier age of onset), we report that in those patients with treatment resistant depression (TRD), reduced LAC was predicted by emotional neglect, and being a female.
Conclusions: Our new findings suggest a role for LAC, an epigenetic modulator of glutamatergic function with insulin-sensitizing properties, as marker to delineate a biologically-defined MDD phenotype, providing a potential biological target for a precision medicine approach and rational path forward for development of novel pharmaceutical agents. Future studies will test whether such severe phenotype of MDD could benefit by treatment with LAC. Given our earlier reported association of reduced LAC with IR in animals with depressive-like behaviors, it will also be important to study if LAC acts as a gating biomarker for depression encompassing other disease targets.
Keywords: Depression, Treatment Resistant Depression, Childhood Trauma, Sex Differences, Insulin Resistance
Disclosure: Nothing to Disclose.

T29. A Double-Blind Randomized Placebo-Controlled Study of Aspirin and N-Acetyl Cysteine as Adjunctive Treatments for Bipolar Disorder Patient: Preliminary Findings

Isabelle Bauer*, Charles Green, Gabriela D. Colpo, Kathryn Durkin, Sudhakar Selvaraj, Antonio L. Teixeira, Giovana B. Zunta-Soares, Jair C. Soares

University of Texas Medical School, Health Science Center at Houston, Houston, Texas, United States

Background: Inflammation, increased oxidative stress and consequent deleterious effect on brain circuitry have been proposed as putative mechanisms involved in the pathophysiology of bipolar disorder (BD). Further support comes from research showing that non-steroidal anti-inflammatory drugs (NSAID) have promising antidepressant effects. Preclinical and clinical findings indicate that the antioxidant N-acetylcysteine (NAC) enhances the effects of NSAID. No study has, however, tested the adjunctive therapeutic benefits of NSAIDs and NAC in BD.
Methods: 24 medicated BD patients aged 18-65 years and with a Montgomery-Åsberg Depression Rating Scale (MADRS) score >20 were randomly assigned to receive either aspirin (1000 mg; 2 capsules of 500 mg), NAC (1000 mg; 2 capsules of 500 mg), combined aspirin and NAC, or placebo (sugar pill). Treatment was adjunctive to the ongoing treatment regimen for a 16-week period. Successful treatment outcome was defined as a $\geq 50\%$ reduction in baseline MADRS scores. We adopted an adjusted adaptive randomization approach at eight weeks to minimize the number of participants assigned to non-viable treatments. A Bayesian analytical method was adopted and posterior probability distributions were calculated to determine the probability of treatment response. This study is registered on ClinicalTrials.gov (#NCT01797575).
Results: Following the first 8-week treatment phase, NAC +aspirin and placebo had similar probability for successful treatment response (70%). They both had greater probability of treatment response compared to NAC+Placebo (60%) and Aspirin+Placebo (50%). Following a 16-week treatment period, NAC+Aspirin was associated with higher probability of treatment response (67%) compared to Placebo (55%), NAC +Placebo (57%), and Aspirin+Placebo (33%). NAC and Aspirin were safe and no life-threatening side effects were reported.
Conclusions: These preliminary findings suggest that the co-administration of NAC and Aspirin may be beneficial in decreasing the severity of depressive symptoms in BD patients. The Bayesian approach allowed us to draw probabilistic inferences, even in the context of small sample sizes, and provided an estimate of the relative success of the respective treatments.
Keywords: Aspirin, N-Acetyl-Cysteine, Bipolar Disorder, MADRS, Neuroinflammation
Disclosure: Nothing to Disclose.

T30. Emotion Regulation, Brain Structural Connectivity, and Affective Behavior in Comorbid Major Depressive Disorder and Obesity

Matthew Sacchet*, Adam Pines, Monica Kullar, Jun Ma, Leanne M. Williams

Stanford University, Stanford, California, United States

Background: Depression is a common and disabling psychiatric condition that is often comorbid with prevalent physical conditions such as obesity, with reciprocal adverse effects on treatment of either. Emotion regulation is a core component of behavior change related to treatment response in affective disorders and obesity. Expressive suppression is an emotion regulation technique that is associated with depression and poor outcomes to treatment. Studies in non-disordered populations suggest that brain structure, including the white matter of the uncinate fasciculus, underlies emotion regulation abilities. Here, in a cohort of obese depressed adults, we aimed to understand targets for behavior change that might facilitate improved treatment response in affective disorders and obesity by quantifying relations across levels of analysis related to emotion regulation. We assessed relations among structural integrity of the uncinate fasciculus as quantified by fractional anisotropy (FA) computed from diffusion weighted imaging, the tendency to use an expressive suppression emotion regulation technique, behavioral indices of emotion processing, and symptom severity.
Methods: This study uses baseline data from a neuroscience study that is operationally linked with a type 1 effectiveness-implementation hybrid design clinical trial of an integrated, technology-enhanced, collaborative care model for treating adult patients with comorbid obesity and depression in primary care. Whole-brain diffusion-weighted and T1-weighted images were collected. FA of the uncinate fasciculus was quantified using automated Fiber Quantification (AFQ), a recently developed approach to automated DWI tractography clustering. Trait use of suppression strategies for regulating emotion was measured using the Emotion Regulation Questionnaire (ERQ). Behavioral measures of emotion processing were obtained from a web browser-based task designed to evaluate emotional biases by quantifying the speed at which individuals identify facial emotions in images (happy, sad, disgust, anger, fear, and neutral). "Negative" emotion reaction time was the average reaction time to the four presented negative emotions: sadness, disgust, fear, and anger. In the event of significant relations between aggregate negative emotions subsequent analyses were conducted for each emotion individually. The Symptom Checklist (SCL-20) and Patient Health Questionnaire-9 (PHQ-9) were used to measure depression severity, and the Generalized Anxiety Disorder-7 (GAD-7) was used to measure generalized anxiety disorder. Relations among variables were assessed using Pearson or Spearman correlations depending on data normality.
Results: Seventy-seven participants (22-76 years old) had complete neuroimaging data for this study. AFQ identified left and right uncinate for 75 of these participants. The software's failure to do so on these two participants could have been due to noise in the data, crossing fibers, abnormal anatomy, or a limited number of fibers detected within the tracts of interest. One participant did not complete the cognitive battery, and one did not complete the questionnaire battery. Right uncinate FA was significantly positively correlated with ERQ Expressive Suppression scores ($r = 0.27$, $p = 0.022$), but not left uncinated FA ($r = -0.08$, $p = 0.500$). Right uncinate FA was also significantly correlated with reaction time to negative faces ($r = -0.26$, $p = 0.026$) such that higher FA corresponded to faster reaction times. Left uncinate FA was not related to reaction times to negative faces ($r = -0.21$, $p = 0.073$). Subsequent analyses revealed that right uncinate fasciculus FA was significantly negatively correlated with reaction time to fearful faces ($r = -0.35$, $p = 0.003$) and sad faces ($r = -0.30$, $p = 0.010$). Right uncinate FA did not significantly correlate with reaction time to disgust faces ($r = -0.15$, $p = 0.218$) or to anger faces ($r = -0.14$, $p = 0.241$). As age may confound FA and reaction time correlations, we also ran partial correlations between our variables of interest while controlling for age. Right uncinate FA and averaged negative face reaction time ($r = -0.25$, $p = 0.037$), fear reaction time ($r = -0.30$, $p = 0.011$), and sad reaction time ($r = -0.24$, $p = 0.047$) remained significant. Neither right nor left uncinate FA correlated with scores on the SCL-20, PHQ-9, or GAD-7 ($|r| \leq 0.16$, $p \geq 0.180$).
Conclusions: We found that FA of the right uncinate fasciculus was significantly positively correlated with the tendency to use expressive suppression emotion regulation, and with faster reaction times to negative faces, in particular fearful and sad faces. These findings suggest that FA of the uncinate fasciculus tracks maladaptive emotion regulation that is relevant to affective disorder psychopathology and obesity, and that interventions that account for these multi-level associations may prove useful for improving clinical outcomes for these conditions.
Keywords: Depression, Obesity, Emotional Regulation, Brain Structure, Behavior
Disclosure: Nothing to Disclose.

T31. Striatal Neuromodulation: From Single Synapse to the Proteome

Yevgenia Kozorovitskiy*, Nicholas Bannon, Vasin Dumrongprechachan

Northwestern University, Evanston, Illinois, United States

Background: The vertebrate brain is composed of intricate circuits of neurons that communicate via electrical signals, mediated by fast excitatory or inhibitory neurotransmission and ion channel opening. Superimposed over this circuit layout is a diverse set of instructive neuromodulatory signals, acting primarily through G protein coupled receptors (GPCRs). Neuromodulatory effects are temporally and mechanistically diverse: ranging from milliseconds to hours, they include the opening of nearby ion channels, kinase-driven phosphorylation of proteins, and altered gene expression.
To interrogate neuromodulation at many scales our lab uses and develops new optical and proteomic techniques, complementing them with classical electrophysiology and anatomical assays. We focus primarily on the basal ganglia, subcortical nuclei essential for reward-based learning and complex motor actions, which reside in a rich neuromodulatory niche.
Methods: We rely 2-photon "uncaging" of glutamate and 2-photon imaging of dendrites to create de novo new dendritic spines and synapses on genetically targeted striatal spiny projection neurons (SPNs) located within their circuits. My prior work has delineated the function of glutamate release in the genesis of neuronal connections and suggested the

importance of modulation in choosing where and how readily connections form. Green or red fluorescent proteins, sometimes together with calcium reporters, are virally expressed in striatal direct or indirect pathway SPNs. We focus on dopamine and adenosine signaling in SPNs, genetically targeted using mouse lines expressing Cre recombinase under the promoters of Drd1 type dopamine receptor and A2a type adenosine receptor. Imaging and spinogenesis are carried out in the acute slice using a dual laser 2-photon microscope. The propensity for focal de novo synaptogenesis and the longevity of new dendritic spines are evaluated under distinct neuromodulatory conditions, at refined spatiotemporal scales.

To delve into the broader, longer time-scale consequences of neuromodulatory activity we develop and utilize methods for genetically targeted neuroproteomics – a level of analysis that is inaccessible by other technical methods. We adapted a new flexible in vivo tagging method to bring genetically targeted proteomics to any mammalian neural circuit, together with refined control of neural circuit activity. This tool capitalizes on Cre recombinase-driven expression of a biotinylating soybean peroxidase Apex. Following induction of biotinylation in the acute slice, tagged proteins are affinity-enriched, identified and quantified using isobaric tagging or label-free quantification mass spectrometry.

Results: We found that separate neuromodulatory cascades acting through protein kinase A (PKA) compete in regulating the birth of new neuronal connections. This occurs on sub-cellular spatial scales and involves GPCRs that bidirectionally control PKA activity (those coupled to Gαi and Gαs type G proteins). Our pilot mass spectrometry runs show that activating Drd1 type dopamine receptors on dSPNs increases the abundance of many proteins associated with neuronal signaling and plasticity (e.g., ankyrin, homer, cofilin, synaptophysin, β-catenin, etc.).

Conclusions: We are in the process of defining the full logic of GPCR signaling control over dendritic spine and synapse genesis at high spatiotemporal scales. In addition to rapid signals that alter the structure and function of striatal neurons, modulators like dopamine also reshape the overall neuronal proteomic landscape. Over the long term, these studies will help facilitate the development of therapeutic applications, harnessing the power of neuromodulators to functionally reconfigure, and sometimes even literally rewire, neural circuits.

Keywords: Striatum, Neuromodulation, Proteomics, GPCRs, Two-photon

Disclosure: Nothing to Disclose.

T32. Patterns of Variability in Dopamine Synthesis and Receptor Availability in Healthy Humans are Associated With the Valuation of Time

Angela Ianni*, Daniel Eisenberg, Sara Constantino, Catherine Hegarty, Philip Kohn, Tim Behrens, Karen Berman

National Institutes of Health, Bethesda, Maryland, United States

Background: Human behavior is guided by an intrinsic desire to maximize rewards, or positive experiences, and minimize negative outcomes. An accurate valuation of time is essential to accomplish this. Prior work has implicated dopamine in the neural circuitry underlying time estimation (Buhusi and Meck, 2005). Specifically, it has been shown that both dopamine D2 receptor antagonists and disorders affecting the dopamine system such as Parkinson's disease and schizophrenia affect interval timin (Buhusi and Meck, 2005; Meck 1983). However, it remains unclear whether presynaptic dopamine function or D1 receptors play a role in interval time estimation.

Another way to measure the neural basis of time valuation is by studying foraging behavior, in which how an animal decides to spend its time searching for food is critical for survival. Computational work has predicted that tonic dopamine encodes the average reward rate of the environment, and thus the loss of potential gain when time is wasted, but, experimental confirmation is lacking (Niv et al, 2007). Here, we used PET to characterize variation in pre- and post-synaptic measures of the dopamine system in healthy humans, and tested the relationship of these patterns of dopamine function to behaviors associated with the valuation of time: interval timing and foraging behavior.

Methods: Fifty-seven healthy adults (mean age 35.4 ± 10.3 years; 29 women) were screened by a physician to rule out psychiatric, neurological, and other major medical illnesses. In these participants, we used PET imaging to directly measure three dopamine-related parameters: presynaptic synthesis capacity (Ki), related to tonic dopamine, (with [18F]-DOPA); D1 receptor binding potential (BPND) (with [11C]-NNC112); and D2/3 receptor BPND (with [18F]-Fallypride). In addition, we measured reward-guided foraging behavior using a computer-based task in which participants gathered apples from trees in four different reward environments. The reward environments varied in two parameters: travel time between trees and decay rate of apples within a tree. Participants decided between continuing to gather apples at the current tree or traveling to a new tree. The threshold at which the decision to leave occurs is called the exit threshold. Furthermore, since successful foraging depends on weighing the benefits of leaving for a new tree with the costs of losing time while traveling, we also quantified subjects' ability to estimate a three second interval (interval timing accuracy) using a computer task that mimics a stop-watch.

PET data were extracted from basal ganglia regions of interest (dorsal putamen, dorsal caudate nucleus, ventral striatum, and midbrain), residualized for age and converted to z-scores. A principal component analysis (PCA) was run on the z-normalized PET values to identify patterns of variation in the three dopamine-related parameters. Finally, to identify which patterns of dopamine variation contributed to the reward-guided behavior, SPSS was used to run Pearson correlations between the behavioral parameters and PCA components that had eigenvalues greater than 0.5. A statistical threshold of $p < 0.05$ was used to assess significance.

Results: The PCA analysis yielded five patterns of the PET data that account for 36, 30, 16, 7, and 6 percent of the variance, respectively. The components can be generally described as follows: (1) low tonic dopamine with high D1 and D2/3 BPND, (2) high tonic dopamine and D2/3 BPND with low D1 BPND in the ventral striatum, (3) high D1

BPND and low D2/3 BPND throughout the striatum along with high tonic dopamine in the putamen, (4) high dorsal striatum tonic dopamine with low ventral striatum and midbrain tonic dopamine and low D1 BPND in the ventral striatum, and (5) high midbrain tonic dopamine and D2/3 BPND in the putamen with low tonic dopamine in the caudate nucleus and low D1 and D2/3 BPND in the ventral striatum.

Individuals changed their exit threshold between the four different reward environments: highest threshold (5.61.7) in the most rewarding environment and the lowest threshold (4.21.4) in the least rewarding environment ($p = 3.7e\text{-}7$). There was a negative correlation between component 3 score and the amount that individuals changed their exit threshold between the reward environments with a shallow reward decay rate compared to those with a steep decay rate ($r = -0.365$, $p = 0.022$). In addition, individuals decreased their reaction time in the most rewarding environment compared to the others (all $p < 0.01$). The amount that reaction time decreased between the least and most rewarding environments was negatively correlated with both components 1 ($r = -0.383$, $p = 0.016$) and 4 ($r = -0.431$, $p = 0.006$). Finally, there was a positive correlation between interval timing and component 4 scores ($r = 0.404$, $p = 0.013$) such that individuals with higher component 4 scores had a longer perception of time.

Conclusions: We identified five patterns of variation in the basal ganglia of dopamine synthesis capacity and receptor availability in healthy humans. In line with prior work (Buhusi and Meck, 2005), we found an important role of striatal dopamine in interval timing. Furthermore, we found a dopaminergic basis to changes in both the foraging exit threshold and the reaction time between the least and most rewarding environments. This work provides insight into the neural mechanisms underlying reward-guided behavior and may have implications for disorders involving dysfunction of the dopamine system.

Keywords: Dopamine, Timing, Patch Foraging

Disclosure: Nothing to Disclose.

T33. COX-1 is Constitutively Expressed and COX-2 is Inducible: In Vivo PET and In Vitro Immunostaining Studies in Acute Model of Neuroinflammation in Rhesus Macaque

Stal Shrestha*, Robert Innis

National Institute of Mental Health, Bethesda, Maryland, United States

Background: Our laboratory recently developed two PET radioligands: 11C-PS13 for COX-1 and 11C-MC1 for COX-2, each of which potently and selectively inhibits the cognate enzyme in whole blood assays from monkey and human. Based on studies in peripheral organs, COX-1 is typically present at baseline (i.e., constitutively expressed) but not induced (i.e., upregulated) by inflammation. In contrast, COX-2 is minimally expressed at baseline in several peripheral tissues but markedly upregulated by inflammation at the level of both gene transcription and protein synthesis. In our presentation last year at the ACNP, we reported a) the pharmacological characterization of the COX-1 and COX-2 radioligands using in vitro enzymatic assays in monkey and human blood, and b) exploratory studies of these two radioligands in a healthy vs. inflamed rhesus monkey brain. We showed that PS13 was potent and selective for COX-1 (IC50 = 1 nM) compared to COX-2 (IC50 > 1,000 nM). Conversely, MC1 was potent and selective for COX-2 (IC50 = 3 nM) compared to COX-1 (IC50 > 1000 nM). Both 11C-PS13 and 11C-MC1 showed good uptake in monkey brain (peak concentrations of 3 – 5 SUV) and washed out relatively quickly (demonstrating that the binding was reversible, as expected). The purpose of this study was to determine whether a model on neuroinflammation (i.e., intracerebral injection of lipopolysaccharide (LPS)) would upregulate expression of COX-2 but not COX-1.

Methods: To induce transient inflammation, LPS (from Escherichia coli O26:B6) was injected into the right putamen of monkeys ($n = 6$). Prior to injection, we obtained a T1-weighted MRI to obtain anatomical information to guide the placement of the cannula. We injected a total dose of 10 μg of LPS at a concentration of 1 μg/μl, and an infusion rate of 0.5 μl/min. Thus, the entire dose was administered at a final volume of 10 μl over a period of 20 minutes. The infusion cannula was left in place for an additional 10 min post-injection. This technique allows the LPS to diffuse ~ 3 mm from the site of injection. About 220 MBq of each radioligand was injected intravenously into rhesus monkeys. Dynamic PET scans of brain were acquired for two hours pre- as well as post-LPS infusion. Standardized uptake value (SUV) was calculated from 60-120 min, and was used as the outcome measure for calculating 1) specific uptake at baseline, and 2) per cent difference between pre- vs. post-inflammation. Full quantitation of distribution volume (VT) was done with arterial sampling for accurate measurement of the concentration of parent radioligand in plasma. Fluorescent in situ hybridization, immunostaining, and western blot were done in post-mortem tissues to confirm the in vivo PET findings.

Results: 11C-PS13 showed specific uptake in healthy monkeys with 45% of the binding displaced by non-radioactive PS13 (0.3 mg/kg). However, compared to pre-injection, there was no increase in uptake post-injection of LPS on either Day 1 or 3. On the contrary, 11C-MC1 showed no noticeable specific uptake in healthy monkeys (i.e. pre-injection). Most importantly, after post-injection of LPS the per cent specific uptake of the COX-2 radioligand was 41% on Day 3, and 35% on Day 8. In addition, using full quantitation, VT of MC1 increased by 45% post-LPS, while VT of PS13 showed no significant change. Since both of the COX-1 and COX-2 radioligands are exploratory markers of neuroinflammation, we also confirmed the location of the edema using a T2-weighted MRI and inflammation using 11C-PBR28 radioligand for translocator protein (TSPO). COX-2 mRNA and protein were upregulated in inflamed brain and located primarily in neurons.

Conclusions: Our results suggest that COX-1 is constitutively expressed and that COX-2 is induced in monkey brain by intracerebral injection of the inflammagen, LPS. We used brain uptake of the radioligand to measure binding to COX, and confirmed with full quantitation. 11C-MC1 is the first radioligand to show promise as a marker of COX-2, and 11C-PS13 is the first direct-acting radioligand for COX-1. COX-2 mRNA and protein are upregulated in inflamed brain

and located primarily in neurons. First in-human studies are currently ongoing and can be used to measure COX-1 and COX-2 in disorders thought to involve neuroinflammation and to measure the delivery to the brain of non-steroidal anti-inflammatory drugs (NSAIDs) as well as their selectivity for the two COX isoforms.
Keywords: Neuroinflammation, PET Imaging, Biomarker
Disclosure: Nothing to Disclose.

T34. Cerebellar D1DR-Expressing Neurons Modulate the Frontal Cortex During Timing Tasks

Kelsey Heslin, Kyle Walsh, Jonah Heskje, Erik Carlson, Krystal Parker*

University of Iowa, Iowa City, Iowa, United States

Background: Cerebellar stimulation improves cognitive function in patients with schizophrenia and in our rodents with D1 dopamine receptors (D1DRs) blocked in the medial frontal cortex (MFC). However, the precise cerebellar contribution to cognition remains unknown. We study cerebellar dependent cognitive processing using an interval timing task. Timing is an essential biological process that serves as a window into cognitive function as it requires executive processes such as working memory, attention, and planning and is known to rely on both the frontal cortex and cerebellum. We have previously shown that cue-evoked ramping activity, or monotonic increases or decreases of firing rate over time in single neurons in the MFC and lateral cerebellar nuclei (LCN) are essential for efficient time estimation. It has been suggested that a specific subtype of neurons in the LCN expressing D1DRs are essential for cognitive function. To further explore how the cerebellum influences the frontal cortex and the role of D1DR-expressing neurons in the LCN, we block D1DRs in the LCN in animals performing an interval timing task and record activity in the frontal cortex. We hypothesize that blocking D1DRs in the LCN will impair performance on the task and attenuate ramping activity in single neurons in the MFC indicating a potential role for this population of cerebellar neurons in cognition.
Methods: We use an interval timing is a task to study cognitive function which requires rats to make a motor indication (lever press) of their estimation of the passage of a specified duration of time (12 seconds). A house light on in the chamber signifies the start of the trial and any presses that occur before 12 seconds have elapsed are not rewarded or punished. A water reward is provided for the first lever press that occurs following the end of the 12 second interval. Animals well trained in the task are implanted with a microwire array in the left MFC and with an infusion cannula in the right LCN. To investigate how cerebellar D1DR-expressing neurons influences frontal cortical activity and performance on the timing task, 0.5 ul of saline or D1 dopamine blocker SCH23390 is infused in the LCN prior to the start of the experiment. Interval timing performance is evaluated based on the efficiency of response times, which is calculated based on the number of lever presses that occur during the 11-12 second interval versus the total number of responses. We use two data driven approaches to quantify ramping activity – linear regression and principal component analysis (PCA).
Results: Animals with LCN D1DRs blocked are significantly impaired on the interval timing task and estimate the passage of the interval earlier than following saline infusion. Additionally, following saline infusions in the LCN, the first principle component (PC1), which explains the most variability in the dataset, was a characteristic ramping activity in single MFC neurons. The amount of variability in MFC neurons as explained by PC1 was significantly decreased following LCN D1DR blockade. This result was also supported by a significant decrease in the number neurons fit by linear regression.
Conclusions: LCN D1DR-expressing neurons modulate single neurons in the MFC and are essential for interval timing performance. These data provide insight into how the cerebellum influences medial frontal networks and indicate that D1DRs may play an essential role in cognitive processing.
Keywords: Cerebellum, D1 Dopamine Receptors, Schizophrenia, Cognition, Timing
Disclosure: Nothing to Disclose.

T35. Convergent Neurobiological Correlates of Schizophrenia-Associated Genes in Zebrafish

Summer Thyme*, Lindsey Pieper, Eric Li, Yiqun Wang, Carrie Sha, Edward Soucy, Shristi Pandey, Joo Won Choi, Owen Randlett, Steve Zimmerman, Steve McCarroll, Alexander Schier

Harvard University, Cambridge, Massachusetts, United States

Background: Large-scale genome-wide association studies have begun to uncover numerous candidate genes linked to schizophrenia. Yet it remains unclear how these genes function and how they contribute to the underlying molecular, cellular, developmental and behavioral processes disrupted in the disorder.
Methods: Recent technological breakthroughs in zebrafish – targeted genome editing with CRISPR/Cas9, whole-brain activity imaging, brain atlas registration, behavioral profiling – combined with the ease of studying large numbers of animals make it an ideal system for analyzing psychiatric disease genes. Combining these technologies, I have generated zebrafish mutants for over 100 schizophrenia-associated genes and am analyzing them for differences in neurological activity and morphology, as well as altered behavior. To assay these mutants for functionally altered brains, I am using a high-throughput antibody staining technique that reports integrated neuronal activity in freely swimming larvae. To determine whether these mutants have altered behavior, I am characterizing their movement rates during the day and night (sleep) and in a stressful heat condition, startle responses to both light changes and sounds, habituation to a stimulus (primitive form of learning), and level of pre-pulse inhibition.
Results: Over one third of these zebrafish mutants display phenotypes in brain activity, brain morphology, or behavior. Comparing brain activity maps between mutants, I have discovered that multiple schizophrenia-associated genes alter activity in the same brain regions. Such shared phenotypes can clarify ambiguity at multi-gene loci: associated genomic

regions containing multiple genes only one of which is likely involved in disease pathology. Observed brain abnormalities in mutants resemble known schizophrenia patient phenotypes, such as the loss of GABAergic inhibitory neurons. Behavioral abnormalities observed in mutant animals are also reminiscent of motor behaviors observed in patients. I have identified mutants with reduced prepulse inhibition, a hallmark of schizophrenia pathology, including for several genes that not previously been linked to this behavior. These findings strengthen the connection between these genes and the disorder, and can also clarify ambiguity at multigene loci.

Conclusions: The finding of shared phenotypes suggests that seemingly unrelated schizophrenia-associated genes may be involved in common pathways. The identification of several genes responsible for maintaining prepulse inhibition directly links these genes to schizophrenia and enables further characterization of the molecular mechanisms underlying this relevant behavior. Our work illustrates how studies in a simple animal model nervous system can help uncover these pathways and elucidate gene function. Understanding the molecular, cellular, developmental and behavioral processes regulated by schizophrenia-associated genes will provide the foundation to understand the causes of schizophrenia and develop new diagnostics and therapies.

Keywords: GWAS LOCI, Zebrafish, Schizophrenia, Animal Models, Prepulse Inhibition

Disclosure: Nothing to Disclose.

T36. Role of ErbB4 Splicing in Parvalbumin Interneuron Maturation and Schizophrenia

Daniel Chung*, Kenneth Fish, David Volk, Zachary Wills, David Lewis

University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania, United States

Background: Proper working memory performance depends on the efficient excitatory drive to parvalbumin-containing (PV) GABAergic interneurons in the dorsolateral prefrontal cortex (DLPFC). In adolescence, synaptic pruning eliminates unwanted or imprecise excitatory inputs, suggesting that working memory maturation during this period may require the pruning of excitatory inputs to PV neurons. In schizophrenia, excitatory drive to PV neurons is thought to be reduced, suggesting that working memory impairment in this illness may be due to the loss of excitatory inputs to PV neurons. The formation of excitatory synapses on PV neurons is mediated by receptor tyrosine kinase ErbB4. ErbB4 transcript is alternatively spliced and each splice variant is associated with different functional effects. In schizophrenia, total ErbB4 expression is unaltered, but splicing of ErbB4 is abnormally shifted from major to minor variants. Thus, ErbB4 splicing may provide a molecular mechanism for modulating the excitatory synapse number on PV neurons.

In this study, we tested the hypotheses that 1) the pruning of excitatory synapses on PV neurons occurs in the DLPFC during adolescence and is associated with a developmental shift in ErbB4 splicing, 2) the loss of excitatory synapses on PV neurons occurs in schizophrenia and is associated with a pathogenic shift in ErbB4 splicing and 3) ErbB4 major and minor variants have distinct tyrosine kinase activity and differentially regulate excitatory synapse formation in PV neurons.

Methods: To investigate PV neurons during adolescence, DLPFC sections of rhesus monkeys from pre-pubertal age ($n=7$) and post-pubertal age ($n=6$) were used. To investigate PV neurons in schizophrenia, DLPFC sections of schizophrenia subjects and matched unaffected comparison subjects ($n=20$ pairs) were used. To assess excitatory synapse on PV neurons, the density of VGlut1+/PSD95+ puncta on PV+ neurons was measured. To assess ErbB4 splicing in PV neurons, the transcript levels of PV, ErbB4 major and minor variants and pan-ErbB4 were quantified. To assess the functional effect of ErbB4 splice variants, the major and minor variants were overexpressed and the density of VGlut1+/PSD95+ puncta on PV+ neurons in rat primary neuronal culture or tyrosine kinase activity in HEK293 cells was measured.

Results: In monkey DLPFC, the density of VGlut1+/PSD95+ puncta on PV+ neurons was lower in post-pubertal relative to pre-pubertal group, demonstrating pruning of excitatory synapses on PV neurons during adolescence. The puncta levels of VGlut1 and PSD95 proteins were higher in post-pubertal group and positively predicted activity-dependent PV levels across all monkeys, suggesting a greater strength of the remaining synapses after pruning. Finally, higher minor-to-major variant ratios predicted lower PSD95+ puncta density on PV+ neurons across all monkeys, suggesting ErbB4 splicing regulates synaptic pruning in PV neurons.

In human DLPFC, the density of VGlut1+/PSD95+ puncta on PV+ neurons was lower in schizophrenia relative to comparison subjects, whereas the puncta levels of VGlut1 and PSD95 proteins were not altered, demonstrating the loss of excitatory synapses on PV neurons without changes in the remaining synapse in schizophrenia. Moreover, the density of VGlut1+/PSD95+ puncta on PV+ neurons positively predicted PV levels selectively across schizophrenia subjects, suggesting the loss of excitatory synapse results in reduced PV neuron activity in the illness. Finally, higher minor-to-major variant ratios predicted lower VGlut1+/PSD95+ puncta density on PV+ neurons selectively across schizophrenia subjects, implying that the loss of excitatory synapses on PV neurons in the illness may be due to an excessive pruning process.

In cell culture, the major variant, but not the minor variant, increased the density of VGlut1+/PSD95+ puncta on PV+ neurons; the major variant also displayed greater kinase activity than the minor variant, suggesting that shift in ErbB4 splicing results in lower ErbB4 signaling, which in turn contributes to fewer excitatory synapse on PV neurons.

Conclusions: Working memory dysfunction in schizophrenia is proposed to arise from alterations in prefrontal cortical PV neurons during development. However, the pathological basis, developmental origin and pathogenic mechanism of PV neuron dysfunction in this illness are not well understood. Using a top-down translational approach across different species and experimental conditions, our study 1) identifies fewer excitatory synapses on PV neurons as a novel synaptic pathology of schizophrenia, 2) implicates an exaggerated pruning of excitatory synapses on PV neurons during adolescence as a developmental origin of this

pathology and 3) demonstrates shifts in alternative splicing of ErbB4 transcripts as a molecular mechanism that can modulate the excitatory synapse number on PV neurons in development and disease. Thus, the results from our study collectively reinforce the view that schizophrenia is a neurodevelopmental disorder with disturbances in the normal maturation process of prefrontal inhibitory circuits.
Keywords: Schizophrenia, Synaptic Pruning, Parvalbumin Interneuron, Alternative Splicing
Disclosure: Nothing to Disclose.

T37. Investigating White Matter Maturation in Genetic High Risk for Schizophrenia Populations

Amanda Lyall*, Nathaniel Somes, Fan Zhang, James Robertson, Lauren O'Donnell, Yogesh Rathi, Ofer Pasternak, Zachary Fitzgerald, Peter Savadjiev, Elena Molokotos, Raquel Mesholam-Gately, Joanne Wojcik, Ben Brent, Heidi Thermenos, Susan Whitfield-Gabrieli, John Gabrieli, Matcheri Keshavan, Lynn Delisi, Larry Seidman, Marek Kubicki

Harvard Medical School, Boston, Massachusetts, United States

Background: Schizophrenia (SZ) has been recognized as a neurodevelopmental disorder with language and working memory deficits. Previous neuroimaging studies of SZ patients report decreased fractional anisotropy in anatomical white matter tracts serving these cognitive functions. However, the timeline of white matter (WM) abnormalities and their relationship to the development of cognitive deficits is poorly understood. This study aims to utilize diffusion imaging and neurocognitive assessments to investigate the relationship between potential structural alterations in WM tracts associated with language and working memory scores in two independent populations of individuals at genetic high risk (GHR) for SZ compared to matched controls (HC).
Methods: 3T diffusion-weighted images of children aged 7 to 12 (24 HC and 16 at GHR for SZ) and young adults aged 19 to 29 (26 HC and 43 GHR) were collected at the Massachusetts Institute of Technology. Whole brain two-tensor tractography was performed and two bilateral WM tracts of interest, a language tract (the arcuate fasciculus (AF)), and a working memory tract (the cingulum bundle (CB)), were extracted utilizing an unsupervised fiber clustering algorithm. The fractional anisotropy of the tissue (FA-t), a novel more biologically-specific measure of WM tissue microstructure, was obtained. We carried out group comparisons of FA-t between GHR and HCs utilizing Mann-Whitney-U tests and Cohen's d effect sizes for each WM tract of interest. Spearman correlations between performance on the language-specific and working memory-specific task were completed with the FA-t values in the AF and CB, respectively, for each population.
Results: Preliminary analyses reveal no significant differences in FAt between GHR and HC in the bilateral AF or CB for either the child or adult population. However, the decreased FAt values observed in the bilateral CB in the GHR children did exhibit medium to large effect sizes (CB-left, $d = 0.51$; CB-right, $d = 0.79$). GHR children also exhibit significant reductions in performance on the working memory-specific task (WISC Letter Number Sequencing, $p = 0.013$), though this was not significantly correlated with FAt in the CB-left or CB-right. GHR children show no differences for the language-specific task. GHR adults show no significant differences for either the language or working memory-specific tasks, nor was FAt in the AF or CB correlated with their respective cognitive tasks.
Conclusions: The preliminary results of this analysis suggest that GHR children and adults do not show significant differences in a maturational measure of WM in the cognitive domain-specific tracts chosen for this study when compared to HCs. However, the significant difference between groups in the child GHR population for the working-memory specific task indicates that a cognitive phenotype may exist. This finding, paired with the Cohen's d in the bilateral CB, could indicate that greater structural differences in the CB in GHR subjects may potentially develop later in the maturational trajectory. The lack of structural or cognitive differences in the adult GHR population, however, may indicate that young adults with an increased genetic risk for the disorder that did not convert to psychosis may exhibit resiliency. These findings are akin to the study by Gogtay et al (2007), which showed amelioration of regional gray matter deficits and functional outcomes occurring by 20 years of age in the healthy siblings of patients with childhood-onset SZ. Taken together, the results of this study provide possible evidence that subtle divergences from a healthy WM maturational trajectory may occur in early adolescence. Additionally, this study may help isolate early-to-late adolescence as a critical period of increased vulnerability.
Keywords: Diffusion MRI, Familial Risk, Cognitive Phenotypes
Disclosure: Nothing to Disclose.

T38. Explication of FKBP5 Methylation and Transcription Patterns in Postmortem Human Cortex

Natalie Matosin*, Silvia Martinelli, Yvonne Awaloff, Tobias Wiechmann, Andrea Schmitt, Thomas Arzberger, Elisabeth Binder

Max Planck Institute of Psychiatry, Munich, Germany

Background: FKBP51, encoded by the FKBP5 gene, is a key co-chaperone of the glucocorticoid receptor in response to stress that is linked to the development of severe psychiatric disorders. The FKBP5 gene codes four indexed alternative transcripts, and data from our laboratory suggests FKBP5 transcription might be regulated by DNA demethylation at key FKBP5 enhancers. It is likely that transcriptional and epigenetic regulation of the FKBP5 gene is tissue and cell-type specific. These patterns of regulation, that may be relevant for understanding disease risk, are yet to be determined in the human brain.
Methods: Postmortem brain samples from BA9 of 24 individuals without a history of neurological or psychiatric conditions were assessed. Ages of the subjects were evenly distributed from 37 to 88 years. Fresh frozen tissues were cryosectioned at 16μm and post-fixed in 4% PFA. For immunohistochemistry (IHC), samples were incubated with

antibodies specific for a neuronal marker (TUJIII/MAP2), glia marker (GFAP) and full-length FKBP51 protein. For in situ hybridization (ISH), the RNAscope Multiplex kit was used to amplify probes targeting the FKBP5 long and short variants. Both IHC and ISH experiments were imaged with a Leica SP8 confocal microscope. An additional subset from BA18/19 of three control subjects were additionally FACS sorted into neuronal and non-neuronal subtypes with NeuN/DRAQ5 antibodies. DNA was extracted from NeuN+/- nuclei or bulk tissue, bisulfite converted and DNA methylation was assessed using a targeted bisulfite sequencing approach for the FKBP5 locus and the Infinium MethylationEPIC BeadChips.

Results: Our IHC data suggest that FKBP51 total protein expression might be limited to neuronal subtypes, although these data are undergoing validation. Similarly, ISH experiments show that FKBP5 long and short proteins are both expressed in BA9, with distributions in both nuclear and cytoplasmic compartments. These data are now being assessed for differences in levels and distributions of FKBP5 protein and transcripts in association with age. Total DNA methylation at FKBP5 regulatory CpGs (i.e. in promotor or enhancer regions) were significantly higher in neuronal populations compared to non-neuronal populations ($P = 0.011$) and compared to bulk tissue ($P = 0.002$). When assessing methylation at individual FKBP5 CpGs of interest, large differences in methylation were observed in CpGs close to regulatory regions of the FKBP5 gene, such as CTCF binding sites (e.g. cg06087101) and transcription start sites (e.g. cg01294490). These data are being confirmed in the larger BA9 cohort and with targeted bisulfite sequencing. Cell-type specific effects of methylation on transcription will also be assessed using qPCR.

Conclusions: These data provide initial evidence that the FKBP5 methylome and transcriptome are regulated in a cell-type specific manner in the human cortex. Further analyses are now being conducted to validate cell-type specific RNA and protein expression of FKBP5, and to determine associations of FKBP5 methylation and transcription. In addition, these results will be integrated with evidence from other brain cohorts to validate and determine if case-control differences exist in psychiatric disorders.

Keywords: Postmortem Brain Tissue, FKBP5, Gene Regulation, Epigenetics, Early-Life Stress

Disclosure: Nothing to Disclose.

T39. The Longitudinal Trajectory of Serum Brain-Derived Neurotrophic Factor (BDNF) Levels in Psychotic Patients: The Impact of Clinical and Genetic Variables

Mirko Manchia*, Diego Primavera, Luca Deriu, Edoardo Caboni, Novella Iaselli, Davide Sundas, Massimo Tusconi, Donatella Congiu, Roberto Collu, Alessio Squassina, Maria Scherma, Paola Fadda, Walter Fratta, Bernardo Carpiniello

University of Cagliari, Cagliari, Italy

Background: There is consistent evidence indicating a decline of serum brain-derived neurotrophic factor (BDNF) levels in chronic and medicated schizophrenic (SCZ) patients, as well as in first episode and medication naïve SCZ patients, compared to unaffected subjects. The decrease of peripheral BDNF levels could be constant, with pre-morbid levels roughly similar to those found in unaffected individuals, linearly declining during the course of SCZ. Alternatively, BDNF peripheral levels might fluctuate in association with acute psychopathological phases of the disorder. In addition, pharmacological treatment, such as antipsychotics, might influence BDNF levels. In this context, we aimed to test 1) whether longitudinal changes in BDNF serum levels were associated with psychopathological, cognitive, and treatment variables, and 2) whether genetic variation within BDNF gene has a role in modulating the longitudinal trajectory of serum BDNF levels. To this end, we followed-up a cohort of SCZ and schizoaffective disorder (SAD) patients over a period of 24 months, measuring serum BDNF levels, as well as clinical and treatment variables, every 6 months.

Methods: Our sample population encompassed a cohort of 105 SCZ and SAD patients followed at the community mental health center of the Psychiatry Research Unit, University of Cagliari. To be included in the study patients had to have 1) an age comprised between 18 and 65 years, 2) a diagnosis of SCZ or SAD according to Diagnostic and Statistical Manual of Mental Disorders-IV-Text Revision (DSM-IV-TR) criteria, and 3) stability of symptoms during the six months before recruitment. Psychopathological measures included the Positive and Negative Symptom Scale for Schizophrenia (PANSS), and the Clinical Global Impression Scale for Schizophrenia (CGI-SCH), while cognitive function was assessed with the Brief Assessment of Cognition in Schizophrenia (BACS) scale. BDNF serum levels were evaluated using human BDNF enzyme-linked immunosorbent assay (ELISA) Kit. High molecular weight genomic DNA was extracted from peripheral blood leukocytes by NaCl precipitation and genotyping of rs1519480 of the BDNF gene was performed using the TaqMan 5'exonuclease method. Mixed-effects linear regression models (MLRM) were used to analyze longitudinal data. In all analyses, we regressed independent variables (both categorical and continuous) on BDNF serum levels (dependent variable). These analyses were performed with packages "lme4" and "multcomp" implemented in R.

Results: Seventy-four patients (70.5%) out of 105 were male. The MLRM analysis correcting for sex and age showed a statistically significant decrease of BDNF levels over time ($Z = -4.09$, $p = 7.4$ x 10-7). Concerning psychopathological measures, the severity of negative and depressive symptoms assessed with the CGI-SCH was associated with a longitudinal decrease of serum BDNF levels, correcting for age and sex ($Z = -2.1$, $p = 0.04$ and $Z = -2.8$, $p = 0.004$). This result remained significant even when we corrected for presence of oral or depot/long-acting injectable (LAI) antipsychotic treatment ($Z = -2.65$, $p = 0.008$). In addition, higher BDNF serum levels were associated with higher scores in verbal fluency (controlled oral word association test) of the BACS ($Z = 3.1$, $p = 0.002$). Again, this result remained significant after correcting for presence of oral or depot/long-acting injectable (LAI) antipsychotic treatment ($Z = 2.6$, $p = 0.01$). There was no effect of the rs1519480 of the BDNF gene on the longitudinal trajectory of serum BDNF ($Z = -1.1$, $p = 0.26$).

Conclusions: In our prospective study, we found a general decline of BDNF serum levels over 24-month. Further, the

longitudinal trajectory of BDNF serum levels decreased over time in relation to the severity of depressive symptoms and increased with higher scores of cognitive performance. Although awaiting replication in independent studies, these results point to distinct trends in the longitudinal variation of BDNF levels, which may help setting up predictive clinical models in major psychoses. The role of genetic polymorphism will be further investigated with the genotyping of the tag SNPs covering the BDNF gene.

Keywords: Longitudinal Study, Schizophrenia, BDNF, Genetics, Cognition

Disclosure: Nothing to Disclose.

T40. Auditory-Based Targeted Cognitive Training Improves Verbal Learning and Reduces Auditory Hallucinations in Chronic Schizophrenia Patients in Locked Residential Care

Yash Joshi*, Michael Thomas, Andrew Bismark, Emily Treichler, Melissa Tarasenko, Lauren Cardoso, Juan Molina, Kristine Tiernan, Neal Swerdlow, Gregory Light

University of California, San Diego School of Medicine, La Jolla, California, United States

Background: Targeted cognitive training (TCT) is an emerging computerized intervention for remediating neurocognitive deficits in patients with schizophrenia (SZ). TCT is designed to enhance "bottom-up" information processing through adaptive and intensive exercises that target low-level auditory discrimination in SZ. Previous studies of TCT have shown that TCT improves cognitive functioning, particularly in the domain of verbal learning and memory. However, these studies were conducted largely in relatively high functioning, younger patients recruited in academic outpatient settings. The effectiveness of TCT for severely disabled SZ patients in the community is not yet established. Here we report preliminary findings from an ongoing randomized clinical trial investigating the effectiveness of TCT in SZ patients in a locked transitional care facility specializing in long-term rehabilitation after acute psychiatric hospitalization.

Methods: Patients with SZ were recruited from a locked residential care program with an average length of stay of 182 days and randomized to treatment as usual (TAU; $n = 24$, mean age = 34.54y) or treatment as usual + TCT ($n = 22$, mean age = 35.73y). TCT and TAU groups did not significantly differ in demographic, clinical, or cognitive variables at baseline. Average duration of illness was approximately 15 years, and did not differ between TCT and TAU groups (TAU = 15.23y; TCT = 16.12y, NS). Auditory discriminability (Word-In-Noise test, WIN), cognitive functioning (MATRICS Consensus Cognitive Battery, MCCB), as well as negative and positive symptoms scores (Scale for the Assessment of Negative Symptoms, SANS; Scale for the Assessment of Positive Symptoms, SAPS) were assessed before and after a full course of TCT. Data were analyzed using linear mixed effects models.

Results: At follow up, TCT was associated with significant improvements in auditory discriminability (WIN d = 0.63, $p < 0.04$), verbal learning and memory (d = 0.82 $p < 0.01$), and positive symptoms (SAPS d = -0.62, $p < 0.05$). Negative symptoms (SANS) did not significantly differ between TCT and TAU groups. Follow-up analyses of SAPS global ratings revealed that improvements in positive symptoms were driven by a significant reduction in auditory hallucinations (d = -0.90, $p < 0.01$). No differences in chlorpromazine equivalents (CPZ) were noted in either group at baseline or at follow up (TAU, pre/post CPZ = 984 mg / 988 mg; TCT pre/post CPZ = 1253 mg / 994 mg, d = - 0.20, NS). Age was a significant moderator of verbal learning gains with TCT; older patients exhibited greater improvements ($p < 0.01$).

Conclusions: TCT significantly improved signal-to-noise discrimination of speech sounds, enhanced verbal learning and memory, and reduced the severity of auditory hallucinations in SZ patients in locked residential care. In contrast to the widespread belief that TCT is most effective in younger, early-illness SZ patients, preliminary results indicate that older patients may also benefit from TCT. This pattern of results demonstrates that improving the fidelity of low-level auditory sensory information processing yields clinically meaningful outcomes, even in patients with chronic illness. Ongoing studies will examine whether TCT-associated improvements can be predicted by neurophysiologic biomarkers and other demographic, clinical and cognitive characteristics assessed at baseline.

Keywords: Computerized Cognitive Training, Clinical Trial, Auditory Processing

Disclosure: Nothing to Disclose.

T41. Pretreatment Predictors and Moderators of Antipsychotic-Related Weight Gain in the Treatment of Early-Onset Schizophrenia Spectrum Disorders (TEOSS) Study

Jerome Taylor*, Ewgeni Jakubovski, Daniel Gabriel, Michael Bloch

University of Pennsylvania, Philadelphia, Pennsylvania, United States

Background: We sought to identify predictors and moderators of antipsychotic-related weight gain in the Treatment of Early-Onset Schizophrenia Spectrum Disorders (TEOSS) study.

Methods: TEOSS randomized 119 youths (age 8–19 years) with schizophrenia or schizoaffective disorder to 8 weeks of antipsychotic treatment with molindone, risperidone or olanzapine and assessed treatment response and weight change. We used multivariable linear regression and receiver operating characteristics analysis to identify predictors and moderators of change in body mass index Z-score (BMIZ) and weight (measured as percent change in weight) from baseline to week 8.

Results: Treatment assignment predicted BMIZ change (DF = 2, F = 14.33, $p < 0.001$) and weight change (DF = 2, F = 15.66, $p < 0.001$): mean BMIZ change was -0.07 (95CI: -0.20–0.06) for molindone, 0.26 (95CI: 0.12–0.40) for risperidone and 0.55 (95CI: 0.33–0.78) for olanzapine; mean percent weight increase was 0.71% (95CI: -1.63%–3.05%) for molindone, 6.70% (95CI: 4.01%–9.40%) for risperidone and 12.54% (95CI: 8.81%–16.27%) for olanzapine. Pretreatment BMIZ and fasting low density lipoprotein (LDL) cholesterol

significantly moderated changes in BMIZ (DF = 2, F = 9.95, p-interaction < 0.001; DF = 2, F = 3.45, p-interaction = 0.04) and weight (DF = 2, F = 4.61, p-interaction = 0.01; DF = 2, F = 4.65, p-interaction = 0.02), such that lower pretreatment BMIZ and LDL predicted larger increases in BMIZ and weight for youths randomized to risperidone or olanzapine, but not molindone.
Conclusions: BMIZ increase and weight gain were severe for olanzapine and moderate for risperidone, while molindone was weight-neutral. When treated with olanzapine or risperidone, youths with markers of good health (lower BMIZ and LDL) pretreatment had the largest increases in BMIZ and gained the most weight. ClinicalTrials.gov Identifier: NCT00053703.
Keywords: Schizophrenia, Antipsychotics, Children, Side Effects, Clinical Trial
Disclosure: Nothing to Disclose.

T42. Hallucinations, Short-Term Neuroplasticity, and Prediction Error Signaling in Schizophrenia

Amanda McCleery*, Jonathan Wynn, Michael Green

University of California, Los Angeles Semel Institute for Neuroscience and Human Behavior, Los Angeles, California, United States

Background: Auditory hallucinations, a hallmark symptom of psychosis, are experienced by most people with a diagnosis of schizophrenia at some point in their illness. Auditory hallucinations can be understood as a failure in predictive coding, whereby abnormalities in sensory/perceptual processing combine with biased cognitive processes to result in a dampening of normal prediction error signaling. In this study, we used a roving auditory mismatch negativity (MMN) paradigm to optimize evaluation of prediction error signaling and short-term neuroplasticity in people with schizophrenia with and without recent hallucinations and healthy comparison participants. In the roving MMN paradigm, a series of standards is presented, followed by a deviant, as in a typical MMN. However, the deviant tone is then presented repeatedly, becoming the new standard. This new standard sequence is subsequently interrupted by a new deviant, and the process repeats. The number of standard presentations in the sequence is varied throughout the task. As the number of standards in a row increases, the amplitude of the ERP to the standard stimulus becomes more positive (called "repetition positivity"). Repetition positivity is hypothesized to reflect stimulus-specific adaptation of neuronal activity to the standard stimulus, a form of short-term neuroplasticity. Likewise, response to the deviant stimulus and the resultant MMN becomes increasingly negative as the number of standards preceding it increases. This increase in MMN amplitude reflects the discrepancy between the expected tone and the actual tone, that is, the prediction error. The increase in MMN with increasing number of standard tones is referred to as the memory trace effect.
Methods: MMN amplitude, prediction error signaling (i.e., memory trace effect), and short-term neuroplasticity (i.e., repetition positivity) were compared in 30 people with schizophrenia ($n = 16$ with and $n = 14$ without recent auditory hallucinations) and 20 healthy comparison participants. The first series of analyses compared the entire schizophrenia group to the healthy comparison participants. The second series of analyses compared the schizophrenia participants with recent hallucinations to those without recent hallucinations.
Results: There were no statistically significant differences between the healthy comparison group and the combined schizophrenia group on any of the roving MMN indices. Compared to the schizophrenia group without hallucinations, the recent hallucinations group exhibited an abnormal roving MMN profile [$F(2,27) = 3.98$, $p = 0.03$], significantly reduced prediction error signaling [$t(28) = -2.25$, $p = 0.03$], and a trend for diminished short-term neuroplasticity [$t(28) = 1.80$, $p = 0.08$].
Conclusions: These findings are consistent with a predictive coding account of hallucinations in schizophrenia, which posits reduced prediction error signaling in those who are prone to hallucinations. These results also suggest that plasticity-mediated formation and online updating of predictive coding models may also be disrupted in individuals with recent hallucinations.
Keywords: Event-Related Potentials, Mismatch Negativity, Auditory Hallucinations, Schizophrenia, Prediction Error
Disclosure: Part 1: MedAvante, Employee.

T43. Long-Lasting Impact of Adolescent Cannabinoid Exposure on Reward-Related Behaviors: Potential Interaction With Schizophrenia

Jibran Khokhar*, Travis Todd, Wilder Doucette, David Bucci, Alan Green

Dartmouth College, Lebanon, New Hampshire, United States

Background: Adolescent cannabis use occurs commonly, affects neurodevelopment, and is a risk factor for schizophrenia, as well as future substance use. Schizophrenia, itself, is associated with high rates of alcohol and drug use as well as motivational and reward-learning dysfunctions. Thus, we were interested in whether Δ9-tetrahydrocannabinol (THC) exposure during adolescence would influence reward related behaviors in adulthood, especially in the context of neurodevelopmental risk for schizophrenia.
Methods: All animal studies were approved by Dartmouth College's Institutional Animal Care and Use Committee and were conducted in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals. Using a neurodevelopmental model of schizophrenia, the neonatal ventral hippocampal lesioned (NVHL) rat, we assessed the effects of adolescent THC (or vehicle) treatments (post-natal day 28-42; 6 mg/kg i.p.) on: (a) free-access 2-bottle choice alcohol (20% v/v) drinking; (b) context-based instrumental food reward acquisition, extinction, renewal and reinstatement; and (c) limited access sweet-fat food binge-like eating ($n = 5$/group). In a subsequent study, we assessed the effects of adolescent THC (or vehicle) treatment on the acquisition and extinction of Pavlovian autoshaping (sign-tracking) behavior ($n = 15$-16/group).
Results: Neither NVHL-lesion status nor adolescent THC treatment altered free-access alcohol intake in adulthood.

Adolescent THC treatment, however, significantly impaired the motivation to lever press for a food reward in both the NVHL and sham animals; both THC treated groups showed decreased responding throughout the entire acquisition period, consistent with decreased motivation to work for a reward in adolescent cannabis smokers. In contrast, only the THC-treated sham animals showed reduced food cup entries, while extinction or renewal of lever pressing did not differ between groups. Conversely, the NVHL animals displayed impaired reinstatement of lever-pressing if given food pellets in the extinction context. Lastly, THC-treated NVHL and sham animals displayed decreased binge-like sweet-fat food consumption in a limited-access paradigm. In the autoshaping study, adolescent THC treatment significantly increased sign-tracking compared to vehicle treatment (consistent with increased cue-reactivity in adolescent cannabis smokers).

Conclusions: This study suggests the adolescent THC exposure may produce long-term changes in reward-related behaviors, independent of risk for schizophrenia. The discordant findings between instrumental and Pavlovian conditioning may also provide important clues regarding the neurobiological and behavioral underpinnings of the potential "gateway drug" effects of adolescent cannabis use as it relates to the risk for future substance use.

Keywords: delta9-tetrahydrocannabinol, Adolescent Development, Schizophrenia-Like Behavior, Drug Addiction, Cannabis Use

Disclosure: Nothing to Disclose.

T44. Nicotine Reverses Hypofrontality in Animal Models of Addiction and Schizophrenia Through a Hierarchical Inhibitory Circuit

Fani Koukouli*, Uwe Maskos

Institute Pasteur Paris, Paris, France

Background: Genome-wide association studies (GWAS) identified genetic variants within the CHRNA3–CHRNA5–CHRNB4 nAChR gene cluster that increase the risk of habitual smoking. Among the series of polymorphisms composing this human haplotype, the rs16969968 single-nucleotide polymorphism (SNP) leads to the substitution of aspartic acid at residue 398 by asparagine (D398N) in the human α5 subunit. Recently, the same genetic locus was associated with schizophrenia in a major GWAS. Because genetic deletion of the α5 nicotinic acetylcholine receptor (nAChR) subunit in mice leads to prefrontal cortex (PFC) linked behavioral deficits and altered cholinergic excitation, we developed a mouse line expressing the human α5 variant (α5SNP mice) to investigate the impact of this variant on behaviors associated with schizophrenia. We then aimed to determine the circuitry mechanisms underlying cognitive impairments in α5SNP mice and understand a possible causal relationship between nicotine addiction and schizophrenia.

Methods: Mice engineered to harbor the α5 D398N variant were obtained via homologous recombination. Wild-type and α5SNP mice were tested for deficits in two behavioural paradigms: the three-chamber social interaction test and the prepulse inhibition (PPI) task. Using in vivo two-photon calcium imaging, CRISPR/Cas9 technology and pharmacological interventions in the PFC of awake mice, we determined the circuit mechanisms underlying the cognitive deficits attributed to dysfunction of α5-containing nAChRs and the human SNP.

Results: We showed that mice expressing a human α5SNP exhibit neurocognitive behavioral deficits in social interaction and sensorimotor gating tasks and different nAChR subunits control spontaneous PFC activity through a hierarchical inhibitory circuit. Specifically, in mice expressing the human α5SNP associated with nicotine addiction and schizophrenia and α5 knockout (KO) mice, lower activity of vasoactive intestinal polypeptide (VIP) interneurons resulted in an increased somatostatin (SOM) interneuron inhibitory drive over pyramidal neurons in layer II/III of PFC. Chronic nicotine administration reversed the hypofrontality observed in α5SNP mice through desensitization of β2 subunits in SOM interneurons. We also present unpublished data showing that α5 nAChR subunits play a key role in the generation of ultra-slow fluctuations (USFs) in the PFC and are specifically required for synchronized activity patterns. These USFs are similar to activity described in the human brain, which is linked to cognitive processing. Finally, we show original data showing impaired pyramidal cell activity in deep layers of PFC in α5SNP mice.

Conclusions: Endogenous ACh distinctly recruits specific interneuron types within an interneuron hierarchy that controls pyramidal neurons in PFC, through differential expression of nAChR subunits. Chronic nicotine treatment of mice carrying the α5SNP restores their neuronal activity deficits. This work paves the way towards new therapeutic strategies targeting nicotinic receptors in mental disorders.

Keywords: Prefrontal Cortex, Nicotinic Acetylcholine Receptors, Two-photon, Nicotine Addiction, Schizophrenia

Disclosure: Nothing to Disclose.

T45. Quantifying Dopamine Receptor Availability in the Retina of Humans Using [11C]-(+)-PHNO

Fernando Caravaggio*, Jun Ku Chung, Philip Gerretsen, Ariel Graff-Guerrero

Centre for Addiction and Mental Health, University of Toronto, Vaughan, Canada

Background: [11C]-(+)-PHNO is an agonist radiotracer for dopamine (DA) D2/3 receptors (D2/3R). We have observed significant uptake of [11C]-(+)-PHNO into the retina, a region rich in extrasynaptic DA receptors. We wished to determine whether this uptake in the retina could be quantified with two reference tissue methods: the simplified reference tissue model (SRTM) and Ichise's Multilinear Reference Tissue Model (MRTM). We then explored whether [11C]-(+)-PHNO BPND in the retina differed between neuroleptic naïve patients with schizophrenia and age and gender matched healthy controls.

Methods: [11C]-(+)-PHNO data from 49 healthy controls (μage: 39.96 ± 14.36; 16 female) and 12 antipsychotic-naïve patients with schizophrenia (μage: 25.75 ± 6.25; 4 female) were analyzed. Time activity curves were extracted from the

retina and cerebellum from manually drawn ROIs (author FC).
Results: The average measure interclass correlation coefficient (ICC) between the manually drawn SRTM and MRTM retina BPND estimates was .91, with a 95% CI from .84 and .95 ($F(48,48) = 11.70$, $p < .0001$). Retina BPND did not differ between patients and matched controls measured with the SRTM ($W = 2$, $p = .97$) and MRTM ($W = 16$, $p = .57$).
Conclusions: It is possible to quantify DA receptor availability in the retina with [11C]-(+)-PHNO. Notably, BPND's and associated model fits fell within previously accepted ranges for other ROIs, such as the thalamus (average BPND, 95% CI): .36, .26-.491; .44, .35-.542. Future studies should conduct displacement studies to determine whether D3R is expressed in the human retina. While it is possible to quantify retina BPND using reference tissue models, arterial kinetic modelling is warranted.
Keywords: PET Imaging, Antipsychotic-Naïve First-Episode Schizophrenia, Retina
Disclosure: Nothing to Disclose.

T46. Adjunctive Treatment With Pentosan Polysulfate Sodium in Patients With Schizophrenia

Olaoluwa Okusaga*

University of Texas Health Science Center at Houston, Houston, Texas, United States

Background: Schizophrenia is associated with immune system dysfunction including abnormal cytokine levels. Elevated plasma cytokines and C-reactive protein have been negatively correlated with cognitive and psychotic symptoms in patients with schizophrenia. Therefore, if peripheral inflammation influences brain function in schizophrenia, it stands to reason that pathways of communication most likely exist between the periphery and the brain. Manipulation of such communication pathways could potentially be of therapeutic value in patients. One such communication pathway that has been poorly studied involves immune-mediated brain endothelial activation which includes the expression of P-selectin (a cell adhesion molecule). Plasma soluble P-selectin levels have been shown to be elevated in patients with schizophrenia. This study is an open-label add-on treatment with pentosan polysulfate sodium (PPS), a heparin-like molecule with potent P-selectin blocking property.
Methods: After IRB approval, two patients with schizophrenia (diagnosed using SCID-5) were recruited to receive PPS 100mg by mouth three times per day in addition to their antipsychotic medication. The first patient (19-year-old Black lady) received PPS for 2 weeks and the second patient (26-year-old Black man) received PPS for 8 weeks. We evaluated PPS blockade of endothelial P-selectin by measuring plasma soluble P-selectin at baseline, week 2, 4, 6 and 8. PPS effect on neurotrophic factors was evaluated by measuring plasma brain derived neutrophic factor (BDNF) at baseline, week 2, 4, 6 and 8. Plasma soluble P-selectin and BDNF were measured by ELISA.
Results: In the first patient, plasma soluble P-selectin decreased from baseline levels after 2 weeks treatment with PPS (123.8 ng/ml and 97.2 ng/ml respectively) while BDNF levels increased (3742.9 pg/ml and 4957.1 pg/ml respectively). There was a less consistent pattern in the second patient; plasma soluble P-selectin at baseline, week 2, 4, 6 and 8 were 59.5 ng/ml, 59.3 ng/ml, 69.3 ng/ml, 63.3 ng/ml, and 54.1 ng/ml respectively while BDNF levels were 5314.3 pg/ml, 14671.4 pg/ml, 11742.9 pg/ml, 5528.6 pg/ml, and 11028.6 pg/ml respectively.
Conclusions: This study demonstrates the feasibility of adjunctive treatment with PPS in patients with schizophrenia. Soluble P-selectin decreased in the first patient, suggestive of target engagement by PPS. The less consistent change in the levels seen in the second patient might be due to suboptimal adherence because unlike the first patient who received PPS while on the inpatient, the second patient received PPS on an outpatient basis. Though speculative, the increase in plasma BDNF levels in both patients might be a reflection of decreased brain inflammation due to reduced spread of inflammation from the periphery after endothelial P-selectin blockade by PPS. Spread of inflammation from the periphery to the CNS via P-selectin-involved endothelial activation deserves further study in patients with schizophrenia. The potential role of PPS in the treatment of schizophrenia should also be evaluated using an experimental therapeutics paradigm.
Keywords: Schizophrenia, Inflammation, P-selectin, Pentosan Polysulfate Sodium
Disclosure: Nothing to Disclose.

T47. Differential Associations Between Brain MRS-Measured Glutamate, GABA, and Glutamine and Recent Heavy Drinking in Individuals With Alcohol Use Disorder and Social Drinkers

James Prisciandaro*, Joseph Schacht, Andrew Prescot, Perry Renshaw, Truman Brown, Raymond Anton

Medical University of South Carolina, Charleston, South Carolina, United States

Background: Proton Magnetic Resonance Spectroscopy (1H-MRS) studies have demonstrated abnormal levels of a variety of neurometabolites in inpatients and outpatients with Alcohol Use Disorder (AUD) following acute alcohol withdrawal relative to healthy controls. Furthermore, differences in glutamate levels between these populations have been shown to depend heavily on participants' recent levels of alcohol drinking. In contrast, few studies have compared neurometabolite levels between less severe, treatment-naïve AUD individuals and controls or examined relationships between neurometabolites and alcohol drinking in these individuals. The present study compared prefrontal glutamate, glutamine, and GABA levels between treatment-naïve AUD individuals and social drinkers (SD) and examined associations between prefrontal glutamate, glutamine, and GABA levels and recent heavy drinking within each of these groups.
Methods: Twenty non-treatment seeking AUD individuals (M[SD] age = 26.8[6.2], male $n = 15$), who met criteria for DSM-IV alcohol dependence and reported ≥ 1 heavy drinking day (and ≥ 25 drinks total) in each of the 2 weeks

preceding the study, and 21 SD comparators (M[SD] age = 26.6[9.0], male $n = 11$) were recruited via community advertisements. Exclusion criteria included current DSM-IV Axis I disorder and/or positive urine drug or alcohol breath screens on scan-day. Participants completed an MRI scan including a two-dimensional j-resolved proton MRS scan (Siemens-3T) in a 25x25x30mm voxel in the dorsal anterior cingulate cortex. Neurometabolite concentrations were estimated using the ProFit algorithm, scaled to water, and corrected for voxel cerebrospinal fluid content. Glutamate, glutamine, and GABA concentrations were entered as dependent variables in separate general linear models, with participant group as the primary predictor. Pearson correlations between glutamate, glutamine, and GABA concentrations and number of heavy drinking days (HDD), collected with a timeline followback calendar method, in the 14 days preceding MRS were estimated within AUD and SD participant groups.

Results: AUD and SD participants did not differ on age, sex, race, or smoking status (ps > .10), however, AUD (M = 14.4) had fewer years of education than SD (M = 16.3, $p < .01$). Additionally, AUD (M = 61.1%) had a lower within-voxel gray matter to brain matter (GMBM) ratio than SD (M = 64.9%, $p < .01$). AUD participants had significantly lower concentrations of GABA (Cohen's D = 0.79, $p = 0.016$) and glutamine (Cohen's D = 0.88, $p = 0.008$), but not glutamate (Cohen's D = 0.34, $p = 0.290$), relative to SD. Education and GMBM ratio were explored as covariates across all statistical models, but were excluded from final model estimation as they were not significantly associated with any of the dependent variables (ps > .10). Only glutamate level was significantly associated with number of recent HDD in the AUD group (r = -0.49, $p = 0.028$); none of the examined neurometabolites were significantly associated with recent HDD in the SD group (rs < [0.33], ps > 0.10).

Conclusions: The primary findings from the present study were that treatment-naïve, AUD individuals had significantly lower levels of prefrontal GABA and glutamine than SD, and that glutamate alone was significantly associated with number of recent heavy drinking days in the AUD participant group. Taken in context with the extant literature, these findings support the interpretation that differences in glutamate levels in AUD relative to SD may be state-dependent (e.g., dependent on recent heavy alcohol drinking) whereas differences in other neurometabolite levels between these groups (e.g., GABA and glutamine) are not. The results from the present study additionally extend past findings of abnormal neurometabolite levels in AUD relative to SD downward to younger, less-severe, treatment-naïve AUD individuals.

The present study was supported by NIAAA P50 AA010761 (PI: Becker). Dr. Prisciandaro was supported by K23 AA020842, Dr. Schacht was supported by K99 AA021419, and Dr. Anton was supported by K05 AA017435.

Keywords: Glutamate GABA, Alcohol Use Disorder, Proton Magnetic Resonance Spectroscopy

Disclosure: Nothing to Disclose.

T48. Increased miR-137 in the Adult Amygdala Leads to Epigenetic Remodeling and Increased Anxiety Following Adolescent Alcohol Exposure

Evan Kyzar*, Huaibo Zhang, Subhash Pandey

Psychiatric Institute, University of Illinois at Chicago, Chicago, Illinois, United States

Background: Binge drinking during adolescence is a crucial risk factor for the development of alcoholism and comorbid psychiatric disorders including anxiety in adulthood. Epigenetic mechanisms such as histone acetylation and DNA methylation, particularly in emotional brain circuitry such as the amygdala, are altered by adolescent alcohol exposure and contribute to the observed negative affective states. MicroRNAs (miRNAs) are small non-coding RNAs that bind to and regulate mRNA transcripts post-transcriptionally. As miRNAs are involved in neurodevelopment and epigenetic reprogramming, we investigated the role of miRNAs in adolescent intermittent ethanol (AIE) exposure-induced adult psychopathology.

Methods: Adolescent male rats were exposed to 2g/kg ethanol (2 days on/off; AIE) or intermittent n-saline (AIS) during post-natal days (PND) 28-41 and allowed to grow to adulthood (PND 92-102) to investigate the lasting effects of AIE. We measured anxiety-like behaviors using the elevated plus maze (EPM) test following an acute challenge with 2 g/kg ethanol in adulthood in both AIS and AIE adult rats. We performed a microarray on miRNAs in the amygdala of adult AIS and AIE rats. qPCR and chromatin immunoprecipitation (ChIP) assays were used to validate miRNA and mRNA levels of various target genes and the histone methylation status of brain-derived neurotrophic factor (BDNF) in the amygdala. In addition, we cannulated AIS and AIE rats directly into the central nucleus of amygdala (CeA) and, after recovery, rats were infused with a locked nucleic acid antagonist of miR-137 (antagomiR-137) prior to anxiety testing in the EPM.

Results: Profiling of miRNAs revealed differential expression of various miRNAs in the amygdala of AIS and AIE adult rats. miR-137, a miRNA that is crucial for neurodevelopment, was non-significantly increased in the microarray analysis but significantly increased in AIE rats after qPCR validation. AIE rats display increased anxiety-like behavior in the EPM, and this behavior, along with amygdala levels of miR-137, returned to control-like levels following an acute challenge with ethanol in adulthood. Methyl-CpG binding protein 2 (MeCP2) is known to regulate miR-137 expression, and we observed decreased Mecp2 mRNA and decreased MeCP2 binding to the promoter of miR137 in the AIE adult amygdala compared to AIS rats. We have previously shown that lysine-specific demethylase 1 (Lsd1) and Lsd1+8a (a neuron-specific splice variant) are decreased in the AIE adult amygdala, and both transcripts are targets of miR-137. LSD1 binding to the BDNF exon IV promoter is also decreased in the amygdala of AIE adult rats. Interestingly, infusion of antagomiR-137 directly into the CeA leads to a rescue of the increased anxiety-like behaviors seen in AIE adult rats.

Conclusions: These novel results indicate that adolescent alcohol exposure causes enduring effects on specific miRNAs, such as miR-137 that are both regulated by

epigenetic effectors and cause downstream effects on chromatin remodeling and behavior. Our results highlight miR-137 in the amygdala as a potential therapeutic target for anxiety susceptibility and alcohol use disorders. (Funded by NIAAA-NADIA UO1-AA019971, U24-AA024605 and P50-AA 022538 grants to SCP and F30-AA024948 to EJK).
Keywords: Adolescent Binge Drinking, Epigenetics, Micro-RNA, Amygdala
Disclosure: Nothing to Disclose.

T49. Inhibition of the Rostromedial Tegmental Nucleus Reverses Withdrawal-Induced Negative Affect

Elizabeth Glover*, E. Margaret Starr, Nathan Burnham, Ying Chao, Thomas Jhou, L. Judson Chandler

Medical University of South Carolina, Charleston, South Carolina, United States

Background: Alcohol withdrawal is associated with a hypodopaminergic state and increased negative affect, both of which are thought to play a significant role in the propensity for relapse. The rostromedial tegmental nucleus (RMTg) exerts inhibitory control over midbrain dopamine neurons and activity within this region is associated with the aversive properties of cocaine and alcohol. Together these data suggest that the RMTg plays a role in mediating drug-induced aversive states.
Methods: To investigate the role of the RMTg in withdrawal and withdrawal-induced negative affect, adult male Long-Evans rats were rendered ethanol dependent using chronic intermittent exposure to ethanol vapor. Measures of neuronal activity and withdrawal-induced negative affect were evaluated across the time course of acute withdrawal (0, 6, 12, 24 hr after final ethanol exposure). cFos expression was measured using standard immunohistochemical methods. Anhedonia-like behavior was measured by evaluating changes in reward sensitivity using the curve-shift method in an intracranial self-stimulation paradigm. Anxiety-like behavior was measured using a battery of tests including light-dark box, elevated plus maze and open field.
Results: A significant increase in cFos expression was observed in the RMTg across the time course of acute withdrawal with peak expression occurring at the 12 hr time point when withdrawal symptom severity is also thought to be at its peak ($p \leq 0.01$). A similar pattern of cFos expression was observed in the lateral habenula – a region that sends prominent glutamatergic projections to the RMTg ($p \leq 0.01$). Compared to ethanol-naïve rats, ethanol dependent rats trained to self-administer intra-cranial electrical stimulation exhibited a significant rightward shift in frequency-rate responding during acute withdrawal indicative of a decrease in reward sensitivity, or anhedonia-like behavior ($p \leq 0.05$). Of note, the time course of this rightward shift mimicked the time course of RMTg cFos induction. Ongoing studies also suggest that inhibition of the RMTg can attenuate this withdrawal-induced decrease in reward sensitivity. Acute withdrawal was also associated with a significant increase in anxiety-like behavior that was significantly attenuated following inhibition of the RMTg ($p \leq 0.05$).
Conclusions: Together these data suggest that the RMTg plays an important role in withdrawal-induced negative affect and therefore may be critically involved in the neurobiological mechanisms underlying relapse.
Keywords: Addiction, Reward and Aversion, Anhedonia, Anxiety
Disclosure: Nothing to Disclose.

T50. Increased Activation in Precuneus and Parahippocampus When Smokers View Tobacco Retail Outlets: Results of a GPS + fMRI 'Community Neuroscience' Study

Jason Oliver*, Matthew Hallyburton, Maggie Sweitzer, Kurt Ribisl, F. Joseph McClernon

Duke University, Durham, North Carolina, United States

Background: Tobacco retail outlets (TROs) are ubiquitous and unavoidable in the lives of current, former and future smokers. Each point of contact with these outlets represents an opportunity to acquire tobacco products for immediate or deferred use, potentially eliciting craving responses and motivating smoking behavior. Indeed, recent studies indicate proximity to TROs is associated with craving among current smokers and frequent contact with TROs is associated with smoking relapse among recent quitters. As such, TROs are arguably the single greatest environmental risk factor for cancer in the United States. Moreover, tobacco purchasing is an inherent step in the progression from smoking lapse to relapse, but has largely been ignored by the existing clinical literature. Despite all of the above, surprisingly little research has been conducted to understand the behavioral or neural mechanisms through which TROs influence behavior. The present project aims to evaluate these mechanisms using a novel combination of GPS-enabled location tracking, mapping of community TROs, and fMRI assessments.
Methods: Smokers ($N = 17$) and non-smokers ($N = 17$) carried GPS loggers for a period of 14 days. Individual GPS tracks were intersected with a county-wide geodatabase of TRO and non-TRO retail stores (e.g. clothing stores) to identify outlets that fell INSIDE ($\geq$ 1 point within a 200m Euclidean buffer from store entrance during tracking) and OUTSIDE (<1 point within a 200m Euclidean buffer during tracking) activity space for each participant. The twelve most frequently encountered TROs and non-TROs inside activity space and a random selection of twelve TROs and non-TROs outside activity space were identified for each participant for subsequent presentation. These were presented along with twelve static TROs and non-TROs located in another county (CONTROL) with similar demographics to the study location. Following 24-hour smoking abstinence, participants underwent BOLD fMRI scanning while viewing images of store exteriors. Participants rated their familiarity with each outlet during scanning (1 = not at all familiar; 8 = extremely familiar). In a separate session, smokers also rated their likelihood of purchasing tobacco from each outlet.
Results: Main effects of Activity Space were observed in the precuneus (Left: $F = 21.7$, $p < .001$; Right: $F = 23.5$, $p < .001$), parahippocampus (Left: $F = 4.7$, $p = .009$; Right: $F = 4.5$, $p = .012$) and right ventral striatum ($F = 4.8$, $p = .009$). In all cases, these main effects indicated greater BOLD response to stores falling INSIDE activity space relative to stores OUTSIDE activity (p's $< .05$) space or CONTROL

(p's < .05) stores, with no difference in activation between stores OUTSIDE and CONTROLs (p's > .05). Smoking Status x Store Type interactions were also present in the precuneus (Left: F = 11.1, p = .001; Right: F = 10.2, p = .001) and parahippocampus (Left: F = 5.8, p = .016; Right: F = 3.5, p = .061). These interactions indicated smokers had significantly greater BOLD response to TROs relative to non-TROs in both precuneus (p's < .001) and parahippocampus (p's < .05), whereas non-smokers did not exhibit Store Type effects in either region (p's > .3). These effects were additive with Activity Space main effects, resulting in smokers exhibiting substantially greater BOLD responses to INSIDE-TROs relative to all other categories. Findings for familiarity ratings were parallel to imaging findings, with a main effect of Activity Space (F = 25.8, p < .001) and a Smoking Status x Store Type interaction (F = 32.0, p < .001) exhibiting the same overall pattern of findings. As expected, smokers reported having a greater likelihood of purchasing tobacco from TROs relative to non-TROs (F = 461.9, p < .001), but a main effect of Activity Space (F = 4.8, p = .011) indicated greater likelihood of purchasing tobacco from stores INSIDE activity space relative to OUTSIDE (p = .03) and CONTROL (p = .004) stores.

Conclusions: Findings confirm the presence of a previously unstudied biomarker (i.e. brain reactivity to personally relevant TROs) that could serve as a novel intervention target for smoking cessation. That is, neural regions related to spatial memory (parahippocampus) and self-relevance (precuneus) exhibited differential patterns of activation as a function of activity-based contact, store type and smoking status. Parallel behavioral findings strongly suggest these effects relate to store familiarity and likelihood of making tobacco purchases. Together, these findings provide insight into a mechanism linking smoking lapse to relapse. To our knowledge, this is the first study to use GPS tracking to ideographically identify community-level cues for use in a functional imaging study, as well as the first study to examine neural responses to exterior storefronts. Despite the efficacy of pharmacotherapy for smoking cessation and other behavioral problems, we still have an extremely limited understanding of how pharmacotherapy impacts how patients process information in the real world. The 'community neuroscience' approach developed and employed here can be easily adapted for many other purposes across a range of mental and physical illnesses (e.g. alcohol use, eating/obesity). Accordingly, this study represents a critical advancement in our mechanistic understanding of how community-level features influence human motivation and behavior.

Keywords: Tobacco, GPS, Functional MRI (fMRI), Smoking, Translational Neuroscience

Disclosure: Nothing to Disclose.

T51. Performance in a Reinforcer Devaluation Task is Causally Linked to Prelimbic Cortex to Nucleus Accumbens Core Transmission Suggesting a Mechanism for Cocaine-Induced Impairments in Flexible Behavior

Elizabeth West*, Mark Niedringhaus, Heather Ortega, Rachel Haake, Regina Carelli

University of North Carolina, Chapel Hill, North Carolina, United States

Background: A history of cocaine impairs the ability to adjust behavior away from reward-predictive cues following reward devaluation, a canonical test of flexible behavior. Several brain regions including the prelimbic cortex (PrL) and the nucleus accumbens (NAc) are necessary for flexible behavior. Phasic activity in the NAc core, a predictor of flexible behavior (West and Carelli, 2016) and a primary target of PrL, is abolished in cocaine-exposed rats during Pavlovian conditioning (Saddoris and Carelli 2014). Thus, we sought to determine how a history of cocaine alters neural cell firing upstream from the NAc in the PrL and impairs flexible behavior, and whether the PrL–>NAc pathway is causally linked to flexible behavior.

Methods: For experiment 1, we used electrophysiology (multineuron) methods to record activity from PrL in cocaine-exposed rats ($n = 7$; 14 days of 2-hr cocaine self-administration; 0.33 mg/inf, ~25 mg/kg) and controls ($n = 10$, 14 days of saline/water self-administration) during Pavlovian conditioning. Briefly, after 3 weeks of abstinence, rats were presented with two distinct cues as conditioned stimuli (CS+; one predicting a sugar pellet and one predicting a food pellet) and two cues that did not predict a reward (CS-); 10 trials each. After 10 sessions, rats underwent a devaluation procedure to induce a conditioned taste aversion to the sugar pellets. Rats were then tested on the same Pavlovian task (under extinction) to evaluate their ability to avoid CS+ associated with the devalued outcome. For experiment 2, rats were infused with either a Cre-On, AAV virus expressing halorhodopsin-mCherry (AAV-EF1a-DIO-eNpHR3.0-mCherry, halo group) or a Cre-On, AAV virus expressing mCherry alone (AAV-EF1a-DIO-mCherry, control group) bilaterally into the PrL. To selectively express halorhodopsin in PrL neurons that directly project to the NAc core, we injected a Cre-GFP virus bilaterally into the NAc core. After 8 weeks of recovery, we placed optical fibers into the PrL and following 1 additional week of recovery, we ran these animals in the same Pavlovian task as above except a 550 "lime" LED (10 mW) was transmitted via patch cable during all the cues during conditioning and testing.

Results: We found that on the last day of conditioning, control and cocaine-exposed rats spent significantly more time in the food cup during both CS+ compared to the CS- [day X cue interaction: controls, $F(3,24) = 16.5$, cocaine $F(3,15) = 15.2$, $p < .05$], showing successful discrimination of rewarded vs unrewarded cues. Post-devaluation, controls successfully avoided the CS+ associated with the devalued reward; however, cocaine rats continued responding to both CS+ equally suggesting impaired behavioral flexibility (determined by a Devaluation Index, DI with DI = 0 reflecting inflexible behavior, DI = 0.30 for controls vs. 0.003 for cocaine; $t = 2.7$, $^*p < .05$). Recordings of PrL neurons on the last day of Pavlovian conditioning revealed distinct populations of cells that were excited or inhibited during cues (classified as "phasic"). In control rats, the % of phasic (excitations and inhibitions) in the PrL predicted how well rats could flexibly shift behavior measured by the corresponding DI ($R^2 = 0.41$, $p < 0.05$) while no such correlation existed in cocaine-exposed group ($R^2 = 0.01$, $p > 0.1$). The percentages of phasic PrL neurons were similar in cocaine-exposed and controls (57% controls rats and 63% cocaine-exposed rats); however phasic PrL neurons in the controls were predominately excited (23/39) whereas phasic

PrL neurons in cocaine rats were predominately inhibited (16/24). We hypothesize the shift to an inhibition of PrL neurons following cocaine drives the loss of phasic activity in the NAc core following a history of cocaine, leading to an inflexible phenotype. In support, when the PrL–>NAc pathway was inhibited during cue presentations during Pavlovian conditioning using optogenetics, behavioral flexibility was impaired (control, $n = 4$, DI = 0.28; halo, $n = 4$, DI = 0.02), similar to that observed following cocaine.

Conclusions: A history of cocaine does not impair the ability of rats to direct behavior towards a reward predictive cue. However, a history of cocaine interferes with the ability of rats to alter behavior after reward devaluation and this inability to change behavior is linked to neural activity in the PrL. Critically, selectively blocking transmission from PrL to NAc core is sufficient to disrupt behavioral flexibility to a similar degree as cocaine-exposed rats suggesting that this connection is required during cue-outcome associative learning to drive behavioral flexibility. As such, a loss of transmission through this pathway may underlie behavioral deficits following a history of cocaine.

Keywords: Cocaine, Prelimbic, Reward Devaluation, Nucleus Accumbens Core

Disclosure: Nothing to Disclose.

T52. Alcohol Dependence Alters IL-1 Regulation of GABA Transmission in the Mouse Prelimbic Cortex

Florence Varodayan*, Michael Steinman, Sarah Montgomery, Amanda Roberts, Michal Bajo, Marisa Roberto

The Scripps Research Institute, La Jolla, California, United States

Background: Neuroimmune signaling is considered critical in the transition to alcohol dependence, as ethanol exposure can activate the neuroimmune system and cytokines can modulate alcohol-related behaviors. In particular, the interleukin-1 (IL-1) system has emerged as a key regulator of the brain's response to alcohol. The pro-inflammatory cytokine IL-1β alters GABAergic transmission in several addiction-related brain regions (e.g. central amygdala, basolateral amygdala, hypothalamus and cerebellum), and promotes alcohol-induced neuroinflammation. Moreover, IL-1β levels are increased in human alcoholics and ethanol-dependent rodents, and this IL-1β neuroinflammatory response in rodents is associated with significant cognitive deficits. As the medial prefrontal cortex (mPFC) governs cognitive function, with the prelimbic mPFC implicated in drug-seeking behaviors and the infralimbic mPFC in impulse control/habit formation, here we investigated how ethanol dependence produces neuroadaptation in the mPFC IL-1 system to disrupt inhibitory synaptic transmission.

Methods: We investigated IL-1 regulation of GABA transmission in layer II/III pyramidal neurons in the prelimbic cortex (PrL) of naïve and ethanol-dependent C57BL/6J mice using whole-cell voltage-clamp electrophysiology to record spontaneous inhibitory GABAA-mediated postsynaptic currents (sIPSCs).

Results: We found that the agonist IL-1β (50 ng/mL; 15 min application) regulates the basal activity of layer II/III pyramidal neurons in the PrL of naive and non-dependent mice by decreasing their inhibitory GABAergic input, and this effect was reversed in ethanol-dependent mice. Conversely, the anti-inflammatory endogenous antagonist IL-1ra (100 ng/mL; 15 min application) increased GABA release in naïve mice, but decreased it in the dependent mice. To uncover the neuroadaptive mechanisms that underlie these chronic ethanol-induced changes, we next investigated whether IL-1 regulation of GABAergic transmission is sensitive to acute ethanol. After pretreatment with ethanol (44 mM; 15 min application), co-application of IL-1β increased GABA release in naïve mice, similar to the effects of IL-1β alone in dependent mice. Importantly, this IL-1β-induced increase in naïve mice required the presence of acute ethanol; after a 15-min washout of ethanol, the effect of IL-1β to decrease GABA release was restored. Potential intracellular mechanisms for this IL-1β/ethanol interaction are currently being assessed, as are possible changes in the expression of IL-1 signaling molecules after chronic ethanol exposure.

Conclusions: Overall, our data indicate that IL-1 signaling interacts with acute ethanol to reverse its effects on inhibitory neurotransmission in the PrL of naïve mice. Chronic ethanol exposure produces a sustained adaptation in the IL-1 system, such that the effect of IL-1β alone in the PrL of ethanol-dependent mice resembles the interaction of IL-1β and acute ethanol in naïve mice. Therefore, ethanol dependence-induced adaptive changes in neuroimmune signaling, such as the IL-1 system, may reflect a persistence in the dynamic effects of acute ethanol and thus, may represent novel targets for the treatment of alcoholism.

Keywords: Alcohol Dependence, GABA Transmission, Neuroimmune, Interleukin-1, Prelimbic

Disclosure: Nothing to Disclose.

T53. Extinction Enables Transient Synaptic Potentiation of D2-MSNs in Nucleus Accumbens Core and Attenuates Cocaine Seeking

Douglas Roberts-Wolfe*, Ana-Clara Bobadilla, Jasper Heinsbroek, Daniela Neuhofer, Peter Kalivas

Medical University of South Carolina, Charleston, South Carolina, United States

Background: Future therapies for drug addiction may benefit from a better understanding of the endogenous mechanisms protecting against relapse. Extinction training, in which drug seeking is repeatedly uncoupled from drug reward, attenuates subsequent drug seeking, but the underlying therapeutic mechanisms are poorly understood. Nucleus accumbens (NA) core is a key node in the neural circuitry underlying drug seeking, and is modified by extinction training. During drug seeking, medium spiny neurons (MSNs) in NA core undergo transient synaptic potentiation (tSP), in which AMPA receptors are inserted rapidly and transiently into post-synaptic dendritic spines, leading to increased AMPA:NMDA ratio. The time course of AMPA:NMDA ratio increase parallels the time course of active drug seeking behavior, and the mechanisms necessary for tSP in NA core are also necessary for reinstatement of drug seeking, suggesting that tSP is an important mechanism

underlying drug seeking. Two separate classes of MSNs expressing D1R or D2R dopamine receptors exert opposite influences on drug seeking: D1-MSNs promote and D2-MSNs suppress drug seeking. We hypothesized that extinction would suppress tSP during drug seeking, possibly in a cell-type specific manner.

Methods: Sprague Dawley rats or transgenic mice (Drd1a-tdTomato/Drd2-eGFP) self-administered cocaine infusions paired with audiovisual cues. Following 10 days of self-administration, animals engaged in 2-3 weeks of extinction training (in which neither drug nor cues were available) or home-cage abstinence (with daily handling but no return to the operant chamber). Finally, animals underwent a brief test of drug seeking, in which active lever presses delivered cocaine-associated audiovisual cues only. Animals were sacrificed and coronal slices containing NA core were prepared for whole cell patch clamp electrophysiology. A separate group of "baseline" extinguished or abstinent animals were sacrificed without return to the operant chamber. Electrically evoked "dual" AMPAR/NMDAR EPSCs were recorded from cells depolarized to +40 mV. AP5 was added to isolate AMPAR EPSCs, and AMPAR EPSCs were subtracted from dual EPSCs to calculate NMDAR EPSCs. AMPA:NMDA ratios were calculated using the peak amplitude of each current.

Results: As previously described, we observed transient synaptic potentiation (tSP) during drug seeking after extinction, in which AMPA:NMDA ratios in rats actively engaged in drug seeking were elevated relative to AMPA:NMDA ratios in baseline extinguished rats. To our surprise, tSP did not occur during drug seeking after abstinence. We hypothesized that D1- and D2-MSNs, visualized using transgenically expressed fluorescent reporter proteins, might exhibit different patterns of tSP. D1-MSNs expressed tSP during drug seeking regardless of the mice's extinction history. D2-MSNs, in contrast, were potentiated during drug seeking only in mice with a history of extinction training. In addition to enabling D2-MSN tSP, extinction training attenuated drug seeking in both rats and mice.

Conclusions: Here we find that D1-MSNs display tSP during drug seeking and that this is not modified by prior extinction training, while a history of extinction training potentiates synapses onto D2-MSNs in mice while they are seeking drug. Overall, these results suggest that extinction confers a form of metaplasticity to D2-MSNs that enables them to rapidly undergo synaptic potentiation during drug seeking. In parallel with D2-MSN synaptic potentiation, prior extinction training attenuates drug seeking. Future research will determine the mechanism of extinction induced metaplasticity, and the causal role of D2-MSN tSP in suppressing drug seeking.

Keywords: Synaptic Plasticity, Cocaine, Extinction, Nucleus Accumbens, D2 Receptor

Disclosure: Nothing to Disclose.

T54. Stress-Induced Reinstatement of Nicotine Conditioned Place Preference in Mice: Sex Differences and Effects of Guanfacine

Angela Lee*, Yann Mineur, Marina Picciotto

Yale University School of Medicine, New Haven, Connecticut, United States

Background: Relapse to drug use after periods of abstinence is a critical problem in addiction, including the nicotine addiction that drives tobacco use. Stress is an important factor driving relapse to nicotine seeking, and clinical and epidemiological research indicate that women are particularly vulnerable to stress-induced relapse. Further, research in human smokers has shown that targeting the noradrenergic system with guanfacine, an alpha2-adrenergic agonist, can reduce stress-precipitated smoking in a laboratory setting. Therefore, pharmacological interventions that can successfully target this stress response may be especially beneficial for women in reducing relapse to smoking. Preclinical models of nicotine addiction can be particularly useful in unraveling the brain mechanisms contributing to sex differences in stress-induced reinstatement of nicotine seeking. In these studies, we describe the validation of a model of stress-induced nicotine seeking in male and female C3H/HeJ mice, and evaluate the effects of guanfacine in this model.

Methods: We used a conditioned place preference (CPP) model in C3H/HeJ mice to evaluate sex differences in stress-induced nicotine seeking. Male and female mice were trained with nicotine (0.5, 0.75, or 1.0 mg/kg, subcutaneously) and saline in alternating chambers of the CPP apparatus in a balanced design. Control mice were trained with saline in both chambers. After CPP acquisition, mice were left in their home cages for two weeks, after which they began extinction training. During extinction, mice were allowed free access to all three chambers of the CPP apparatus. Following extinction, reinstatement was tested with a 6-minute forced swim stressor. The effect of noradrenergic manipulation was tested by injecting guanfacine (0.15 mg/kg, intraperitoneally) or saline 30 min prior to the forced swim stressor. C-fos staining was measured throughout the brain in separate cohorts of male and female mice to identify any regional sex differences in neuronal activation that might contribute to the behaviors evaluated. Mice were exposed to the same regimen of nicotine and saline injections, but without the CPP training, followed by two weeks in the home cage, and were then administered saline or guanfacine 30 min prior to a forced swim stressor. Brain tissue was then collected and processed for immunohistochemistry and c-fos expression was quantified in several brain areas of interest, including the prefrontal cortex (PFC), nucleus accumbens (NAcc), hippocampus (HPC), basolateral amygdala (BLA), ventral tegmental area (VTA), locus coeruleus (LC), and nucleus of the solitary tract (NST).

Results: We observed a dose-dependent preference for the nicotine-paired chamber that differed in male and female mice, with male mice responsive to lower doses of the drug. Both male and female mice subsequently demonstrated successful extinction learning after repeated exposure to the CPP chambers in the absence of nicotine, and reinstatement of preference for the nicotine-paired chamber following a forced swim stress. Interestingly, our preliminary data suggest that guanfacine blunts this stress-induced reinstatement in a sex-dependent manner, with female mice more sensitive than male mice to guanfacine administration.

Conclusions: We have validated a behavioral paradigm that can be used to evaluate nicotine-seeking behavior in male and female mice following extinction in response to stress. Our results show sex differences in the dose-response

function for nicotine CPP acquisition and stress-induced reinstatement in C3H/HeJ mice. This preliminary work with guanfacine will be followed up with additional experiments to identify the mechanisms that contribute to sex differences in response to noradrenergic manipulations. Further immunohistochemical studies will also help determine the neurobiological substrates involved in these sex differences.
Keywords: Nicotine Addiction, Stress-Induced Reinstatement, Sex Differences, Guanfacine
Disclosure: Nothing to Disclose.

T55. Preliminary Evidence That Cannabis Use Moderates the Relationship Between Alcohol Consumption and Circulating Cytokines

Hollis Karoly*, L. Cinnamon Bidwell, Raeghan Mueller, Kent Hutchison

University of Colorado, Boulder, Boulder, Colorado, United States

Background: Alcohol use disorders (AUDs) are associated with significant morbidity, mortality and socioeconomic costs in the United States. Despite decades of research on the topic, the best currently available treatments are only modestly effective. This is likely due, in part, to the neurobiological complexity of AUDs. Although the molecular mechanism(s) driving the effects of alcohol on the brain and body are not fully understood, human and animal studies in recent years have converged to underscore the important role of alcohol-related aberrations within inflammatory signaling pathways in both the brain and peripheral tissues. Briefly, alcohol binds to cell-surface receptors located on immune cells in the brain (i.e., microglia) and periphery (i.e., macrophages), and triggers signaling pathways via the oxidant-sensitive pro-inflammatory transcription factor NF-kB, that ultimately lead to an increase in pro-inflammatory cytokines. Chronic activation of these pathways may be associated with significant tissue damage, which may contribute to numerous negative effects of heavy alcohol use, such as damage to frontal control regions of the brain. Conversely, cannabis has been associated with decreased inflammatory signaling. However, to date, research on this topic is limited. Given that alcohol and cannabis use frequently co-occur, the impact of cannabis use on inflammatory signaling in AUDs warrants examination. Thus, the present study explored the relationship between self-reported alcohol and cannabis use and circulating levels of the pro-inflammatory cytokines IL-6, IL-8 and IL-1B in the blood.
Methods: We collected a sample of $N = 66$ regular drinkers, with a mean age of 30.08 (SD = 4.7, range 25-40). Subjects ranged from light to heavy alcohol users, with mean drinks per drinking day = 3.48 (SD = 2.4, range = .3-10.9) and mean Alcohol Dependence Scale (ADS) total score = 6.17 (SD = 4.4, range = 1-20). Across the entire sample, we examined circulating cytokine levels in the blood using high-sensitivity ELISA assays. We also administered a battery of questionnaires assessing self-reported alcohol consumption and days of cannabis use over the past 90 days, as well as relevant demographic information. Using a series of Ordinary Least Squares (OLS) multiple regression models, we examined whether alcohol consumption, days of cannabis use and gender were associated with changes in circulating cytokine levels, and whether there was a significant interaction between alcohol consumption and cannabis use predicting circulating cytokine levels.
Results: A significant main effect of alcohol consumption emerged in the model for which IL-6 was the criterion ($b = .353$ $t(64) = 2.529$, $p = .014$), indicating that alcohol consumption is positively associated with circulating IL-6. In the model for which IL-1B was the criterion, a main effect of cannabis consumption emerged ($b = -.453$ $t(59) = -3.650$, $p = .001$), suggesting a negative relationship between cannabis use and circulating IL-1B. In the model in which IL-8 was the criterion, no main effects were observed for alcohol consumption or cannabis use. Follow-up moderation analyses indicated a significant cannabis use by alcohol consumption interaction predicting circulating IL-6 ($b = -.434$ $t(64) = -3.722$, $p < .001$), but not circulating IL-1B. Further examination of this interaction demonstrated that, covarying for gender, individuals who did not use cannabis in the 90 days prior to the study ($n = 29$) showed a strong correlation between alcohol and IL-6 (partial $r = .724$, $p < .001$), whereas those who reported using cannabis at least once in the 90 days prior to study participation ($n = 29$) did not demonstrate a significant association between alcohol and IL-6 (partial $r = -.138$, $p = .452$).
Conclusions: These findings are consistent with previous work suggesting a positive association between alcohol consumption and circulating pro-inflammatory cytokines in the blood, as well as prior research indicating that cannabis use is associated with decreased inflammatory signaling. We further demonstrated that cannabis may serve to moderate the relationship between alcohol and circulating cytokines. However, given the relatively small sample size, as well as the lack of prior research demonstrating convergence between peripheral circulating cytokines and inflammation in the brain, these results should be considered preliminary. Replication of these findings in brain tissue in animal studies would provide compelling evidence for the role of neuroinflammation in alcohol use disorders, as well as highlight the potential anti-inflammatory properties of cannabinoid compounds. These findings also suggest that cannabinoids should be further explored for their anti-inflammatory potential in humans, particularly in the context of alcohol use disorders. Overall, despite the preliminary nature of these results, our findings provide promising initial data to inform future investigations, with the goal of ultimately leveraging knowledge of the role of inflammatory signaling in alcohol use disorders to develop more effective treatments focused on novel targets within the immune system.
Keywords: Alcohol, Inflammation, Cannabis Use
Disclosure: Nothing to Disclose.

T56. Krüppel-Like Factor 9 mRNA in Hippocampus Dentate Gyrus is Elevated in Women in Association With Major Depression and Stressful Life Events, and Decreased With Aging in Women and Men

Cynthia Zizola, Suham Kassir, Antoine Besnard, Andrew J. Dwork, Gorazd B. Rosoklija, Rene Hen, J. John Mann, Amar Sahay, Maura Boldrini*

Columbia University, New York, New York, United States

Background: Adult neurogenesis in the ventral dentate gyrus (DG) regulates emotional behavior and antidepressant action in mice (Wu and Hen, 2014). In postmortem anterior human DG, homologous to rodent ventral DG, we found fewer mature granule neurons in untreated major depressive disorder (MDD) vs. controls (Boldrini, 2013), and more neurogenesis in MDD treated with antidepressant medications (Boldrini, 2014; 2012). We identified krüppel-like Factor 9 (Klf9) as a transcriptional regulator of DG granule cell maturation blunting dendritic spines, and Klf9 null mice showed normal stem cell proliferation, but impaired differentiation into adult-born neurons and impaired neurogenesis-dependent synaptic plasticity (Scobie, 2009; McAvoy, 2016).

Corticosteroids increase Klf9 mRNA expression in Xenopus tropicalis tadpole brain (Shewade, 2017), postnatal mouse hippocampus, and mouse hippocampal cell line HT-22 (Bagamasbad, 2012; Datson, 2011), and Klf9 is down-regulated in response to chronic restraint stress, suggestive of adaptation (Datson, 2011). The expression of Klf9 mRNA in human adult DG has never been studied. We aimed to examine Klf9 mRNA levels in untreated women and men with MDD and non-psychiatric controls, and effects on expression related to recent stressful life event severity and age.

Methods: Klf9 mRNA was quantified postmortem by radioactive in situ hybridization and autoradiogram densitometry in anterior DG from untreated MDD subjects who died from suicide and age, sex and postmortem interval-matched non-psychiatric controls ($n = 11$/group), including women ($n = 10$) and men ($n = 12$). All brain samples were characterized by diagnostic and other clinical data from a psychological autopsy, using our method validated for DSM axis I and II diagnoses (Kelly and Mann, 1996). The St. Paul-Ramsey scale was used to assess life events stress load in three months prior to death, and brain and blood toxicology and neuropathology examination helped to rule out recent psychotropic drug exposure or neuropathology. We used a Klf-9 riboprobe generated from the 3′-untranslated region of Klf-9 (NM 010638) corresponding to nucleotides 1500–1780 (Scobie, 2009). Linearized plasmid was labeled using [35S]-CTP, and, after in situ hybridization, slides were exposed to sheet film (Hyperfilm, B-Max", Amersham) in x-ray film cassettes. Then film was developed, and autoradiograms digitized using Densita software (MBF Inc.). Calibrations were done by sampling [14C]-146 standards (American Radiolabeled Chemicals) co-exposed with the slides, given the linear relationship between 35S and 14C radioactivity (Miller, 1991). Slide background density was sampled and subtracted from DG density. Regression analysis assessed correlation between age and Klf9 mRNA density. Age and stress severity were used as covariates in an univariate analysis of variance where group and sex were fixed factors and Klf9 mRNA was the dependent variable.

Results: There was a sex*group effect ($F1,22 = 4.676$, $p = .042$), an effect of age ($F1,22 = 12.361$, $p = .002$), an effect of stress ($F1,22 = 3.625$, $p = .031$) and a sex*stress effect ($F1,22 = 8.057$, $p = .002$) on Klf9 mRNA density. Female MDD had more Klf9 mRNA vs. female controls ($F1,22 = 7.166$, $p = .028$), with no change between male MDD and controls. Higher Klf9 mRNA correlated with worse stressful life event severity in women ($r2 = .814$, $p < .001$), not men. Klf9 mRNA declined with age in both women ($r2 = 0.487$, $p = .012$) and men ($r2 = 0.391$, $p = .017$).

Conclusions: We suggest the age-related decline in Klf9 expression in human brain may result in impaired differentiation of newborn DG neurons and decreased neurogenesis-dependent synaptic plasticity, as shown in mice, together with increased anxiety-like behavior and impaired contextual fear discrimination learning (Scobie, 2009). Klf9 regulation of genes that could favor the integration of newborn neurons in the DG may support the creation of new neuronal networks in the aging hippocampus (McAvoy, 2016).

Stress- and MDD-associated higher Klf9 expression in women might be a pathogenic pathway to MDD found in women and not men and contribute to the higher prevalence of depression in women (Vikram, 2004). Greater Klf9 expression in women could result in decreased dendritic spines and less progenitor cell activation, as observed in mice (McAvoy, 2016). In line with our findings, corticosteroids increase Klf9 mRNA (Bagamasbad, 2012; Shewade, 2017) and elevated corticosteroids are found in MDD (Vythilingam, 2004), while repeated stress exposure may contribute to the development of depression (Krugers, 2010). Hippocampus plasticity in response to stress shows sexual dimorphism: exposure to stressors increases CA1 synaptic spine density in males and decreases them in female rats (Shors, 2001). Women have higher corticotropin releasing factor (CRF)-induced adrenocorticotropic hormone and cortisol compared to men in dexamethasone suppression-CRF stimulation test (Bourke, 2012).

Moreover, Klf9 overexpression in mouse hippocampal neuron cell line, as we observed in the DG of women with MDD, suppresses Wnt (Wingless) signaling (Knoedler, 2017), a pathway that increases dendritic arborization in mice (Hiester, 2013; Rosso, 2004). In humans, antidepressants increase neuroplasticity via Wnt pathways (Sani, 2012). The functional relationships of Klf9 expression with HPA axis, Wnt signaling, neurogenesis and dendrite plasticity needs further investigation in human DG in MDD and relative to antidepressant treatment effects.

Keywords: Neuroplasticity, Aging, MDD, Life Stress, Women's Mental Health

Disclosure: Nothing to Disclose.

T57. Cognitive Control Network Connectivity Specifically Predicts Executive Functioning Among Healthy Older Adults

Sara Weisenbach*, Vincent Koppelmans, Joseph Kim, Anna Arp, Nathalie Li, Victor Patron, Natalie Grubman, Robert Welsh, Jon-Kar Zubieta, Scott Langenecker

University of Utah, Salt Lake City, Utah, United States

Background: There has been a growing interest in the neuroimaging literature into the functioning and coherence of brain networks, in contrast to single regions of interest. Three primary brain networks that have been identified

include the Default Mode Network (DMN), Salience Network (SN), and Cognitive Control Network (CCN). While there is evidence that the connectivity within these networks predicts performance variations among individuals with psychiatric and neurologic diseases, there is relatively less knowledge about how within-network connectivity predicts cognitive performance during healthy aging. This preliminary study sought to elucidate relationships between resting state network connectivity of the DMN, SN, and CCN and performance on a comprehensive battery of neuropsychological tests.

Methods: Nine never-depressed, cognitively normal older adults (M age = 69.9, SD = 5.4) completed a comprehensive neuropsychological test battery and an 8-minute resting state fMRI scan. Independent component analysis (ICA) was used to segregate DMN, SN, and right and left CCN for each participant using the GIFT Toolbox. Values for connectivity within each network were extracted for each participant. A series of multiple regression analyses were conducted with DMN, SN, or CCN as the predictor variable, covarying for estimated premorbid IQ. Age and education-corrected (when available) neuropsychological test scores composed outcome variables.

Results: Left CCN connectivity positively predicted performance on measures of concept formation (DKEFS-Sorting; Adj. R2 = .72, p < .01), cognitive flexibility (Wisconsin Card Sorting Test Perseverative Responses; Adj. R2 = .85, p < .01) and semantic fluency (Animal Naming; Adj.R2 = .61, p < .05). Right CCN connectivity positively predicted performance on measures of visual abstract reasoning (Matrix Reasoning, Adj. R2 = .74, p = .01) and category generation (DKEFS-Categories; Adj.R2 = .61, p < .05). Left nor right CCN connectivity was significantly associated with performance on measures of verbal (Hopkins Verbal Learning Test-2) and visual memory (Brief Visuospatial Memory Test-R), processing speed (Trails A), vocabulary knowledge (Wechsler Adult Intelligence Scale-4-Vocabulary), face-emotion perception accuracy (Face Emotion Perception Test), or psychomotor speed (Purdue Pegs). DMN connectivity positively predicted cognitive flexibility only (Adj. R2 = .72, p < .05). SN connectivity was not significantly associated with performance on any cognitive measure.

Conclusions: This preliminary study of healthy older adults reveals that connectivity of the CCN is positively associated with performance on higher-order executive functioning measures, but not related to performance in other domains, including memory, processing speed, vocabulary knowledge, face emotion perception accuracy, and psychomotor speed. In contrast, SN connectivity did not predict cognitive performance on any measure, while DMN connectivity was associated only with cognitive flexibility. These results suggest that even in the context of healthy aging, resting state connectivity of regions within the CCN (and to a lesser extent the DMN) is related specifically to performance variations in aspects of executive functioning. Ongoing work is currently underway to increase the sample size of healthy adults, and make comparisons to older adults with depression and amnestic Mild Cognitive Impairment.

Keywords: Aging, Resting State Functional Connectivity, Cognition, Executive Function

Disclosure: Nothing to Disclose.

T58. Meta-Analysis of Psychosocial Treatment Effects on Cancer Survival and Sources of Heterogeneity

David Spiegel*, Spela Krizanec, Helena Kraemer, Booil Jo, Mona Ershadi, Eric Neri, Bita Nouriani, Linn Aasly

Stanford University School of Medicine, Stanford, California, United States

Background: There are currently eight other meta-analyses that address the question of whether or not psychosocial intervention for cancer patients affects survival, but they reach divergent conclusions. The aim of this study was to perform a definitive meta-analysis that also helps us to understand the reasons for the heterogeneity among prior meta-analyses.

Methods: Databases (September-December 2015) were searched, using summary data, to identify valid randomized controlled trials that compared psychosocial intervention with usual care. The main outcome was survival. Hazard ratios and their confidence intervals were pooled to estimate the strength of the treatment effect on survival time and z-tests were performed to assess possible heterogeneity of effect sizes associated with different patient and treatment characteristics.

Results: Twelve trials were retrieved (including 2,471 patients) of which four were cognitive-behavioral (CBT), three supportive-expressive (SEGT), three psychoeducational, and two supportive interventions. The overall effect favored the treatment groups having longer survival, with a HR of 0.71 (95% Cl [0.58-0.88], $p = 0.002$, I2 = 63%). An effect size favoring survival among the treatment groups was observed in studies sampling patients with lower social support (NNT = 4.3 versus NNT = 15.4), at early stage in Cognitive Behavioral Therapy treatment (NNT = 2.3 versus NNT = -28.6) and in patients older than 50 years (NNT = 4.2 versus NNT = -20.5).

Conclusions: Psychosocial interventions may have an important effect on cancer survival. Although subgroup exploratory results should be taken with caution, they provide promising directions for future studies and crucial information for clinical decision-making.

Keywords: Cancer Survival, Psychosocial Treatment, Meta-Analysis

Disclosure: Nothing to Disclose.

T59. Impact of Age- and Stress-Related Neuroendocrine Dysfunction on Executive Functions and GABAergic Synaptic Markers in the Prefrontal Cortex

Joseph McQuail*, Sahil Ghay, Matthew Bruner, Eric Krause, Barry Setlow, Deborah Scheuer, Jennifer Bizon

University of Florida, Gainesville, Florida, United States

Background: Executive functions, including working memory and cognitive flexibility, depend upon coordinated excitatory (glutamatergic) and inhibitory (GABAergic) signaling within the prefrontal cortex (PFC). Indeed, previous work from our lab has revealed that age-related changes to the PFC complement of NMDA and GABAB receptors are mechanistically linked to executive impairments. The PFC is also enriched for receptors that bind the

stress hormone corticosterone (CORT) and this brain region innervates limbic and hindbrain nuclei that modulate the hypothalamic-pituitary-adrenal (HPA) axis to dynamically regulate circulating CORT levels. Consequently, a long-standing theory posits that the cumulative effects of stressful experiences encountered over the lifespan precipitate deleterious endocrine, neuronal and cognitive changes that emerge at advanced ages. However, little is known about the mechanisms that relate stress and/or HPA axis dysfunction to age-related impairment of PFC-dependent cognition. The current study integrated behavioral, pharmacological, and physiological approaches in young adult and aged rats to 1) characterize reciprocal relationships between CORT/HPA axis and PFC-dependent cognition and 2) to determine whether chronic stress and aging produce similar changes in PFC receptor expression.

Methods: All experiments used male, F344 rats at 6 months (young adult) or 24 months (aged) of age. In Experiment 1, young adult ($n = 11$) and aged ($n = 16$) rats were characterized for working memory performance on a PFC-dependent, delayed response test followed by determination of CORT release elicited by 1 hour of restraint stress. CORT was measured in blood samples obtained by tail-bleed during and after restraint. In Experiment 2, young adult rats were randomly assigned to unstressed (UNS, $n = 15$) or chronic, variable stress (CVS, $n = 16$) treatment conditions. The CVS regimen comprised twice daily exposure to an unpredictable schedule of stressors that included forced swims, cage flood, restraint stress and exposure to predator urine. All rats were concurrently tested for working memory performance (up to 21 days, $n = 15$) or set-shifting ability (after 28 days of UNS/CVS, $n = 16$). In Experiment 3, PFC of young adult rats exposed to UNS or CVS was harvested for measurement of glutamatergic and GABAergic synaptic markers using Western blotting techniques.

Results: In Experiment 1, aged rats were found to exhibit delay-dependent decrements in working memory relative to young; however, within the aged group, individual differences in peak CORT elicited by 1 hour of restraint stress were positively correlated with better working memory performance. In Experiment 2, 21 days of CVS exposure was sufficient to produce delay-dependent decrements in working memory performance relative to age-matched, young adult UNS controls. Surprisingly, 28 days of CVS exposure facilitated set-shift performance (vs UNS). Subsequent analyses determined that improved learning of the new rule was associated with impaired memory for the old rule, suggesting this difference was due to CVS-related mnemonic impairment. Importantly, these behavioral differences coincided with increased circulating CORT and adrenal hypertrophy in CVS rats. In Experiment 3, CVS treatment was observed to produce marginal, though not significant, reductions in protein levels of NMDA receptor subunits and no change in VGluT1, a cortex-specific marker of glutamatergic terminals. In contrast, expression of GABABR1a, a presynaptic GABA autoreceptor, and VGAT, the presynaptic vesicular GABA transporter, were significantly reduced in the PFC of CVS rats.

Conclusions: The present findings indicate that stress-related HPA dysfunction robustly impairs PFC-dependent working memory of young rats in a manner that mirrors deficits that occur normally with aging. Further, in aged rats, greater dynamic range of HPA axis response elicited by an acute stressor is a correlate of preserved working memory. Differences in working memory of chronically stressed or normally aging rats relative to young, unstressed control animals coincide with modifications to synaptic proteins that localize to PFC interneurons. Until now, much work has focused on stress- and age-related modifications to PFC pyramidal neuron dendritic geometry and spine density, leaving contributions from GABAergic interneurons comparatively under-studied. Collectively, our data suggest that PFC interneuron dysfunction is a hallmark of stress- and age-related working memory impairment and, thus, identify a common substrate through which these processes may act in concert with one another.

Keywords: Aging and Dementia, Chronic Stress, Working Memory, Hypothalamic-Pituitary-Adrenal Axis, GABA
Disclosure: Nothing to Disclose.

T60. Genetically Predicted Gene Expression in the Brain and Peripheral Tissues Associates With PTSD

Laura Huckins, Kiran Girdhar, Gabriel Hoffman, Tanja Jovanovic, Michael Breen, Sanne van Rooij, Douglas Ruderfer, Laura Sloofman, Panos Roussos, Menachem Fromer, Amanda Dobbyn, Laramie Duncan, Adam Maihofer, CommonMind Consortium n/a, Psychiatric Genomics Consortium PTSD Workgroup, Elisabeth Binder, Aliza Wingo, Caroline Nievergelt, Murray Stein, Solveig Sieberts, Bernie Devlin, Nancy Cox, Hae Kyung Im, Joseph Buxbaum, Pamela Sklar, Kerry Ressler, Eli Stahl, Nikolaos Daskalakis*

McLean Hospital, Harvard Medical School, Belmont, Massachusetts, United States

Background: Post-traumatic stress disorder (PTSD) is a debilitating psychiatric disorder occurring in individuals exposed to trauma. To date, little is known about the genetic aetiology of the disorder, although the latest GWAS (carried out by the PGC-PTSD working group) demonstrates that genetic heritability is in line with other psychiatric disorders. PTSD development involves multi-systemic dysregulation in multiple brain regions and diverse peripheral tissues. Some peripheral systems are particularly interesting since epidemiologic evidence suggests that PTSD patients commonly have cardiovascular, metabolic and immune dysregulation.

Methods: Transcriptomic Imputation approaches use machine-learning methods to impute gene expression from large genotype data using curated eQTL reference panels. These offer an exciting opportunity to compare gene associations across neurological and peripheral tissues. Here, we apply CommonMind Consortium (CMC) and Genotype-Tissue Expression (GTEx) derived gene expression prediction models to the PGC-PTSD data (9,245 cases/ 24,285 controls). Models included 12 brain regions, five cardiovascular tissues, 2 endocrine tissues, the tibial nerve, adipose tissue and whole blood.

Results: We identified 24 significant gene-tissue associations, of which 5 were in peripheral tissues (adrenal gland, heart atrial appendage, tibial artery, tibial nerve). We stratified analyses according to trauma type (civilian vs. combat trauma), sex, and self-defined ancestry. Our three strongest

associations were identified in military cohorts only, which supports the hypothesis that there is substantial genetic heterogeneity between civilian and combat PTSD risk.
We used the PsychENCODE neuronal and non-neuronal reference map for two histone marks associated with transcription and open chromatin (3-trimethyl-lysine 4 (H3K4me3) and H3-acetyl-lysine 27 (H3K27ac) to understand patterns of histone modification among our PTSD-associated genes. Preliminary analyses indicate a significant correlation between PTSD-association statistics and the presence of both histone marks (correlation with neuronal H3K4me3, Pearson rho = 0.87, $p = 3.99x10\text{-}5$). We intend to expand this analysis to include a wider range of histone modification marks.
Conclusions: We will further expand these analyses to identify tissue specific gene clusters and enriched pathways across tissues or in specific tissues. Finally, we will use neuroimaging data and physiological cardiovascular data to functionally validate our results.
Keywords: Genetics, Post-Traumatic Stress Disorder, Gene Expression, Epigenetics, Imputation
Disclosure: Nothing to Disclose.

T61. Chronic Psychosocial Stress in Mice Alters Brain Myelination in a Genetic Background-Dependent Manner

Mikaela Laine, Kalevi Trontti, Zuzanna Misiewicz, Natalia Kulesskaya, Suvi Saarnio, Aino Heikkinen, Eija Jokitalo, Dario Greco, Ingrid Balcells, Ewa Sokolowska, Iiris Hovatta*

University of Helsinki, Helsinki, Finland

Background: Chronic psychosocial stress is a well-established risk factor for anxiety disorders and major depression. Mechanisms by which chronic stress impacts susceptibility and resilience to psychiatric disorders are largely unknown. The chronic social defeat stress (CSDS) mouse model allows identification of factors underlying resilience and susceptibility to chronic psychosocial stress, in a controlled manner not possible in human settings. It involves daily 5-10 minute confrontations of two conspecific male mice using the resident-intruder paradigm for 10 days. This naturalistic stress model leads to long-term plastic changes in the brain, and consequently the stress susceptible mice show increased depression and anxiety-like behavior. We used this model and unbiased genome-wide gene expression profiling to identify biological pathways most strongly affected by CSDS. Since we expected genetic background to influence the transcriptomic response to chronic stress, we used two inbred mouse strains, C57BL/6Crl (B6; a non-anxious inbred strain) and DBA/2Crl (D2; an anxious inbred strain).
Methods: To identify brain gene expression changes taking place after chronic psychosocial stress, we used CSDS, and carried out RNA-seq from medial prefrontal cortex (mPFC), ventral hippocampus (vHPC), and bed nucleus of stria terminalis (BNST). To establish whether and how myelin thickness and structure are altered after stress, we carried out transmission electron microscopy (TEM). We also performed immunohistochemical staining with anti-CNPase antibody and measured the thickness of corpus callosum, a large white matter track.
Results: The two mouse strains showed a distinct behavioral response to stress, as measured by the social avoidance test carried out after social defeat. 69 % of the B6 mice but only 11% of D2 mice were resilient to stress, the remainder being susceptible. We discovered that the expression levels of several myelination-related genes differed substantially due to chronic stress in both brain regions, and 'myelination' pathway was over-represented in gene-set enrichment analysis. Using TEM we established that the B6 stress resilient mice had significantly thicker myelin of small diameter mPFC axons compared to the B6 control mice. In the vHPC susceptible B6 mice had thinner myelin compared to B6 control mice. D2 resilient mice had thinner myelin in both vHPC and mPFC compared to controls or susceptible mice, and in the vHPC this effect was specific to small diameter axons. Corpus callosum thickness did not differ between stressed and control animals as measured by immunohistochemical staining, suggesting that stress effects on myelination are regionally selective.
Conclusions: Our findings suggest significant white matter plasticity in response to chronic psychosocial stress in mice. Such differences have previously been observed in response to early-life stress or social isolation in adult mice. Our results extend these previous findings to psychosocial stress, and demonstrate that the pattern of myelination differences is dependent on the genetic background and varies across brain regions.
Keywords: Psychosocial Stress, Myelin, RNA-sequencing, Inbred Mouse Strain
Disclosure: Nothing to Disclose.

T62. Examining the Anxiolytic Effect of Floatation-REST

Justin Feinstein*, Sahib Khalsa, Obada Al Zoubi, Hung-wen Yeh, W. Kyle Simmons, Murray Stein, Martin Paulus

Laureate Institute for Brain Research, Tulsa, Oklahoma, United States

Background: Floatation-REST (Reduced Environmental Stimulation Therapy) reduces sensory input to the nervous system through the act of floating supine in a pool of water saturated with Epsom salt. The float experience is calibrated so that sensory signals from visual, auditory, olfactory, gustatory, thermal, tactile, vestibular, gravitational and proprioceptive channels are minimized, as is most movement and speech. This open-label study aimed to examine whether Floatation-REST would attenuate symptoms of anxiety, stress, and depression in a clinical sample.
Methods: Fifty participants were recruited across a spectrum of anxiety and stress-related disorders (posttraumatic stress, generalized anxiety, panic, agoraphobia, and social anxiety), most ($n = 46$) with comorbid unipolar depression. Measures of self-reported affect were collected immediately before and after a 1-hour float session, with the primary outcome measure being the pre- to post-float change score on the Spielberger State Anxiety Inventory. A subset of the sample ($n = 30$) underwent a second floatation session during which

time indices of blood pressure and heart rate variability were measured using wireless and waterproof sensors.
Results: Overall, the procedure was well-tolerated, with no major safety concerns or adverse events. Irrespective of diagnosis, Floatation-REST substantially reduced state anxiety (estimated Cohen's d > 2). Moreover, participants reported significant reductions in stress, muscle tension, pain, depression and negative affect, accompanied by a significant improvement in mood characterized by increases in serenity, relaxation, happiness and overall well-being ($p < .0001$ for all variables). Further analysis revealed that the most severely anxious participants reported the largest effects. Physiological measures collected during the float session showed signs of a strong relaxation response, including a significant ($p < .0001$) reduction in diastolic blood pressure (on the order of 10 mm Hg) and an increase in normalized high-frequency heart rate variability.
Conclusions: The findings from this initial study need to be replicated in larger controlled trials, but suggest that Floatation-REST may be a promising technique for transiently reducing symptoms in those with anxiety and depression.
Keywords: Anxiety, Anxious Depression, Novel Therapeutics, Non-Pharmacological Therapy
Disclosure: Nothing to Disclose.

T63. Computational Modeling of Effects of Acute Methylphenidate on Component Decision-Making Processes During Fear Extinction and Affective Processing

Jonathon Howlett*, Cédric Hysek, Murray Stein, Martin Paulus

University of California, San Diego School of Medicine, La Jolla, California, United States

Background: A major challenge for psychopharmacology is the discovery of clinically relevant, quantifiable medication targets at the individual level in humans. For example, despite decades of study, the effects of the stimulant methylphenidate (MPH) on complex cognitive and affective decision processes remain poorly understood. The use of computational modeling to decompose these complex processes into quantifiable subcomponents is a promising avenue to elucidate these effects. The present study uses a drift diffusion modeling approach to examine the effects of MPH on decision-making in healthy adult males in different behavioral contexts.
Methods: 18 healthy males participated in two experimental sessions in which either placebo or MPH 40 mg was administered. Each subject received both MPH and placebo in separate sessions in a crossover design, with order of medication and placebo randomized in double-blinded fashion. Subjects performed an attentional bias task (a modified dot probe detection task measuring engagement and disengagement from affective stimuli). As part of the same study, 10 subjects underwent a fear conditioning and extinction procedure and performed a continuous performance task during fear extinction. Trial-by-trial responses and reaction times were used to fit a drift diffusion model based on the fast-dm-30 algorithm, which assumes that decisions are made via a noisy process in which information is accumulated over time until one of two response boundaries is reached, at which time a response is initiated. Drift diffusion modeling of the modified dot probe detection task and the continuous performance task determined for each subject, task and condition (a) a drift rate representing rate of information accumulation, (b) a decision threshold determining the speed-accuracy tradeoff, and (c) a non-decision time representing stimulus registration and motor execution. Model parameters and mean reaction times (RTs) were compared across conditions using paired t-tests.
Results: In the attentional bias task ($n = 18$), there were no significant differences in mean RT ($p = 0.11$), drift rate ($p = 0.30$), decision threshold ($p = 0.80$), or non-decision time ($p = 0.46$) between the MPH and placebo conditions. In the continuous performance task performed during fear extinction ($n = 10$), mean RT was significantly shorter ($p < 0.01$) and drift rate was significantly higher ($p = 0.03$) in the MPH compared to the placebo condition, while there were no significant differences in the decision threshold ($p = 0.99$) and non-decision time ($p = 0.25$).
Conclusions: MPH significantly increased mean RT and rate of information accumulation assessed by computational modeling in a continuous performance task performed during fear extinction. However, MPH did not have a significant effect on mean RT or model parameters in an attentional bias task. These results are consistent with the hypothesis that MPH increases information processing in a fear context but not in an approach/avoidance context. More generally, the effect of methylphenidate on component decision processes depends on behavioral and motivational context. Finally, computational psychopharmacology may be useful in helping to generate process models of how medications affect complex cognitive and affective processes.
Keywords: Computational Psychiatry, Methylphenidate, Anxiety, Mood Disorders
Disclosure: Nothing to Disclose.

T64. Repeated Exposure to Experimental Pain Differentiates Combat TBI With and Without PTSD

Irina Strigo*, Andrea Spadoni, Sabra Inslicht, Alan Simmons

University of California, San Francisco, San Francisco, California, United States

Background: Pain contributes to more severe posttraumatic stress disorder (PTSD), as evidenced by greater disability, more frequent use of harmful opioid analgesics and increased pain severity. Furthermore, PTSD is highly co-morbid with chronic pain and is often a precursor to developing chronic pain conditions. Pain in PTSD is associated with greater utilization of medical services, greater use of psychiatric medication and increased total cost of treatment (Moeller-Bertram et al 2012). This suggests that pain presents a significant burden to those suffering from PTSD, as well as to society. However, the phenomenon of pain perception and its putative dysregulation in PTSD is still unknown – PTSD is a complex disorder that commonly presents with Mild Traumatic Brain Injury(mTBI) and other co-morbid conditions (e.g. depression), that, in turn, are associated with increased pain vulnerability and dysregulated

pain-related brain activation (Strigo et al 2008; 2010; 2013; 2014). This is particularly relevant to Veterans involved in Iraq and Afghanistan conflicts where mTBI is considered a "signature injury" (Hoge et al 2008) Thus the contribution of co-morbid conditions needs to be taken into account while deciphering the mechanism of pain after trauma in a complex population, such as a Veteran sample, who carry significantly elevated risk for pain. We have shown that women with PTSD following intimate partner violence show attenuated brain response to repeated experimental pain that was related to symptoms of avoidance (Strigo et al 2010). The aim of the current study was to: 1) provide further evidence for our hypothesis that brain response to repeated heat pain would be related to avoidance symptoms in a diagnostically complex sample of male veterans, and 2) provide the first examination of the neural correlates of the repeated exposure to experimental pain stimuli in men with combat mTBI with and without PTSD while controlling for depression.

Methods: Seventy male veterans gave written informed consent to participate in this study, which was approved by the University of California San Diego Human Research Protection Program and Veterans Affairs San Diego Healthcare System Research and Development Committee. Of the 70 total subjects, 24 had light to moderate combat exposure with no reported history of mTBI (control group), 20 had moderate combat exposure, reported history of mTBI and did not meet DSM-IV criteria for post-traumatic stress disorder(PTSD)(mTBI-only group), while 26 had moderate combat exposure, reported history of mTBI and also met DSM-IV criteria for PTSD (mTBI+PTSD group). The groups did not differ significantly on age ($F(2,67) = 0.107$, $p = 0.808$), race($2 = 0.7$, $p = 0.990$), or education($F(2,67) = 0.55$, $p = 0.947$). All subjects performed functional Magnetic Resonance Imaging(fMRI) while they experienced painful heat stimuli (9cm2 thermode, Medoc TSA-II, Ramat-Yishai, Israel) to the left forearm. As in our prior study, we examined change in brain activity to repeated heat pain with linear mixed effects modeling for group by administration interaction effects.

Results: All subjects in the brain injury groups (mTBI-only and mTBI+PTSD) self-reported a history of blast-related concussion, the number and severity of which did not significantly differ between the two mTBI groups ($p's > 0.05$). The degree of combat exposure was moderate and was not different between the two mTBI groups. As expected, all PTSD symptoms clusters as measured by CAPS4, i.e., re-experiencing, avoidance and numbing, and hyperarousal, were significantly higher in the co-morbid mTBI+PTSD group compared to the mTBI-only group, consistent with the diagnosis. We observed a significant group by administration interaction to repeated heat pain within insular, frontal and parietal cortices such that the control group showed increased activation over time, while mTBI groups (mTBI-only, mTBI+PTSD) showed decreased activation within bilateral anterior insulas(AI) between administrations. Importantly, change in the right AI response was inversely correlated with avoidance symptoms, but only in those with co-morbid mTBI+PTSD. Furthermore, in the comorbid group greater AI attenuation was associated with decreased connectivity with anterior cingulate(ACC).

Conclusions: The current study provides evidence for the hypothesis that combat trauma with mTBI and PTSD shows a complex relationship with neural response to acute experimental pain whereby repeated exposure to brief painful stimuli results in attenuation of insula activation over time. Importantly, we found that although both mTBI groups showed attenuation of insula activation over time, symptoms of avoidance significantly predicted insula attenuation only in those subjects with combat-related mTBI and PTSD. In combination with the existing literature, it appears this relationship bridges across gender, type of trauma, and paradigmatic task differences (Strigo et al 2010b) potentially suggesting a generalized mechanism of maladaptive response to experimental pain in those who develop PTSD following traumatic experiences. Importantly, these findings, in combination with our prior work in which we showed that brain injury without significant psychological symptoms disrupts endogenous pain modulation (Strigo et al 2014), suggests dysfunction within segregated neural systems underlying pain response and modulation in those suffering from comorbid mTBI and PTSD.

Keywords: Concussion, Combat Veteran, BOLD Imaging, Pain Sensitivity, Trauma

Disclosure: Nothing to Disclose.

T65. Amygdala Hypoactivation to Infant but not Adult Faces in New Mothers With a History of Trauma

Aviva Olsavsky*, Andrew Erhart, Rebekah Tribble, Pilyoung Kim

University of Colorado Denver, Aurora, Colorado, United States

Background: Face emotion processing in new mothers is inextricably linked to mother-infant social communication, which is critical for development. History of trauma is associated with altered parenting behaviors and style, making maternal trauma an important area to study to understand factors affecting parenting. Trauma history has been related to risk for insecure attachment and HPA-axis dysfunction in mothers and infants, more predominantly negative attributional style toward the child, risk for child abuse, and Post-Traumatic Stress Disorder vulnerability in youth. Neuroimaging studies in mothers with trauma are limited compared to studies regarding postpartum depression, but evidence suggests increased amygdala reactivity to threat and diminished reactivity to more positive-valence infant social stimuli (e.g. infant videos). Additional data suggests that amygdala activation is associated with affective salience, particularly in the context of mother-child relationships. We hypothesized that mothers who have experienced trauma would have blunted amygdala reactivity to infant faces. Furthermore, the evidence that trauma exposure may disrupt processing of social stimuli and the increased relevance of infant stimuli for women in late pregnancy and the postpartum period led us to hypothesize this group-difference would be present only for infant stimuli. Thus, we hypothesized that we would not see a between-group difference in adult face processing. We further postulated that blunted amygdala reactivity to infant faces in new mothers would be associated with decreased sensitivity to their infants during a standardized dyadic observation.

Methods: Participants included new mothers who performed face emotion processing functional MRI (fMRI) tasks: an infant face task ($N = 47$; Healthy Comparisons (HC) = 29, Trauma = 18) and an adult face task ($N = 46$; HC = 29, Trauma = 17). Subjects were classified with respect to interpersonal trauma history (e.g. - physical or sexual abuse/assault, verbal abuse by parent, witnessed domestic violence) utilizing three validated questionnaires (CRISYS, Risky Families questionnaire, and Life Events Checklist). Demographic and maternal mood variables (Spielberger Trait/State Anxiety Inventory (STAI), Beck Depression Inventory (BDI), socioeconomic status, age) were collected, and between-group differences were evaluated to assess for confounders. Infant and adult fMRI tasks were event-related designs including standardized face emotion photographs (infant - sad, neutral, happy; adult - angry, fearful, neutral, happy). Images were preprocessed and analyzed in AFNI. Beta-weights from bilateral anatomical amygdala ROIs were extracted and analyzed further in SPSS with repeated-measures ANOVA. Additionally, a standardized behavioral observation was conducted to assess mother-infant interaction during a postnatal home visit, coded using the Emotional Availability Scale (EAS), which is validated in similar populations.
Results: In anatomical ROI analyses, there was a main effect of group, driven by blunted left amygdala response to infant faces in mothers with a trauma history vs. HC ($F1,45 = 9.33$, $p < 0.005$), which was not present in the adult faces task ($F1,44 = 0.01$, $p > 0.05$, no significant main effects/interactions). There was no main effect of emotion or interaction between emotion and group in the infant faces task. Post hoc analyses of BDI, STAI, maternal socioeconomic status, and maternal age did not reveal between-group differences. There was no statistically significant effect in right amygdala in either task (ps > 0.05). The EAS Direct Sensitivity subscale was lower in women with history of trauma vs. HC ($F = 6.28$, $p < 0.05$). Furthermore, sensitivity rating was positively correlated with left amygdala activation in the trauma group ($r = 0.512$, $p < 0.05$, $N = 18$).
Conclusions: Our preliminary data reveals that mothers who experienced trauma exhibit blunted amygdala activation to infant face stimuli during the postnatal period, and that they demonstrated lower maternal sensitivity during a home behavioral assessment. Furthermore, in women with a history of trauma, higher maternal sensitivity was associated with higher amygdala activation to infant faces across emotions. By contrast, this correlation did not hold either across both groups, or within controls. Given the strikingly different neural activity between groups, as well as the differences in maternal sensitivity, it may be that the control group represents a more narrow range of both amygdala activation and maternal sensitivity, while in the trauma group, marginal changes in amygdala reactivity to infant stimuli are associated with accompanying improvements in sensitivity. This finding has important implications regarding differences in maternal parenting behaviors, which have been shown to influence long-term infant outcomes. Further studies might include connectivity analyses and assessing the role of neuroendocrine function, including cortisol and oxytocin levels in women with trauma, and their relationship with processing of infant face stimuli.

Keywords: Peripartum, Maternal Sensitivity, Face Emotion Processing, Trauma Exposure
Disclosure: Nothing to Disclose.

T66. DTI-Identified Cerebral Microstructural Changes in Mice Overexpressing CRF in the Forebrain

Jessica Deslauriers*, Mate Toth, Miriam Scadeng, Robert Rissman, Victoria Risbrough, Gregory Brown

University of California, San Diego, La Jolla, California, United States

Background: Corticotropin releasing factor (CRF) regulates endocrine and behavioral responses to stress. Stress exposure in animals induces long-term changes in the CRF system and in hippocampal function. Excess CRF signaling in the hippocampus has been linked to alterations in hippocampal function, spine formation and related cognitive functions. In humans, CRF levels are increased in the cerebrospinal fluid of individuals with childhood trauma history and some mood and anxiety disorders. Mood and anxiety disorders are also characterized by low hippocampal volume and function. By using diffusion tensor imaging (DTI), a methodology translatable across species, we tested the hypothesis that forebrain-specific CRF overexpression induces changes in brain microstructure, specifically in the dorsal hippocampus.
Methods: Male and female mice with inducible forebrain-specific CRF overexpression were generated by crossing two genetically modified mouse lines carrying a CaMKIIα promoter-driven rtTA2 transgene and a doxycycline-regulated tetO promoter fused to the CRF gene to produce double mutant mice. CRF overexpression was induced with doxycycline (DOX) administration in mouse chow at a dose of 3.5 mg/g body weight/day from postnatal day ~90 -120, after which DOX was removed. Since DOX alone did not affect behavioral outcomes in wild-type animals, double mutant littermates without DOX treatment were used as control subjects. In vivo DTI was performed at three time points in each animal: baseline (before DOX treatment), ~3 weeks of DOX treatment, and again ~3 months after DOX treatment was ended. Histology showed significant increases in CRF protein levels in hippocampus 3 weeks after start of treatment, which returned to normal 3 months after treatment had ended. DTI parameters fractional anisotropy (FA) and mean diffusivity (MD) were assessed in dorsal hippocampus.
Results: In male mice, MD increased with time in the dorsal hippocampus of control mice, with significant higher MD 3 months after the first scan. In mice overexpressing CRF in the forebrain, no change in MD was found across time in the same brain region. These effects were not observed in females. Also, no significant effect of CRF-overexpression or time was found on FA. Previous studies have shown that this regimen of CRF-overexpression results in poor hippocampal function as measured by reduced contextual fear learning and novel object place recognition.
Conclusions: Surprisingly, CRF overexpression appeared to block aging-related elevations in MD, suggesting some potential modulatory effects of CRF on hippocampal microstructure. Previous studies implicate CRF both as a potential protective factor and a driver of tau-pathology in

aging-related neurodegeneration. Future studies are required to delineate the mechanism of aging-induced MD and the contribution of CRF in this process.
Keywords: Corticotropin-Releasing Factor (CRF), Diffusion Tensor Imaging, Hippocampus
Disclosure: Nothing to Disclose.

T67. International Collaborative OCD Network (iCON)

Dorothy Grice*, Joseph Buxbaum, Mark Daly, Silvia DeRubeis, Bernie Devlin, Stephen Hyman, Benjamin Neale, Avi Reichenberg, Kathryn Roeder, Stephan Sanders, Sven Sandin

Icahn School of Medicine at Mount Sinai, New York, New York, United States

Background: Obsessive-compulsive disorder (OCD) can be severely impairing and a better understanding of the etiological factors for OCD will enhance the development of more effective interventions. Heritability for OCD is estimated at between 40-80% providing a path to gene discovery as a means of understanding etiology. Available GWAS data for OCD is still modest in size when considered from either the standpoint of necessary sample to identify common variation of small effect, or when compared with other successful GWAS studies in psychiatry (e.g., schizophrenia). Moreover, high-throughput sequencing studies in OCD are in their infancy. We have therefore created the International Collaborative OCD Network (iCON) to enhance etiological discovery in OCD, beginning with study of the genomic architecture.
Methods: iCON will ascertain cohorts of well-phenotyped OCD cases (n > 1,750/year), ideally with information on comorbid disorders and traits. All cases (and additional controls where necessary) will be genotyped. In addition, we will carry out whole-exome sequencing (WES) on a subset of cases (n ~ 1000 in year 1) for which control data exists or can be inferred. With these data, and data from other studies, we will carry out multiple analyses. We will: carry out GWAS for OCD; study CNV in OCD; assess the role of de novo and recent variation in OCD; develop polygenic risk scores for OCD and assess them in relation to traits in the population; and, estimate shared risk with other neuropsychiatric disorders (e.g., Tourette disorder, autism spectrum disorder (ASD), ADHD, etc.). These approaches would also address the degree to which shared risk across neuropsychiatric disorders is due to comorbid diagnoses as opposed to true pleiotropy and model risk as a function of common SNPs, rare standing variation including CNV, and recent mutations.
Results: To date, we have coordinated with other groups including the Psychiatric Genomics Consortium and the Genomic Psychiatry Cohort to maximize efficiency and to improve precision and statistical power. In addition, we have identified several existing OCD cohorts and begun sample collection in Sweden [EGOS (Epidemiology and Genetics of OCD in Sweden), http://ki.se/en/meb/the-egos-study and http://ki.se/meb/egos-studien]. In EGOS, ascertainment is performed through Swedish national health registers (through which we can also obtain comorbid psychiatric diagnoses) using a validated strategy. We have identified ~ 12,000 cases in the registers. To simplify ascertainment and reduce the time and resources, we adopted a strategy where a first e-mail contact is followed by a call and then a visit by research nurses who collect saliva or blood from prospective participants. In addition, we deployed two assessments (FOCI and OCI-R) online to obtain additional clinical information (and to further validate diagnoses). At present, we have collected DNA from over 500 cases and anticipate collecting > 1,500 cases from Sweden. Genotyping is ongoing: 1,100 pediatric controls have been completed, ~ 1,000 OCD samples will be genotyped within the next three months and a similar number queued for WES. Additionally, in Sweden, ~ 10,000 control, ~ 1,100 ASD, ~ 5,000 schizophrenia, and ~ 5,000 bipolar disorder samples are already genotyped.
We previously developed a novel approach (PMID 25038753) that successfully defined the allelic spectrum of ASD in Sweden. We used Treelet Covariance Smoothing (PMID 24587841) to improve SNP-based estimates of pairwise kinship and, including relatives (albeit distant), refine the estimates of total heritability (which now can include heritability due to rare inherited variants). Using SNP genotypic data, we estimated that heritability due to common variation (~50%) greatly exceeded that of rare inherited variants (~3%) in this sample. Common variant heritability also greatly exceeded literature-derived estimates of heritability due to non-additive variants (~4%, e.g., rare recessives) or de novo variants (~3%). As the EGOS Swedish OCD collection is also being ascertained using a national framework, we are poised to carry out similar analyses in OCD. Furthermore, we have developed and applied tools to assess and identify rare variation (PMID 23966865 25363760).
Conclusions: In collaboration with other critical consortia, we are ascertaining, genotyping and sequencing international OCD samples. All data will be shared with contributing sites and consortia, including the PGC. We will continue to develop novel methods to continue to define the genomic architecture of OCD. As iCON expands, combining iCON data with other consortia data will lead to the identification of specific genetic risk for OCD. iCON is actively seeking collaborations to enhance gene discovery in OCD. iCON is supported by a grant from the Stanley Center.
Keywords: Obsessive-Compulsive Disorder (OCD), Genetic Architecture, Heritability
Disclosure: Nothing to Disclose.

T68. Modulating Expression of Conditioned Suppression Responses in Rats Using the Endocannabinoid Reuptake Inhibitor URB597

Laurence P. Simmons, Mahder T. Etuma, Elizabeth M. Bergman, Nicole Moore*

Walter Reed Army Institute of Research, Silver Spring, Maryland, United States

Background: The endocannabinoid system is involved in associative memory processes underlying aversive conditioning. This role suggests that compounds influencing

cannabinergic signaling may have value as pharmacological tools to remediate adverse behavioral responses related to aversive memory. To better understand the role and timing of endocannabinoid tone in threat-associated learning and memory, we used a rodent model of aversive conditioning and conditioned suppression of appetitively-motivated operant behavior. Using URB597 as a reuptake inhibitor, we characterized the effects of enhanced endocannabinoid signaling upon aversive responses at the time of conditioning or at the time of the conditioned response.

Methods: Adult male Sprague Dawley rats (75 days of age and older) were trained and then maintained on a variable interval (32 seconds average) schedule of operant responding. After several weeks and stable responses across days, rats were conditioned to associate a light-and-tone pairing to an aversive footshock. In the time of conditioning group, URB597 (0.1 or 0.3 mg/kg i.p.) was administered 30 minutes before the session. After returning to a stable baseline of operant responding, conditioned suppression was measured. At a random time during the operant session, the light-and-tone cue was presented, and reduced rate of lever pressing was recorded. In the time of conditioned response group, URB597 (0.1 or 0.3 mg/kg) was administered 30 min prior to the start of the session. Measures collected for all groups were time to first lever press and suppression index, a measure of depression of lever-pressing rate after cue presentation.

Results: Early observations show that timing of drug administration affects outcomes, consistent with previous studies of cannabinergic drugs. Learning curves over the five days of extinction differ by time of administration. Time to first press may be a more sensitive measure with this compound compared to the overall suppression of lever-pressing rate.

Conclusions: Determining the optimal timing of administration of a potentially therapeutic cannabinoid compound, relative to experimental events, is an important consideration when evaluating efficacy.

Supported by the Military Operational Medicine Research Program, US Army Medical Research and Materiel Command.

Material has been reviewed by the Walter Reed Army Institute of Research. There is no objection to its presentation and/or publication. The opinions or assertions contained herein are the private views of the author, and are not to be construed as official, or as reflecting true views of the Department of the Army or the Department of Defense. Research was conducted in compliance with the Animal Welfare Act and other Federal statutes and regulations relating to animals and experiments involving animals and adheres to principles stated in the Guide for the Care and Use of Laboratory Animals, NRC Publication, 2011 edition. All procedures were reviewed and approved by the WRAIR Institutional Animal Care and Use Committee, and performed in facilities accredited by the Association for Assessment and Accreditation of Laboratory Animal Care, International.

Keywords: Rat Models, Aversive Conditioning, Endocannabinoid

Disclosure: Nothing to Disclose.

T69. Impact of Gender and Anxiety on Exploratory Activity as Measured by a Human Behavioral Pattern Monitor

Robin Aupperle*, Akalvizhy Elanko, Arpi Minassian, Kathryn Tombridge, Kelly Cosgrove, Elisabeth Akeman, Martin Paulus

Laureate Institute for Brain Research, Tulsa, Oklahoma, United States

Background: Translation of findings from animal experiments to humans is of central importance for the field of behavioral neuroscience. However, the value of translational research has been challenged by the fact that pre-clinical animal findings often do not generalize to human clinical outcome trials. The development and use of translational behavioral tasks that are similar across animal and human research would support a more fluid exchange of scientific ideas, in turn allowing for the development of optimized treatment strategies (Kirlic et al., 2017). Decreases in rodent exploratory behavior on open field tests (such as the behavioral pattern monitor) is often interpreted as a model of anxiety (i.e., Prut & Belzung, 2003). A translational human behavioral pattern monitor (hBPM) has been developed by previous researchers (Perry et al, 2009) to examine motor movement and exploratory activity. The hBPM has been used to identify distinct patterns of motor movement and exploratory activity in patients with bipolar disorder or schizophrenia (Minassian et al., 2010). Here, we describe the use of a modified version of the hBPM to identify whether anxiety results in attenuated exploration of a novel environment.

Methods: As part of an ongoing study, 79 participants (56 females) with varying levels of anxiety and depression completed the hBPM (15 with major depressive disorder only; 8 with anxiety disorders only; 42 with comorbid anxiety and depressive disorders; 14 healthy control). NIH PROMIS® measures were used to estimate severity of anxiety and depression symptoms. The Mini International Neuropsychiatric Inventory was used to diagnose DSM IV/5 psychiatric disorders. The hBPM was set up in a 14.5'x12' room, designed to appear like a child development room with 11 exploratory objects (i.e., toy snake, ball, bobo doll, etc.) dispersed throughout. For the hBPM, participants are fitted with a physiological sensor vest and directed to stay in the room for 15 minutes under the pretense that researchers are obtaining baseline physiological measures. Participants are recorded digitally by a concealed camera. Custom Matlab scripts were used for detection of x-y-coordinates of each participant throughout the task. Amount of movement was quantified as distance traveled during the entire period. Spatial d was calculated as a measure of the degree to which individuals stay in a particular area and explore this area (high spatial d) versus moving from one area to another (low spatial d) as a function of behavioral activity. Number of exploratory interactions was coded manually during visual review of video recordings. Linear mixed models were used to examine main and interaction effects of gender and symptom severity, with age entered as a covariate.

Results: Distance traveled ($r = .70$, $p < .001$) and spatial d ($r = -.66$, $p < .001$) were positively correlated with number of

hPBM object interactions. A relationship was observed between age and total number of interactions ($r = -0.33$, $p = 0.015$), and thus was included as a covariate in the remaining analyses. Linear mixed model results indicated main effects of both gender and anxiety, as well as gender by anxiety interactions on all hBPM variables of interest. The interactions were characterized by anxiety relating to reduced object interactions ($F(1,49) = 7.43$, $p = .009$) and distance traveled ($F(1, 74) = 6.46$, $p = .013$), and higher spatial d ($F(1,72) = 5.40$, $p = .023$) for females, while the opposite relationships were observed for males. The degree of depressive symptoms was not related to measures of exploratory behavior or motor behavior in the hBPM.

Conclusions: The modified version of the hBPM developed in this study was successful in motivating exploratory interactions with novel objects and as expected, automatic tracking of participant movement in the room correlated with the number of object interactions. The gender effects identified in the current study add to an inconsistent literature regarding potential cross-species effects of gender on anxiety-related behaviors (i.e., Aupperle et al., 2011; Basso et al., 2011; Johnston & File, 1991). Such findings may partially underscore the higher prevalence of anxiety disorders for females (Baxter et al., 2013). Further, results from this study suggest that the behavioral manifestations of anxiety may be unique for different sub-groups of patients. Namely, while females may often respond to anxiety with inhibited and avoidant behavior, males may be more likely to respond with increased agitation or approach-oriented behavior. These findings indicate that the behavioral targets for anxiety-related interventions may need to be personalized for the population or individual. In summary, the hBPM offers a realistic, objective assessment to understand behavioral manifestations of anxiety and thus, may be useful in predicting and/or tracking response to treatment.

Keywords: Translational Models, Anxiety, Depression, Exploration

Disclosure: Nothing to Disclose.

T70. Cannabinoid Facilitation of Fear Extinction in Posttraumatic Stress Disorder

Christine Rabinak*, Craig Peters, Farrah Elrahal, Mohammed Milad, Sheila Rauch, K. Luan Phan, Mark Greenwald

Wayne State University, Detroit, Michigan, United States

Background: Exaggerated fear responses are the hallmark of posttraumatic stress disorder (PTSD). Empirically-supported psychotherapy for PTSD, Prolonged Exposure (PE), involves repeated exposure to fear-linked cues to produce "extinction" of fear. PE is generally effective, but many patients exhibit incomplete extinction or fail to sustain extinction learning-related improvements over time. Recall of extinction learning depends on limbic-frontal brain networks (hippocampus [HPC], ventromedial prefrontal cortex [vmPFC]) and PTSD patients show decreased activity in these regions and poor extinction recall. Adjunct interventions that address vmPFC-HPC dysfunction and rescue extinction recall deficits could enhance the efficacy of PE for PTSD. Our prior work suggests that an acute oral dose of Δ9-tetrahydrocannabinol (THC), prior to experimental fear extinction procedures in healthy volunteers, facilitates recall of extinction learning by increasing activation and functional connectivity of vmPFC-HPC. As extinction recall deficits and vmPFC-HPC dysfunction have been observed in PTSD, our preliminary findings indicate the cannabinoid system is a promising target to improve the efficacy and durability of learning during PE in treating PTSD (e.g., shortening treatment while strengthening and prolonging gains). However, direct tests of cannabinoid effects on recall of extinction learning and associated neural circuits have not yet been conducted with PTSD patients.

Methods: This ongoing double-blind, placebo-controlled, randomized-group study uses a Pavlovian fear learning paradigm to examine the effect of THC (7.5mg) vs. placebo (PBO), administered prior to extinction learning, on brain activation (functional magnetic resonance imaging [fMRI]) and skin conductance (SCR) responses in 49 trauma-exposed adult volunteers with ($n = 20$; PTSD) and without ($n = 29$; trauma-exposed controls; TEC) PTSD and 20 non-exposed healthy controls (HCs), testing extinction recall 24 hours after extinction learning.

Results: Preliminary data suggest that acute THC (vs. PBO) administration prior to extinction learning in PTSD patients improves recall of extinction learning. In particular, PTSD patients who received PBO during fear extinction exhibited, as expected, poor extinction recall as evidenced by increased peripheral measures of fear (SCR and US expectancy ratings) to a conditioned stimulus (CS) that was previously extinguished (CS+E). In contrast, PTSD patients who received THC during fear extinction exhibited good extinction recall (significantly lower peripheral measures of fear, compared to PBO) and increased HPC activation to the CS +E during recall of extinction learning. There was no drug effect on extinction recall in either control group (TEC, HC).

Conclusions: These findings provide the first evidence that pharmacological enhancement of recall of extinction learning is feasible in PTSD patients using cannabinoid system modulators. Ultimately, the cannabinoid system may serve as a promising target for innovative intervention strategies in PTSD and other fear learning-related disorders.

Keywords: Cannabinoid, Fear Extinction, Posttraumatic Stress Disorder

Disclosure: Nothing to Disclose.

T71. Comparison of Emotional Cognition Assessed With Fear Conditioning by Interpersonal Conflicts in Patients With Dysthymia, Schizophrenia and Healthy Controls

Hideaki Tani*, Mitsuhiro Tada, Takefumi Suzuki, Masaru Mimura, Hiroyuki Uchida

Keio University School of Medicine, Tokyo, Japan

Background: While emotional cognition has been implicated in various psychiatric illnesses, data are still scarce about how common or variable it is across the diagnoses. We recently developed an interpersonal stimulus for the fear conditioning paradigm to represent real-world social conflict, which has not been adequately taken into account in conventional physically aversive stimulus such as sound. This interpersonal stimulus has successfully been applied to healthy

individuals. The objective of this study was to compare emotional cognition of patients with dysthymia or schizophrenia compared to that of healthy individuals by measuring autonomic response using skin conductance response (SCR) to the interpersonal stimuli.

Methods: Twenty patients with dysthymia (ICD-10) and 20 patients with schizophrenia (ICD-10) and 20 healthy subjects underwent fear conditioning and extinction experiments in response to two types of stimuli: an aversive sound, and a picture of an actor's face with recorded unpleasant verbal messages to generate interpersonal conflicts. All participants were female and the mean ± S.D. age was 28.9 ± 7.2 years. The paradigm consisted of three consecutive phases: habituation, acquisition, and extinction. In the acquisition phase, one conditioned stimulus (CS) was randomly selected from three CSs and the selected CS was then paired with the unconditioned stimulus (US) at a partial reinforcement rate of 50% (i.e. CS+). Other two CSs were presented without pairing the US during the acquisition phase (i.e. CS-). Conditioned response was quantified by differential SCR between CS+ and CS- using paired t-tests during the acquisition as well as early and late extinction phases. Regression analysis was performed to examine the effects of emotion regulation strategy and prescribed medications on the differential SCR.

Results: Dysthymic patients successfully showed fear conditioning in both sound and interpersonal conditions. The mean changes in the SCR amplitudes in response to CS+ were greater than those in response to CS- during the acquisition phase (CS+ = 0.55 and CS- = 0.23 for the aversive sound [$t(19) = 4.03$, $p < .001$]; and CS+ = 0.51 and CS- = 0.23 for the interpersonal stimulus [$t(19) = 5.22$, $p < .001$]). Subsequently, the dysthymic subjects successfully extinguished conditioned fear responses in the sound condition (CS+ = 0.22, CS- = 0.15 [$t(19) = 1.67$, $p = .11$]). In the interpersonal condition, however, those dysthymic patients failed to extinguish the conditioned response during the last three extinction trials (CS+ = 0.23, CS- = 0.12 [$t(19) = 2.85$, $p = .01$]). Four outlier subjects with schizophrenia who did not demonstrate evaluable SCRs ($> 0.03\mu s$) to the CS+ during the acquisition phase to either of two stimuli were excluded from the analysis. The subjects with schizophrenia did not show a fear conditioning in the interpersonal condition (CS+ = 0.56 and CS- = 0.64 [($t(14) = -1.25$, $p = .23$]) or the sound condition (CS+ = 0.37 and CS- = 0.35 [($t(15) = 0.22$, $p = .83$]). No differences in SCRs were observed between CS+ and CS- in extinction phases in both conditions. The healthy subjects successfully showed fear conditioning in both sound and interpersonal conditions (CS+ = 0.56 and CS- = 0.18 for the aversive sound [$t(19) = 7.81$, $p < .001$]; and CS+ = 0.71 and CS- = 0.44 for the interpersonal stimulus [$t(19) = 3.90$, $p = .001$]). Subsequently, the healthy subjects successfully extinguished conditioned fear responses in both conditions (CS+ = 0.09, CS- = 0.08 for the aversive sound [$t(19) = 0.43$, $p = .68$]; and CS+ = 0.16 and CS- = 0.13 for the interpersonal stimulus [$t(19) = 1.42$, $p = .17$]). The differential SCR during the early extinction phase in the interpersonal condition was significantly greater in dysthymic patients than the healthy subjects (0.14 vs 0.05, $p = .035$). Analysis of variance found that a significant difference in differential SCR during acquisition phase in interpersonal stimuli among the three groups ($F(2,53) = 9.71$, $p < .001$); differential SCR was smaller in the patients with schizophrenia than healthy subjects or dysthymic patients. Among the dysthymic subjects in the interpersonal condition, the Emotion Regulation Questionnaire (ERQ) expressive suppression score was positively associated with the differential SCR during the early extinction phase ($\beta = .94$, $p = .01$), and the ERQ cognitive reappraisal score was negatively associated with the differential SCR during the early extinction phase ($\beta = -.88$, $p = .04$). Moreover, the use of antidepressant drugs was negatively associated with the differential SCR during the late extinction phase ($\beta = -.71$, $p = .02$).

Conclusions: Female subjects with schizophrenia failed to get conditioned with the interpersonal stimuli or the aversive sound, which suggests intrinsic impairments in the emotional processing. A greater difficulty of extinction in the interpersonal condition observed in the dysthymic subjects points to a possibility that their unstable interpersonal relationship is derived from aberrant extinction process of interpersonal negative experiences. Emotional regulation strategy and use of antidepressant drugs were found to be associated with successful extinction of conditioned fear by interpersonal conflict in dysthymic patients, which may imply that antidepressant treatment and emotional self-control attainable through cognitive behavioral therapy may serve, at least to some extent, to alleviate personal negative conflicts in this patient group.

Keywords: Emotional Processing, Fear Conditioning, Skin Conductance Responses, Interpersonal Conflict

Disclosure: Part 4: Eli Lilly, Grant.

T72. Dissociation Between Intrinsic Amygdala and BNST Function in Post-Traumatic Stress Disorder

Jennifer Blackford*, Alec Seidenberg, Meagan Buford, Hannah Gardner, Bunmi Olatunji

Vanderbilt University Medical Center, Nashville, Tennessee, United States

Background: Post-traumatic stress disorder (PTSD) affects more than 30% of Veterans returning from Afghanistan and Iraq. Unfortunately, PTSD is often resistant to current therapeutic interventions and a complete remission is uncommon. Current treatments, such as SSRIs and exposure therapy, are thought to work by normalizing function in the amygdala. One possibility for the lack of widespread treatment success is that the neurobiology of PTSD is complex and extends to alterations in other brain regions. Animal models of fear and anxiety suggest that a second brain region–the bed nucleus of the stria terminalis–is a crucial hub for anxiety. Some studies suggest that the amygdala and BNST play different roles in fear and anxiety, evidenced by a dissociation in the amygdala vs BNST response to short-term fear stimuli vs ambiguous or unpredictable anxiety stimuli. To date, most human neuroimaging studies of PTSD have examined only the amygdala-mediated fear neurocircuitry. Here, we test for a difference between intrinsic function in the amygdala and BNST in combat Veterans relative to combat controls and healthy controls.

Methods: Forty-seven medication-free adults (14 Combat Veterans with PTSD, 20 Combat Veterans without PTSD, and 13 Healthy Controls) completed a resting state MRI scan. Amplitude of Low Frequency Fluctuations (ALFF) and fALFF (fractional ALFF) were computed for the amygdala and BNST regions of interest. Repeated measures ANOVAs tested for an amygdala-BNST difference. Exploratory whole-brain analyses were also performed.
Results: Intrinsic amygdala and BNST response differed significantly across the three groups (ALFF: brain region x group interaction, $p = .04$, FALFF: brain region x group interaction, $p = .03$). For both ALFF and FALFF, the combat PTSD group had the smallest amygdala-BNST difference, reflecting relatively similar amygdala and BNST signal. In contrast both control groups had higher BNST function relative to amygdala function. Whole brain analyses revealed increased activation in PTSD in multiple regions involved in fear and anxiety including amygdala, hippocampus, insula, caudate, hypothalamus, and orbitofrontal cortex.
Conclusions: Both combat and healthy controls showed a dissociation between amygdala and BNST function at rest. Importantly, combat Veterans with PTSD failed to this same pattern of dissociation and instead had similarly elevated levels of amygdala and BNST function. This finding highlights the importance of examining multiple components of the fear and anxiety circuits and suggest a role for the BNST in understanding the neurobiology of PTSD.
Keywords: PTSD, Amygdala, BNST, Anxiety
Disclosure: Nothing to Disclose.

T73. Effects of Respiratory-Gated Auricular Vagal Nerve Stimulation on Central Autonomic Regulation and Mood Symptomatology in Major Depression

Ronald Garcia*, Aileen Gabriel, Arielle Stanford, Harlyn Aizley, Riccardo Barbieri, David Gitlin, Vitaly Napadow, Jill Goldstein

Brigham & Women's Hospital, Boston, Massachusetts, United States

Background: Patients with major depressive disorder (MDD) and decreased vagal tone are at increased risk for developing cardiovascular disease (CVD). Conversely, patients with CVD are more likely to have poor outcomes if they also have MDD. Thus, treatments that address comorbid illness will have a significant impact on public health. Recently, transcutaneous auricular vagus nerve stimulation (tVNS) has emerged as a well-tolerated and minimal risk alternative to implanted VNS, and experimental studies have shown promising antidepressant effects and modulatory actions on cardiac autonomic physiology. The mechanisms of action and neural pathways mediating these effects are still unclear. However, neuroimaging and physiological studies have suggested that tVNS effects are mediated by modulation of nucleus tractus solitarii (NTS), from which projections synapse with brain regions involved in mood and autonomic regulation [central autonomic network (CAN)]. Further, as NTS operates in response to changes in cardiopulmonary function (i.e. receiving inhibitory influences from ventral respiratory group medullary neurons during inhalation and firing facilitation during exhalation), we proposed that gating vagal afferent stimulation to exhalation will yield a novel neuromodulation approach that may effectively optimize tVNS effects on reduction of depressive symptomatology and modulation of cardiovascular physiology in patients with MDD.
Methods: Eighteen women (29.7 ± 4.2 years) with recurrent MDD, unmedicated and in an active episode were scanned twice within one week to assess the effects of tVNS gated to exhalation vs inhalation in the modulation of CAN, depressive symptomatology and peripheral autonomic function. Electrodes were placed in the cymba concha of the left ear (vagal-innervated ear regions) and electrical stimuli were administered at a frequency of 30Hz, duration of 0.5s, pulse width of 450 μS, and current intensity set to achieve moderate (but not painful) sensation. Functional MRI (fMRI) data were acquired on a Siemens MAGNETOM Skyra 3T MRI scanner with a 32-channel head coil (TR/TE = 1250 ms/33 ms, slice thickness: 2 mm). Subjects underwent a mild visual stress challenge task comprising presentation of blocks of negative valence/high arousal, neutral valence/low arousal, and fixation images adapted from the International Affective Picture System. The stress challenge preceded and followed a 20-minute session of respiratory-gated tVNS. Brain response to the stress challenge collected in pre- and post-tVNS runs were calculated in a first level GLM analysis (FEAT, FSL) using a regressor for negative valence/high arousal images blocks convolved with the canonical hemodynamic response function (Double-Gamma). The results of these analyses (ie, parameter estimate and variance) were transformed into standard space (MNI152), and submitted to group analyses (Randomise, FSL) using permutation-based nonparametric tests (5000 permutations) to evaluate the effects of tVNS on modulation of brain activity (2-sample paired t-test: post-tVNS vs pre-tVNS). Significant voxels resulting from this analysis were used to calculate mean percent signal changes and statistical differences between exhalatory-gated and inhalatory-gated stimulation. An adaptive point-process algorithm was applied to electrocardiogram recordings to compute variations in the High Frequency component of heart rate variability (HF-HRV, 0.15 to 0.4 Hz), an indicator of parasympathetic cardiac tone. In addition, a Beck Depression Inventory (BDI) was administered before and after each fMRI session. A paired Student's t-test was used to evaluate significant group differences in HF-HRV and BDI changes after stimulation (exhalatory-gated vs inhalatory-gated tVNS).
Results: Depressed women showed a significant reduction in depressive symptomatology following exhalatory-gated tVNS compared to baseline measurements (20.6 ± 8.2 vs 28.3 ± 7.68, $p < 0.01$), whereas this difference was not observed after inhalatory-gated stimulation (24.3 ± 8.7 vs 27.2 ± 7.5, $p = 0.3$). Mean decrease in BDI scores after the exhalatory-gated sessions was significantly greater compared to inhalatory-gated tVNS (-7.65 ± 6.4 vs -2.88 ± 4.4, $p = 0.01$). Exhalatory-gated stimulation was also associated with significant activation of NTS, anterior insula, thalamus, orbital prefrontal cortex, and anterior and mid-cingulate cortex when compared to inhalatory-gated stimulation. In addition, a significant increase in cardiac vagal output was observed following exhalatory-gated stimulation compared

to inhalatory-gated tVNS (HF-HRV % change = 46.3 ± 33.8% vs 22.1 ± 30.7%, $p = 0.03$).

Conclusions: Our results indicate that exhalatory-gated tVNS effectively modulates shared brain circuitry implicated in mood and autonomic dysregulation in MDD. Furthermore, our study shows that this physiologically-enhanced neuromodulation technique is associated with significant acute antidepressant effects and modulatory actions on cardiac parasympathetic tone of depressed subjects. Future studies should evaluate the longitudinal effects of exhalatory-gated tVNS, linking stimulus-evoked brain responses and antidepressant and vagal modulatory effects with long-term clinical outcomes in patients with comorbid MDD and CVD.

Keywords: Major Depression Disorder, Transcutaneous Auricular Vagus Nerve Stimulation (taVNS), Respiration, Central Autonomic Network, Cardiac Autonomic Tone

Disclosure: Nothing to Disclose.

T74. Lisdexamfetamine Dimesylate in the Treatment of Adult ADHD With Anxiety Disorder and Depression Comorbidity: Results From a Clinical Trial

Michael Van Ameringen*, William Simpson, Beth Patterson, Jasmine Turna, Katrina Pullia

McMaster University, Hamilton, Canada

Background: Adult ADHD is highly comorbid with mood and anxiety disorders, with 85% of patients having at least one psychiatric comorbidity, and 60% having at least two. Although it has been hypothesized that anxiety may be a feature that is closely tied to the pathogenesis of the ADHD, it has also been proposed that ADHD with comorbid anxiety may represent a subtype of ADHD or that patients with ADHD and comorbid anxiety are phenotypically different from those with the pure disorder. The presence of anxiety comorbidity in adult ADHD has been associated with additive clinical effects, leading to more global impairment, poorer outcome, greater resistance to treatment and increased costs of illness. ADHD with comorbid anxiety disorders has also been associated with greater degrees of inattention. Stimulant medications are the first-line agents in the treatment of adult and childhood ADHD. Few studies have examined the efficacy of ADHD treatments within these comorbid populations, as the bulk of the pharmacotherapy literature has examined the pure disorder. Only two randomized controlled trials have specifically examined adult ADHD with comorbidity. Adler and colleagues (2009) examined the use of atomoxetine in adult patients with ADHD and comorbid social anxiety disorder(SAD) and found that patients treated with atomoxetine had significantly higher reductions in both ADHD ($p < 0.001$) and social anxiety symptoms ($p < 0.001$), when compared to placebo. Rösler et al., 2010 examined the effects of extended release methylphenidate (MPH) on emotional symptoms of ADHD in a 24-week study. Extended-release MPH demonstrated improvements in emotional lability and dysregulation as well as improvements in obsessive compulsive disorder (OCD) symptoms, however no change in anxiety, phobia or major depression(MDD) symptoms were found. Lisdexamfetamine dimesylate (LDX) is a central nervous system (CNS) stimulant with a unique chemical structure, and has been found to be efficacious in both child, adolescent and adult ADHD. Given the high degree of comorbidity between anxiety disorders and ADHD, and the limited evidence to guide clinicians on treatment, a prospective trial examining adult ADHD with comorbidity was conducted using LDX. This poster will present the primary results of this study.

Methods: This study was a double-blind, placebo controlled, flexible-dose, cross-over evaluation of LDX in adult outpatients (aged 18-65) with DSM-IV ADHD with at least one of the following comorbid diagnoses: Panic disorder with/without agoraphobia (PD/PDAG), generalized anxiety disorder (GAD), SAD, OCD, dysthymic disorder or MDD. Participants were randomized to either LDX or placebo for 8 weeks according to a computer-generated randomization. Between weeks 8 and 9, all participants were treated with placebo, and then were crossed over to the opposite treatment condition. Primary outcome was the clinician-rated ADHD-Rating Scale (ADHD-RS); secondary outcomes included the Barkley Adult ADHD Rating Scale (BAARS-IV), the Overall Anxiety Severity and Impairment Scale (OASIS), the Revised Padua Inventory, the Panic and Agoraphobia Scale (PAS), the GAD-7, the Social Phobia Inventory (SPIN), the Quick Inventory of Depressive Symptoms (QID-SR-16), the Clinical Global Impression–Severity Scale (CGI-S), the Sheehan Disability Scale (SDS) and the Weiss Functional Impairment Rating Scale (WFIRS).

Results: Forty-six participants with primary ADHD were randomized to receive drug or placebo. The sample was 54.3% female with a mean age: 34.3 ± 11.1 years. Lifetime prevalence of comorbid DSM-IV diagnoses was high with the most common comorbidities being GAD (91.3%), SAD (60.9%) and MDD (58.7%). Participants receiving LDX or placebo did not differ on the ADHD-RS or CGI-S at baseline. Using LOCF-ITT analysis, LDX group achieved statistical separation from placebo at week 6 during Arm 1 (ADHD-RS -11.3 LDX vs. -3.8 PBO, $p = 0.03$ 95% CI = -12.7 to -0.7). Response rates were modest and not significantly different between groups (17.4% LDX vs. 8.7% PBO, $p =$ NS). During the first arm, 7 participants withdrew, producing an ITT population of 39 (20 PBO, 19 LDX) for Arm 2. Participants did not differ in mean ADHD-RS or CGI-S scores at baseline of Arm 2. Mean scores on the ADHD-RS were lower after crossover compared to Arm 1 baseline (LDX -4.9, $p = 0.06$ vs. PBO -6.2, $p = 0.02$) suggesting the presence of carry over effects. During Arm 2, ADHD-RS scores decreased for the LDX group, but failed to achieve statistically separation from placebo (all $p =$ NS). Similar to Arm 1, response rates were modest (15.8% LDX vs. 5.0% PBO, $p =$ NS) and not significantly different between groups. LDX did not appear to have any effect on comorbid anxiety or depressive symptoms. Mean scores on all secondary outcome measures did not differ between groups at any time point (all $p =$ NS).

Conclusions: These results suggest that LDX was efficacious in treating ADHD symptoms in individuals with comorbidity and did not appear to have a negative impact on anxiety or depressive symptoms. However, a significant difference was not found for rates of response between drug and placebo, which differs from studies examining LDX in ADHD without comorbidity. This may suggest that ADHD with comorbidity is a sub-type of the disorder, which warrants further investigation.

Keywords: ADHD, Psychiatric Comorbidity, Pharmacotherapy
Disclosure: Part 1: Shire Canada, Grant, **Part 4:** Shire Canada, Grant, Self.

T75. Psychopathological and Sociodemographic Features in Treatment Resistant Unipolar, Bipolar I and II Depression: A Comparative Study

Nicolas Nunez, Stefano Comai, Linda Booij, Michael Gabrash, Marie Saint-Laurent, Nancy Low, Pablo Cervantes, Gabriella Gobbi*

McGill University, Montreal, Canada

Background: Major depression is a disabling disorder affecting about 8.2% of people yearly with worldwide prevalence rates ranging from 10% to 19%. According to the STAR*D study, more than 50% of patients suffer from treatment-resistant depression (TRD), since they do not respond to a first trial of an antidepressant. Some authors have suggested that treatment resistant unipolar depression (TRD-UP) should be considered as an undiagnosed bipolar disorder type-I (BP-I) or type-II (BP-II), however no clinical studies have yet compared clinical characteristics between TRD-UP with BP patients. We thus examined sociodemographic and clinical traits that may differentiate TRD-UP from depression in the bipolar spectrum.
Methods: A chart analysis in 194 depressed patients with a diagnosis of TRD-UP ($n=100$), BP-I ($n=52$) or BP-II ($n=42$) was carried out by analyzing sociodemographic and clinical features and depression severity from the Registry of the Mood Disorder Clinic at McGill University Health Center (MUHC), by chart review analysis. Participants' diagnoses were ascertained by the Structured Clinical Interview for Diagnosis (SCID) carried out by skilled professional or by psychiatrists. All patients were assessed during depressive phase, prior to a final and stable treatment (stable for at least 3 months) using the Montgomery-Asberg Depression Rating Scale (MADRS), the Hamilton Rating Scale for Depression (HAM-D17), the Clinical Global Impression-Severity of Illness (CGI-S). Manic symptoms were assessed using the Young Mania Rating Scale (YMRS). TRD-UP subjects were also classified according to the Maudsley Staging Method (MSM). Multinomial logistic regression analysis was conducted to examine clinical predictors independently associated with TRD–UP, BP-I and BP-II.
Results: TRD-UP patients showed higher depression severity, higher prevalence of anxiety and panic disorders, of melancholic features, and of cluster C personality disorders, a later onset of depression and fewer hospitalizations than both BP-I and BP-II patients. They also had less suicide attempts than BP-II. Multinomial logistic regression showed that more than one hospitalization for depression, comorbidity with anxiety disorders, current psychotherapy, HAMD-17 and GAF scores differentiated patients with TRD-UP from those with BP-I. Comorbidity with anxiety disorders, psychotherapies, GAF score, number of previous failed pharmacotherapies were instead differentiating TRD-UP from BP-II. Finally, employment status, more than one hospitalization for depression and history of suicide could discriminate BP-I from BP-II depression.
Conclusions: These data indicate that TRD-UP, BP-I and BP-II have different sociodemographic and clinical traits, suggesting that TRD-UP is a distinct psychopathological condition and not a prodromal state of BP depression.
Keywords: Bipolar I Depression, Treatment Resistant Depression, Diagnosis, Antidepressant
Disclosure: Part 1: Merck, Honoraria, Sunovion, Honoraria, **Part 2:** Cosmas Therapeutics Development, Patent.

T76. De Novo Damaging Coding Mutations are Strongly Associated With Obsessive-Compulsive Disorder and Overlap With Autism

Carolina Cappi, Melody Oliphant, Zsanett Péter, Gwyneth Zai, Catherine Sullivan, Abha Gupta, Ellen Hoffman, Roseli Shavitt, Euripedes Miguel, James Kennedy, Margaret Richter, Thomas Fernandez*

Yale University, New Haven, Connecticut, United States

Background: OCD is an often-disabling developmental neuropsychiatric disorder with onset typically during adolescence or young adulthood and a lifetime prevalence of 1.5-2.5%. The WHO ranked OCD among the 10 most debilitating disorders of any kind, and treatment-refractory disease is common. The causes and underlying pathophysiology of OCD are not well understood, limiting the development of new treatments and interventions. For these reasons, there is an urgent need for more research to elucidate OCD risk factors and mechanisms. Despite strong evidence for a substantial genetic contribution to OCD risk, identification of high-confidence risk genes has been challenging. In contrast, genetic research into other neuropsychiatric disorders (e.g. autism) has seen great progress in recent years, partly attributable to an increasing effort to evaluate the contribution of rare genetic variants, especially de novo sequence variation (arising spontaneously in the parental germ cells or in a zygote shortly after conception). This proven avenue for risk gene discovery in complex neuropsychiatric disorders has yet to be fully leveraged in OCD.
Methods: We performed whole-exome sequencing (WES) and genetic variant calling in 222 OCD and 855 unaffected parent-child trios. We detected de novo mutations (DNMs) in all trios using validated algorithms, and performed the following analyses: (1) compared the rate of DNMs per base pair in cases versus controls; (2) by comparing DNM frequencies, estimated the fraction of observed DNMs contributing to OCD risk and the percentage of cases in whom a de novo mutation is contributing to OCD risk; (3) using the TADA statistical algorithm and previously established false discovery rate (FDR) thresholds, determined whether DNMs in OCD cluster within specific genes; (4) using a maximum likelihood estimation (MLE) method and an "unseen species" method, estimated the number of genes contributing to OCD risk; (5) using candidate gene sets for several disorders from the literature, tested for overlap with our OCD genes harboring DNMs; (6) using this same list of OCD genes, performed exploratory network connectivity and pathway enrichment analyses.
Results: After quality control, our sample size was 184 OCD and 777 unaffected trios. We confirmed our hypothesis that

likely gene disrupting (LGD: stop codon, frameshift, or canonical splice site) DNMs are significantly over-represented in OCD probands (rate ratio 1.93. 95% CI 1.19-3.09, $p = 0.01$). Missense DNMs predicted to be damaging (Mis-D; Polyphen2 HDIV score ≥ 0.957) were also enriched in OCD probands (RR 1.43, CI 1.13-1.80, $p = 0.006$). As a group, damaging DNMs (LGD + Mis-D) occur at a significantly higher rate in coding regions in OCD probands versus controls (RR 1.52, CI 1.23-1.86, $p = 0.0005$). No differences were seen in the rate of synonymous DNMs (RR 0.99, CI 0.75-1.31, $p = 0.5$).

Based on the DNM rates in cases and controls, we estimate that 53.8% of LGD DNMs and 37.6% of Mis-D DNMs contribute to OCD risk. As a group, we estimate that 41.2% of damaging (LGD+Mis-D) DNMs contribute to risk. We also estimate that 14.2% of cases have a Mis-D DNM and 5.7% of cases have an LGD DNM mediating OCD risk. As a group, we estimate that 18.2% of cases have a damaging DNM contributing to risk.

We identified three genes with multiple LGD or Mis-D DNMs in unrelated probands. Using TADA, two of these genes met criteria for high-confidence risk genes (FDR < 0.1): SCUBE1 (Signal Peptide, CUB Domain and EGF Like Domain Containing 1) and CHD8 (Chromodomain Helicase DNA Binding Protein 8). A third gene, TTN (Titin), did not meet this threshold (FDR = 0.7).

Using the MLE method, based on OCD proband vulnerability to damaging DNMs, we determined the most likely number of genes contributing to OCD risk to be 335. Using an alternate ("unseen species") method, this estimate is 317. Using DNENRICH, we found significant overlap between genes harboring damaging DNMs in OCD and similar variants in ASD. There was no significant overlap with genes in intellectual disability or schizophrenia.

Using the GeNets algorithm, OCD genes mapped onto a meta-network with more connectivity than expected by chance ($p = 0.026$). Using IPA and MetaCore, we found significant enrichment in canonical pathways related to immune response, particularly the complement system. Other significant pathways include granulocyte-macrophage colony stimulating factor (GM-CSF) signaling, neurotrophin/tyrosine kinase signaling, B cell receptor signaling, and focal adhesion kinase signaling.

Conclusions: We demonstrate a strong association between damaging DNMs and OCD and highlight a way to leverage gene-level recurrence of DNMs to systematically identify OCD risk genes. Two genes, CHD8 and SCUBE1, meet criteria for high-confidence association with OCD. We estimate that ~40% of damaging DNMs seen in OCD carry risk and that 335 genes confer risk in 17.5% of patients. Genes identified by damaging DNMs in OCD are functionally connected to a greater degree than expected by chance and are enriched in immunologic and cell signaling canonical pathways. Our study strongly reinforces the value of continuing WES in larger cohorts of OCD parent-child trios. Discovering risk genes will change the status quo in OCD genetics by allowing new studies in model systems and network analyses aimed at understanding the spatial, temporal, and cell-level dynamics of OCD pathophysiology; these are all critical prerequisites for the discovery of novel therapeutic targets to alleviate the suffering of those with OCD.

Keywords: Obsessive-Compulsive Disorder (OCD), Whole Exome Sequencing, De Novo Mutation, Autism Spectrum Disorder, Human Genetics
Disclosure: Nothing to Disclose.

T77. Kynurenic Acid Levels in Major Depressive Disorder: A Systematic Review and Meta-Analysis

Shinichiro Luke Nakajima*, Kamiyu Ogyu, Kaoruhiko Kubo, Yusuke Iwata, Yoshihiro Noda, Eric Plitman, Sho Moriguchi, Sakiko Tsugawa, Yuki Omura, Hiroyuki Uchida, Ariel Graff-Guerrero, Masaru Mimura

Keio University, Tokyo, Japan

Background: Kynurenic acid (KYNA) is an endogenous antagonist of glutamatergic N-methyl-D-aspartate (NMDA) receptors that is derived from astrocytes as part of the kynurenine pathway of tryptophan degradation. This pathway is expected to account for the relationship between major depressive disorder (MDD) and both neuroinflammation and glutamatergic dysfunction. Notably, it has previously been suggested that abnormal KYNA levels are involved in the pathophysiology of MDD. Thus, we conducted a systematic review of relevant literature and a meta-analysis to investigate whether any differences in KYNA levels exist between patients with MDD and healthy controls (HCs).

Methods: A literature search was conducted using Pubmed (last search: June 15, 2017) with the search terms: (kynuren* or KYNA or quinolinic or QUIN or QA) AND depressi* NOT bipolar. English language studies measuring KYNA levels using any technique in both patients with MDD and HCs were identified. Cross-reference and hand searches were also performed. The variables recorded from each included study were KYNA levels, diagnoses, age, sex, antidepressant treatment status, duration of illness, symptom severity, and method of KYNA measurement. Standardized mean differences (SMDs) were calculated to determine differences in KYNA levels between groups.

Results: Out of 257 initial records, 18 articles were identified to form the empirical basis of this analysis. Eleven (61.1%) studies examined KYNA levels and 7 (38.9%) studies examined other metabolites in the kynurenine pathway. Among the KYNA studies, nine (81.8%) studies examined serum and two (18.2%) studies examined cerebrospinal fluid (CSF). In the main analysis, KYNA levels were decreased in patients with MDD (n = 433) in comparison to HCs (n = 610) (SMD = -0.61, CI = -1.01 to -0.21, P < .00001).

Conclusions: This meta-analysis suggested that KYNA levels may be decreased in patients with MDD with a moderate effect size. However, given the small number of included studies and heterogeneity of subject characteristics, further research is clearly needed to quantitatively examine the levels of KYNA and other metabolites in the kynurenine pathway in patients with MDD. Further understanding of the role of KYNA and its related metabolites on MDD will assist in elucidating the pathophysiology of MDD and developing novel treatment options.

Keywords: Kynurenine Pathway, Depression, Meta-Analysis, Kynurenic Acid
Disclosure: Nothing to Disclose.

T78. Efficacy of Lurasidone in Preventing Recurrence of Bipolar Disorder: Results of a Multistate Outcome Analysis of Treatments (MOAT)

Mauricio Tohen*, Andrei Pikalov, Yongcai Mao, Joyce Tsai, Antony Loebel

University of New Mexico Health Science Center, Albuquerque, New Mexico, United States

Background: The primary efficacy outcome in the majority of maintenance treatment studies in patients with bipolar disorder is time-to-recurrence of a mood episode evaluated using Kaplan-Meier survival analysis. Risk reduction estimates may then be calculated based on a Cox proportional hazards model. However, point estimates of time-to-recurrence do not provide clinicians with information on the changing topography of affective symptom severity over the course of maintenance therapy. Bowden and colleagues (Molec Psychiatry 2016; 21:237-242) have developed a MOAT analysis methodology designed to provide this type of detailed information on the duration of syndromic and subsyndromic periods of affective symptomatology. The aim of the current analysis was to assess the effect of maintenance treatment with lurasidone on a range of affective symptom severity states, using MOAT methodology, based on a placebo-controlled maintenance trial of lurasidone in patients with bipolar disorder.
Methods: Data were analyzed from a study of patients with bipolar I depression or mania/hypomania that consisted of an open-label stabilization phase (up to 20 weeks on lurasidone adjunctive with lithium or valproate) followed by a 28-week, double-blind maintenance period in which patients were randomized to continue adjunctive lurasidone, or were switched to placebo, adjunctive with lithium or valproate. The primary a priori endpoint was time-to-recurrence of any mood episode. In the current post-hoc analysis, utilizing MOAT methodology, survival time was partitioned into clinically distinct periods that were operationally defined by cut points on the Montgomery-Asberg Depression Rating Scale (MADRS) and the Young-Mania Rating Scale (YMRS). The clinical states analyzed were: remission, subsyndromal (SS) and syndromal (SYN) mania; subsyndromal (SS) and syndromal (SYN) mixed states; and subsyndromal (SS) and syndromal (SYN) depression.
Results: The analyzed sample consisted of patients randomized to lurasidone + Li/VPA [$N=246$; index episode depression (58.5%), mania (22.8%), mixed mania (12.6%), hypomanic (6.1%)], and placebo + Li/VPA [$N=250$; index episode depression (47.6%), mania (24.8%), mixed mania (21.2%), hypomanic (6.4%)]. Lurasidone was found to be associated with a significantly greater (vs. placebo) mean number of days spent in remission (124.2 vs. 105.3; $P<0.01$; t-test using Cochran-adjusted P-value), and significantly greater mean percent of time spent in remission (73% vs. 62%; $P<0.01$; t-test using Cochran-adjusted P-value). Duration of time spent in other clinical states (SYN-depression, SS-depression, SYN-mania, SS-mania, SYN-mixed, SS-mixed) was not significantly different for lurasidone vs placebo. Lurasidone patients experienced a significantly greater mean number of days in the study before a mood recurrence event, discontinuation, or lost-to-follow-up (165.8 vs. 151.3 days; $P=0.018$).
Conclusions: In this post-hoc analysis (using MOAT methodology) of a randomized, double-blind maintenance study of bipolar disorder, treatment with adjunctive lurasidone was significantly more effective than placebo (+ Li/VPA) in maintaining patients in remission. However, there were no between-treatment group differences in the duration of time spent in syndromal or subsyndromal affective states.
Keywords: Longitudinal Analysis, Lurasdione, Bipolar Disorder
Disclosure: Part 1: Sunovion, Advisory Board, Otsuka, Advisory Board, Abbott, Honoraria, Lundbeck, Advisory Board, Neuroscience, Advisory Board, Minerva, Advisory Board.

T79. Effects of Rapastinel (Formerly GLYX-13) on Serum Brain Derived Neurotropic Factor in OCD

Omer Linkovski, Hanyang Shen, Jordana Zwerling, Maria Filippou-Frye, Booil Jo, Elisabeth Cordell, Thomas Cooper, Helen Simpson, Ronald Burch, Joseph Moskal, Francis Lee, Carolyn Rodriguez*

Stanford University School of Medicine, Stanford, California, United States

Background: Serum Brain-Derived Neurotrophic Factor (BDNF) may be a treatment efficacy marker in depression. Rapastinel is an NMDA glutamate receptor modulator, shown to affect BDNF levels in-vitro. It exerts acute therapeutic effects on obsessive-compulsive disorder (OCD) symptoms with a minimal risk of ketamine-like side effects. We analyzed exploratory data regarding serum BDNF samples collected from OCD patients pre- and post- rapastinel infusion. Our goal was to determine whether 1) rapastinel alters serum BDNF levels in the interval from 0 to 230 minutes post-infusion, and 2) whether changes in BDNF levels correlated with changes in OCD symptoms during this period.
Methods: Seven un-medicated outpatients diagnosed with OCD (ages 18–55) were recruited (March 2014 to March 2015) and met criteria for moderate OCD severity. All participants ($N=7$) received a single 3-to 5-minute intravenous infusion of rapastinel (10 mg/kg). Blood was drawn at baseline, 40, 110 and 230 minutes post-infusion. Serum BDNF levels were determined using a Millipore ChemiKine Sandwich ELISA Kit. OCD severity was assessed at baseline and 230 minutes, using the YBOC Challenge Scale, a 10-item self-report form assessing obsessions and compulsions over the previous hour. We assessed BDNF level changes over time using the non-parametric Wilcoxon signed-rank test. A mixed-effects regression model assessed BDNF levels' time trend. Spearman correlation tested whether changes in serum BDNF levels from baseline to 230 minutes post-infusion correlated with OCD symptom severity change. As a negative control, we compared BDNF levels of the current patient sample to an independent cohort of seven OCD patients who received a ketamine infusion, with BDNF serum levels determined in identical intervals, at the same time of day. Both samples sets were processed and analyzed concurrently with identical methods.
Results: In the rapastinel group, baseline median serum BDNF level was 22.3 ng/ml (range 16.8-34.9). At 230 minutes, median BDNF level was 17.55 ng/ml (range 12.6-26) and

decreased in most participants (85.7%, $N=6$). This reduction was statistically significant ($p = .031$). BDNF levels exhibited a significant time-trend, decreasing from baseline to 230 minutes ($\beta = -.019$, $p = .0017$). In contrast, in the ketamine group, the infusion did not significantly decrease serum BDNF levels over time. Baseline BDNF levels did not differ between the rapastinel and the ketamine OCD groups ($p = .16$). Median YBOC Challenge Scale at baseline was 28 (range 24-39), and decreased in all participants ($N=7$) at 230 minutes (median = 15; range 3-20). Changes in serum BDNF levels significantly correlated with changes in YBOC Challenge Scale scores between baseline and 230 minutes ($R = .86$, $p = .012$).

Conclusions: In this small sample, rapastinel reduced OCD patients' serum BDNF levels from baseline to 230 minutes, and these changes correlated with changes in OCD symptom severity. In a control sample, ketamine reduced OCD symptom severity but did not change serum BDNF levels. While both compounds relieved OCD symptoms, they acted differently on serum BDNF levels, suggesting that elucidating their mechanisms of action requires further investigation.

Keywords: Rapastinel, BDNF, Ketamine, GLYX-13, OCD

Disclosure: Part 4: Naurex, Grant, Allergan, Consultant, Rugen Therapeutics, Consultant, BlackThorn Therapeutics, Consultant.

T80. Sensing Depression: Using Smartphone Sensors to Predict Changes in Depression Severity

Ellen Frank*, Michael Merrill, Hane Aung, Claudio Soares, Sidney Kennedy, Mark Matthews, David Kupfer, Aleksandra Lalovic, Tanzeem Choudhury

University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania, United States

Background: A major challenge in the management of depressive illness is the clinician's or clinical researcher's relative inability to assess the patient's clinical state between face-to-face encounters. Yet, there is substantial evidence to suggest that episodes of clinically significant depression are preceded by behavioral changes that are not easily captured with traditional measurement methods; however, many of these changes can be readily measured using the GPS and accelerometer sensors within all commercial smartphones. Most important from the standpoint of their utility as predictors, these data can be acquired on a 24/7 basis over long periods of time without active effort on participants' part. We report on a collaboration between the Canadian Biomarker Integration Network in Depression (CAN-BIND) and HealthRhythms, Inc. to explore the potential of the smartphone as a means of acquiring such behavioral data and examining the relationship between such measures and changes in depression status.

Methods: As an initial step in the collaboration between CAN-BIND and HealthRhythms, we used Measure, a smartphone app intended to collect continuous behavioral data relevant to those disorders in which sleep-wake and circadian dysregulation are implicated to monitor a small group ($N=11$) of adult outpatients with a lifetime diagnosis of major depressive disorder. Participants were asked to download the Measure app and keep it running continuously over a period of 8 weeks. The PHQ-8 was administered at baseline and every two weeks throughout the study. We considered a group of 17 variables, including measures such as sleep duration, walking rate, time spent at home, and travel diameter, extracted from the accelerometer and geolocation sensors. We built a simple model by regressing PHQ-8 scores on the average of the sensed inferences from the preceding two weeks. We used a generalized estimating equation (GEE) to examine associations between sensed variables and PHQ-8 scores.

Results: The 11 study participants included in these analyses provided 11,317 hours of data, constituting 73.4% of the time they were enrolled in the study and indicating good coverage of their behavior during the study. Not surprisingly, we found that sensed measures associated with motor activity, sleep and decreased social interaction (i.e. decreased travel diameter) and a measure we refer to as 'behavioral trend' collected over the two weeks prior to a PHQ-8 assessment are significantly predictive of higher depression scores. Our regression model shows significant relationships with 'activity percent' (the percentage of time the participant spent walking), step count, travel diameter, sleep duration, and our 'behavioral trend' statistic. (p's = 0.012, 0.013, 0.005, 0.007, and 0.002 respectively).

Conclusions: While we are only beginning to analyze the complex relationships in our multidimensional dataset, preliminary findings suggest highly informative relationships between our sensed inferences and self-reported depression levels that are consistent with established knowledge about the precursors of depressive disorders. Furthermore, these initial analyses suggest the utility of our mobile app as a feasible means of collecting clinically-relevant behavioral data than can inform the management – and even the self-management – of depressive disorders. Furthermore, the ubiquity of the smartphone and its almost continuous presence in patients' lives make it an ideal method for behavioral data collection since it requires little to no effort on the part of patients, no special equipment, can be implemented at relatively low cost and appears to be entirely acceptable to patients.

Keywords: Depression, Prediction, Mobile Technology

Disclosure: Part 1: Psychiatric Assessments, Inc., Stock / Equity, Self/Spouse, HealthRhythms, Inc., Stock / Equity, Self/Spouse, Servier, Advisory Board, Self/Spouse, Minerva Neuroscience, Board Member, Spouse, Minerva Neuroscience, Stock / Equity, Self/Spouse, **Part 2:** Minerva Neuroscience, Board Member, Spouse, HealthRhythms, Inc., Employee, **Part 3:** HealthRhythms, Inc., Employee, **Part 5:** HealthRhythms, Inc., Employee.

T81. Influence of Flavor Additives on Adolescent Nicotine Reward: Development of a Mouse Model Using Commercial E-Cigarettes

Theresa Patten*, Erika Perez, Mariella De Biasi

University of Pennsylvania Perelman School of Medicine, Philadelphia, Pennsylvania, United States

Background: E-cigarette (e-cig) use is rapidly rising among adolescents worldwide. The overwhelming popularity of sweet flavorants among adolescent e-cig users and the vulnerability of the brain during this critical stage in

development make it crucial to better understand how these products might affect nicotine reward and the adolescent brain.
Methods: We have established a model to deliver nicotine vapor to awake C57/BL6J mice using a commercial e-cigarette and employing a topography that closely mimics a human "vaping" session. This approach represents an improvement over other delivery methods (e.g. systemic injections of nicotine, osmotic pumps, constant vapor exposure), which do not precisely mirror the pharmacokinetic profile of nicotine inhalation. We have confirmed that a 25-minute exposure to 12 mg/ml nicotine vapor results in plasma cotinine concentrations comparable to those achieved following a 0.5mg/kg intraperitoneal nicotine injection. We use this model of nicotine delivery in combination with the conditioned place preference (CPP) behavioral paradigm to test the hypothesis that sweet flavorants in e-cigs will enhance adolescent nicotine reward. These behavioral experiments are complemented by molecular techniques for the detection of changes in immediate early gene expression and the measurement of dopamine levels in the nucleus accumbens (NAc) following exposure to both flavored and unflavored e-cigarette vapor.
Results: These studies are the first to deliver nicotine with a topography that closely mimics a human "vaping" session in order to measure drug reward. We show that nicotine CPP can be established through delivery of e-cig vapor to adolescent mice. Furthermore, the addition of a fruit flavorant to a subthreshold dose of nicotine vapor increases nicotine reward in adolescent mice. Preliminary data also suggests that there is an increase in c-fos staining following exposure to flavored nicotine vapor, when compared to unflavored nicotine vapor. Finally, dopamine microdialysis in the nucleus accumbens (NAc) further elucidates the influence of sweet flavorants on adolescent nicotine reward.
Conclusions: In 2009, the Family Smoking and Prevention and Tobacco Act banned "characterizing" flavors from being used in tobacco products. However, the FDA's announcement in 2016 to extend regulations on Electronic Nicotine Delivery Systems (ENDS) via its "Deeming Act," did not include a flavor ban. Our data suggests that fruit-flavored nicotine vapor enhances neuronal activity within the mesolimbic dopamine system and potentiates nicotine reward. This data is unique in that it could have important implications on the safety of adolescent e-cigarette consumption, and could impact FDA regulations of e-cigarettes.
Keywords: Electronic Cigarette (e-cigarette), Adolescence, Nicotine, Reward
Disclosure: Nothing to Disclose.

T82. Effect of Citalopram on Emotion Processing in Humans: A Combined 5-HT1A [11C]CUMI-101 PET and Functional MRI Study

Sudhakar Selvaraj*, Christopher Walker, Danilo Arnone, Bo Cao, Paul Faulkner, Philip Cowen, Jonathan Roiser, Oliver Howes

University of Texas Health Science Center at Houston, Houston, Texas, United States

Background: A subset of patients started on a selective serotonin reuptake inhibitor (SSRI) initially experience increased anxiety, which can lead to early discontinuation before therapeutic effects are manifest. The neural basis of this early SSRI effect is not known. Presynaptic dorsal raphe neuron (DRN) 5-HT1A receptors are known to play a critical role in affect processing. We investigated the effect of acute citalopram on emotional processing and the relationship between DRN 5-HT1A receptor availability and amygdala reactivity.
Methods: Thirteen (mean age 48 ± 9 years) healthy male subjects received either a saline or citalopram infusion intravenously (10 mg over 30 minutes) on separate occasions in a single-blind, random order, cross-over design. On each occasion, participants underwent a block design face-emotion processing task during functional magnetic resonance imaging (fMRI) known to activate the amygdala. Ten subjects also completed a positron emission tomography (PET) scan to quantify DRN 5-HT1A availability using [11C] CUMI-101.
Results: Citalopram infusion when compared to saline resulted in a significantly increased bilateral amygdala responses to fearful vs. neutral faces (Left $p = 0.025$; Right $p = 0.038$ FWE-corrected). DRN [11C]CUMI-101availability significantly positively correlated with the effect of citalopram on the left amygdala response to fearful faces ($Z = 2.51$, $p = 0.027$) and right amygdala response to happy faces ($Z = 2.33$, $p = 0.032$).
Conclusions: Our findings indicate that the initial effect of SSRI treatment is to alter processing of aversive stimuli, and that this is linked to DRN 5-HT1A receptors in line with evidence that 5-HT1A receptors have a role in mediating emotional processing.
Keywords: SSRI, Amygdala, Antidepressant, Functional MRI (fMRI), PET Imaging
Disclosure: Nothing to Disclose.

T83. Proactive Cognitive Control as a Double-Edged Sword in Autism Spectrum Disorder

Jeremy Hogeveen*, Marie Krug, Matthew Elliott, Cameron Carter, Marjorie Solomon

MIND Institute, University of California, Davis School of Medicine, Sacramento, California, United States

Background: Whereas cognitive neuroscience research on autism spectrum disorder (ASD) has often focused on social and affective processes, fewer studies have focused on the cognitive symptoms that are classically associated with ASD. ASD is associated with an idiosyncratic tendency to perform restricted and repetitive behaviors (RRBs), including self-injurious, stereotyped, or compulsive behaviors that are maladaptive to the individual. Additionally, roughly half of individuals with ASD are at or near diagnostic criteria for comorbid attention deficit hyperactivity disorder (ADHD), and attention problems in ASD are associated with difficulties in disengaging and shifting attention. It has been hypothesized that impaired cognitive control plays a role in RRBs and attention problems in ASD, but empirical evidence for this connection is limited. One possibility is that individuals with ASD show perseverative behaviors and have difficulties shifting attention because of a disrupted ability to transition between thoughts or actions depending

on the context. The ability to use stimulus context to control cognitive processing is central to the construct 'proactive cognitive control,' which refers to the ability to use contextual information to guide early selection of task-relevant stimuli prior to the occurrence of a cognitively demanding event. Here, we merged a proactive control task with clinical evaluations in children with ASD or typical development (TYP), and tested two related hypotheses: i) that proactive control is impaired in ASD, and ii) that this impairment is associated with elevated RRBs and attention problems.

Methods: 79 participants–34 with ASD (Age = 10.44 ± 1.66 years; 4 F) and 45 with TYP (Age = 10.93 ± 1.34; 8 F)–completed the study. Proactive control was assessed using a children's version of the AX-continuous performance test (AX-CPT). The AX-CPT contains cue-probe sequences of the form AX, AY, BX, or BY, with 1200ms separating cue and probe. AX sequences occur on 70% of trials, and require participants to press a 'target' button. AY, BX, or BY trials each occur on 10% of trials, and require participants to press a 'nontarget' button. Proactive control in this task affords a high hit rate on AX trials, as participants proactively use the A-cue context to prepare a prepotent 'target' response. Conversely, proactive control increases false alarm rates on AY trials, as the Y-probe does not match the anticipated prepotent response plan. Therefore, we computed a measure known as the A-Cue Bias, which averages across hits on AX trials and false alarms on AY trials to describe the degree to which the A-cue context proactively biases participant responses. Additionally, we used the parent-report Child Behavior Checklist (CBCL) to quantify attention problems, and the parent-report Repetitive Behavior Scale-Revised (RBS-R) to measure RRBs in our participants.

Results: Proactive control was matched between children with ASD and TYP. However, among participants with ASD we observed a paradoxical relationship between the degree of reliance on proactive control and clinical symptoms. Specifically, A-Cue Bias was regressed onto group (ASD, TYP) and CBCL attention problems, as well as an interaction term. Robust linear regression models were fit due to skew and heteroscedasticity in the data. There was a significant interaction effect ($p = 0.028$), driven by a negative relationship between attention problems and A-Cue Bias within the ASD group ($p = 0.042$), and no association between these variables in TYP. In a separate model, three-factors derived from the RBS-R (self-injurious behaviors; stereotyped/restricted behaviors; and compulsive/ritualistic/sameness behaviors, CB) and participant group were included as predictors, and a significant main effect of CB was observed ($p = 0.002$). The interaction was not significant, due to the fact that TYPs were at floor across all factors of the RBS-R. Importantly, the main effect of CB was driven by a positive relationship between CB and A-Cue Bias within the ASD sample ($p = 0.001$).

Conclusions: The ability to rely on proactive control appears to represent a double-edged sword in children with ASD. Using robust dimensional analyses, we find that greater proactive control among participants with ASD is associated with an improved ability to control attentional processes. This improvement in attentional control comes at the cost of heightened behavioral inflexibility, as participants with greater proactive control also demonstrate higher CB symptoms. Our work begins to illustrate that psychopharmacological, neural retraining, and neural stimulation interventions in the current treatment armamentarium must be selected thoughtfully based on patient characteristics, given the potential negative side effects (e.g. increasing inflexibility) of these interventions for some. It will be crucial to further investigate the prevalence and neural mechanisms of proactive control in ASD in order to improve our capacity to implement a precision medicine approach when treating affected individuals. Finally, once more is known about the mechanisms governing proactive control and whether there are proactive control profiles based on this cognitive characteristic, it may prove worthwhile to use the level of proactive control as a means of stratifying ASD patients in clinical trials of agents seeking to improve both context maintenance and flexibility—the two sides of the double-edged sword.

Keywords: Autism Spectrum Disorder, Cognitive Control, Attention

Disclosure: Nothing to Disclose.

T84. Sex Specific Effects of Variable Stress on Circulating Cytokines

Jennifer Rainville, Amanda Patterson, Georgia Hodes*

Virginia Polytechnic Institute and State University, Blacksburg, Virginia, United States

Background: Depression affects more than 300 million people worldwide, and the women are at twice the risk of men to experience a depressive episode. Pro-inflammatory cytokines and the activation of inflammatory pathways have been associated with depression. Many autoimmune and inflammatory diseases also have a higher incidence in women, presenting the possibility that sex-specific peripheral immune system responses to stress may influence sex differences in susceptibility to depression. The purpose of this study is to characterize how peripheral cytokine levels reflect differences in stress susceptibility between males and females.

Methods: We used A 6-day variable stress model, in which stressed female mice show a stronger behavioral response to stress (Hodes et al., 2015) to observe sex differences in peripheral cytokine levels. Animals were exposed to a combination of foot shock (0.45 mA/ 2 sec/ 100 in 1 hour), tail suspension and restraint stress over the course of 6 days. Each stressor was applied once a day for an hour and given 2 times over the 6-day period. Blood was drawn from animals within 20 minutes of stress exposure (restraint stress) on the last day of SCVS, prior to behavior via a submandibular bleed. Plasma was obtained for subsequent analysis. Multiplex ELISA was used as an unbiased screen to quantify plasma levels of 32 different cytokines from the same sample.

Results: The overall pattern of expression indicated that male and female peripheral immune systems respond differently to 6 days of variable stress. In general, females expressed an anti-inflammatory profile when exposed to stress compared to males. Eotaxin and IL-10 were oppositely regulated by stress in males and females as indicated by a significant interaction ($p < 0.05$). A main effect of stress indicated that the chemokines RANTES/CCL5 and KC/

CXCL1 were altered in both sexes. Regardless of stress, females expressed an anti-inflammatory profile with sex specific expression of GM-CSF, IL-9, IL-5, MIG/ CLXL9 and MIP1a/ CCL3. We generated z-scores for all animals to correlate behavior with cytokine levels and found that IL-9 and IL-10 positively correlated with stress susceptibility scores. GM-CSF and IL-12(p40) negatively correlated with behavior.

Conclusions: Males and females express different patterns of cytokines when exposed to 6 days of variable stress. Females display an overall anti-inflammatory pattern which is accentuated by stress, whereas males express stronger regulation of pro-inflammatory cytokines. The female response to stress is reminiscent of a Th2 response to parasite infection or allergy.

Keywords: Emotional Stress, Animal Model, Sex Differences, Immune Biomarkers, Cytokines

Disclosure: Nothing to Disclose.

T85. The Janssen Autism Knowledge Engine (JAKE): Results From a Large, Prospective Observational Biosensor Study in Autism Spectrum Disorder

Gahan Pandina*, Manyakov Nikolay, Abigail Bangerter, David Lewin, Shyla Jagannatha, Matthew Boice, Andrew Skalkin, Seth Ness

Johnson & Johnson, Pennington, New Jersey, United States

Background: There is a lack of sensitive, validated measures for assessing change in Autism Spectrum Disorder (ASD). This adversely impacts development of new therapies, and no approved treatments exist for ASD core symptoms. The Janssen Autism Knowledge Engine (JAKE®) has been developed to provide valid, reliable measures for use in assessing treatment outcome, as well as experimental endpoints for detecting change and identifying population subgroups. JAKE was tested in a prospective observational study to establish the clinical validity of a new rating scale measuring the symptoms of ASD, and the value of using experimental biosensors to objectively measure the symptoms of ASD during computer challenge tasks.

Methods: This prospective observational clinical study was conducted at 9 sites in the US. Participants aged ≥ 6 years were consented and enrolled (ASD $N = 144$, mean age 14.6 years; Typically Developing/TD $N = 41$, mean age 16.3 years). ASD subjects were assessed over 8 to 10 weeks using the JAKE Sense at 3 time points, while TD subjects were assessed once for comparison. JAKE components included My JAKE, a web and mobile application for tracking behavior, and; JAKE Sense, an array of experimental biosensors (electroencephalography, eye-tracking, cardiovascular measures, facial affect recognition, and motion/ activity) which records information from subjects during a computer-administered task battery.

Results: Results suggest a range of phenotypic differences between ASD and TD populations in brain activity assessed via electroencephalography (e.g., difference in left/right hemispherical asymmetry [p = 0.02]), attention to social scene assessed via an eye-tracker (e.g., reduced attention to eyes in ASD during joint attention condition [$p < 0.001$]), cardiovascular measures evaluated via electrocardiography (ECG, e.g., increased ASD heart rate [$p < 0.001$]), and facial affect evaluated via automated facial expression analysis software (FACET, e.g., reduced evidence of upturned mouth when making a 'happy face' [$p < 0.001$]).

The ABI, a novel parent-reported scale for assessing change in core and associated ASD symptoms showed good research performance characteristics (Test-retest ICC $>$ 0.85 – 0.95, convergent validity ICC $>$ 0.7). ABI Core, Social Communication (SC), and Restrictive Repetitive Behavior (RRB) scores were able to detect improvements in severity based on category change in Social Responsiveness Scale-2nd Revision Total Score (within-group effect sizes -0.63, 0.48, and 0.43, respectively).

Correlation between experimental biosensors/tasks, the ABI, and clinical gold standard scales used in ASD research showed significant relationships, informing potential biosensors of interest (e.g. eyes closed resting state absolute theta with ASD core symptoms r = -0.4, [$p = 0.001$]). There was evidence of some developmental differences in biosensor/task performance in the ASD population, suggesting that objectively measured aspects of ASD may be influenced by age or stage of illness. (e.g., relationship of emotion production [sad] to symptoms significant in <13 years, $p = 0.005$, not in >13 years).

Conclusions: JAKE measures diverse aspects of ASD, including both traditional clinical endpoints and experimental biosensor/tasks. Correlations between clinical and sensor-based measures, and their ability to measure change over time, will enrich our understanding of ASD. JAKE may suggest useful experimental methods that could enhance the development novel medicines to address the significant unmet medical need in ASD.

Keywords: Autism Spectrum Disorder, Eye-Tracking, Electroencephalography

Disclosure: Part 5: Johnson & Johnson, Employee.

T86. Adolescents and Young Adults With Autism Spectrum Disorder Show Differences in Dynamics and Recruitment of Cognitive Control Networks

Marjorie Solomon*, Marie Krug, Cory Coleman, Matthew Elliott, Jeremy Hogeveen, Tara Niendam, Ragland Dan, Cameron Carter

University of California, Davis, Sacramento, California, United States

Background: Individuals with Autism Spectrum Disorder (ASD) exhibit cognitive control (CC) deficits that appear to persist into adolescence and adulthood. Cognitive control deficits may be associated with the difficulties in social functioning, adaptive functioning and restricted and repetitive behaviors that are characteristic of many individuals with ASD, and are thus critical to understand. Previous fMRI studies with slow event-related versions of the preparing to overcome prepotency (POP) task found evidence that typically developing (TYP) individuals develop a mature LPFC/Parietal network in adolescence, which aids them in successfully inhibiting motor responses on incongruent trials. Individuals with ASD, however, may continue to rely more heavily on a less mature dACC/LPFC network when

presented with a situation that requires the implementation of CC. We use data from the first wave of a five year cohort-sequential study aimed at tracking the neurodevelopment of CC in adolescents and young adults with ASD to: (1) investigate potential group differences in CC by looking at behavioral performance in a rapid event-related version of the POP task in participants ages 12-22 years old, (2) associate behavioral measures with recruitment of brain regions known to play a role in successful implementation of CC, and (3) validate this new version of the POP task by examining brain behavior associations using the NIH Toolbox Cognition Battery.

Methods: Participants included 38 individuals with ASD (mean age = 117.2 years; mean IQ = 101) diagnosed using gold standard measures and 40 individuals with TYP (mean age = 117.2 years; mean IQ = 113). They completed 4 28 trial runs of the Rapid Preparing to Overcome Prepotency (rPOP) task in the fMRI environment. In the rPOP, they were first presented with a fixation cross. This was followed by a cue which signaled whether they should push a button on the same side (cued by a green square) as indicated by the probe, which was an arrow, or to the opposite side relative to where the arrow pointed (cued by a red square). After an inter-stimulus interval, the probe arrow was presented. Both inter-stimulus and inter-trial intervals were jittered with a mean time of 4,500 milliseconds (ms). Data was acquired using a 3 Tesla Siemens Tim Trio with a 32-channel head coil. Data were preprocessed and analyzed using SPM12. Cue and probe phases of red and green trials were modeled separately in the GLM. Error and post-error trials were modeled separately from correct trials and excluded from contrasts. The GLM included translational and rotational movement regressors. Functional connectivity analyses were used to examine ROI to ROI connectivity using the CONN functional connectivity toolbox (http://www.nitrc.org /projects/conn). Dorsal anterior cingulate (dACC) & dorsolateral prefrontal cortex (DLPFC) ROIs were created, centered on coordinates from MacDonald et al. (2000). To examine fronto-parietal connectivity, a structural parietal ROI was created from BA 7 and BA 40. The NIH Toolbox Cognition Battery was used to examine associations between imaging results and performance on common neuropsychological tests.

Results: Behavioral performance on the task as shown by an inverse efficiency score that indexes inefficient performance (IES; RT(ms)/accuracy), demonstrated that there was a main effect of both cue type and diagnosis and a significant cue type X diagnosis interaction ($F = 10.17$, $p < .005$), indicating that the ASD group was less efficient at the task. Whole brain analysis indicated that recruitment in the TYP group for the red-green cue was significantly greater in BA 32 where there was a main effect of cue type and a cue type x diagnosis interaction ($F = 3.42$, $p < .05$). In the ASD group, the IES for red-green trials was significantly negatively correlated with recruitment in both the DLPFC and parietal ROIs. During the presentation of red cues, both ASD and TYP showed significant functional connectivity between the DLPFC and the parietal ROIs, however, for the ASD versus the TYP group, there also was significantly stronger functional connectivity between the dACC and the DLPFC that was negatively associated with task performance. The ASD group performed comparably to the TYP group on the NIH Toolbox List Sorting Working Memory and Picture Sequence Memory tasks, but demonstrated significant relative impairment on the Flanker ($p < .001$) and Dimensional Change Card Sorting ($p < .001$) tasks. Across both groups, the IES for the red-green contrast was significantly negative correlated with performance on these two tasks.

Conclusions: Adolescents and young adults with ASD show impaired efficiency on the rPOP compared to TYP. For individuals with ASD, recruitment of DLPFC and parietal cortex at the cue correlates with better performance on the rPOP task. Participants with ASD show a pattern of greater activation and connectivity of dACC at the cue phase, which is associated with poorer task performance and should be more thoroughly investigated. Performance on the RPOP task is closely related to well-established neuropsychological tasks in the NIH Toolbox Cognition Battery, where individuals with ASD show impairment in CC, but not memory tasks. These early results also suggest that the neural circuitry underlying cognitive control as engaged by rPOP maps well to that predicted by neuropsychological task performance for both groups suggesting this circuitry may be transdiagnostic.

Keywords: Cognitive Control Network, Autism Spectrum Disorder, Adolescent, Functional Magnetic Resonance Imaging, Neuropsychology

Disclosure: Nothing to Disclose.

T87. D-Cycloserine Augmentation of Cognitive Behavior Therapy for Pediatric OCD: Predictors and Moderators of Outcome

Sabine Wilhelm, Noah Berman, Brent J. Small, Eric A. Storch, Rachel Porth, Daniel Geller*

Massachusetts General Hospital, Boston, Massachusetts, United States

Background: Previous research suggests that cognitive behavioral therapy (CBT) is an efficacious treatment for pediatric OCD. However, up to 60% of children with OCD receiving CBT do not fully remit. Researchers have attempted to augment CBT with D-cycloserine (DCS), an N-methyl-D-aspartate (NMDA) partial agonist that has been shown to enhance extinction learning, with mixed results. While results of the largest trial to date (5R01MH093402-02) suggest that DCS did not improve outcomes for pediatric OCD patients, it may be that baseline factors moderate a patients' response to DCS. The present study aimed to identify the predictors and moderators of DCS of CBT, or baseline characteristics that are associated with outcomes and the conditions in which DCS augmentation worked best. Identifying these factors would allow clinicians to better direct pediatric patients with OCD to the most appropriate treatment option.

Methods: Two hundred and six children and adolescents were enrolled at either Massachusetts General Hospital (MGH) in Boston, MA or University of South Florida (USF) in St. Petersburg, FL. One hundred and forty-two patients with an OCD diagnosis per DSM-IV-TR criteria as

determined by the Schedule for Affective Disorders and Schizophrenia for School-Age Children– Present and Lifetime Version (KSADS-PL) (M = 12.79, SD = 2.99; range 7-17-years-old) were randomized to either DCS + CBT (n = 70) or placebo + CBT (n = 72; for full consort diagram, please refer to Storch et al., 2016). Eligible children received 10 sessions of cognitive behavioral treatment, with two sessions per week for the first two weeks. Participants were randomized in a double-blind approach to either CBT augmented with DCS or CBT and placebo before the fourth CBT session. They then received either DCS or a placebo pill one hour prior to exposure sessions. We adopted an exploratory approach, as prior research on the predictors of CBT for pediatric OCD is conflicting. We looked at the extent to which demographics (age, gender, race, ethnicity), OCD-specific features (symptom severity, family accommodation, impairment), comorbidity (number of comorbidities, a comorbid anxiety, depression, or tic disorder, anxiety symptom severity, depression symptom severity, and externalizing behavior symptom severity), and treatment-related factors (treatment alliance at randomization) predicted treatment response. We also looked at how demographic (age and gender) and comorbidity (previous or current history of depression, anxiety and tics) variables interact with DCS augmentation to moderate outcome. All procedures performed in studies involving human participants were in accordance with the ethical standards of the Institutional Review Boards at the USF and MGH. Written parental informed consent and child assent were obtained from all individual subjects participating in the study.

Results: Moderator analyses examined the extent to which age, gender, current history and previous history of depression, anxiety and tics predicted treatment outcomes. Although there were significant improvements in all outcome measures (Children's Yale-Brown Obsessive Compulsive Scale (CY-BOCS) and CGI-Severity), the treatment group by time by moderator term was not statistically significant for any of the outcome/moderator pairs. We next examined the predictors of response across treatment groups, looking at changes in CY-BOCS total, CY-BOCS obsessions and compulsions, and CGI-Severity. However, we found that none of the predictors were related to longitudinal changes.

Conclusions: The results of the present study suggest that there are no factors that moderate DCS augmentation of CBT and that no baseline variables predict outcomes, as pediatric patients with OCD with varying initial symptom severity and baseline characteristics improved during CBT. This finding indicates that CBT is an effective treatment option for pediatric patients with OCD regardless of baseline characteristics and moderators, and that baseline characteristics cannot determine whether DCS augmentation is appropriate. However, because not all pediatric patients with OCD respond equally well to CBT, future research should look at the interactions between other baseline variables, such as positive family history of OCD, oppositional behavior, and poor treatment expectancy.

Keywords: Obsessive Compulsive Disorder, Cognitive Behavioral Therapy, D-Cycloserine, Predictors of Response, Moderators

Disclosure: Nothing to Disclose.

T88. Risk for Depression in Adolescent Girls: Associations With Brain Network Architecture

Amanda Guyer*, Rajpreet Chahal, Alison Hipwell, Kate Keenan, Erika Forbes

University of California, Davis, Davis, California, United States

Background: Adolescence is a time when connections among neural networks become stronger and more efficient, yet it is also a period of depression onset, particularly for girls. Experiencing depressive symptoms may compromise the fine-tuning of these neural networks. Examining brain connectomics may therefore provide novel insight into the emergence and maintenance of depression in girls. We examined the relationship between girls' depressive symptoms in adolescence and patterns of neural network topology in early adulthood using Diffusion Tensor Imaging (DTI). We hypothesized that higher levels of depressive symptoms earlier in adolescence would be associated with restricted network connectivity, reflected by higher average path length and decreased node strength.

Methods: Participants included 115 girls from the Pittsburgh Girls Study-Emotion neuroimaging sub-study. Girls' depressive symptoms were assessed with the Adolescent Symptom Inventory at 16 and 18 years of age. A diffusion tensor imaging scan was collected from the girls at age 18 and whole brain networks were reconstructed from this scan. AAL atlas-based brain regions were used as network nodes ($n = 94$) and tractography streamline counts as weighted and directed edges. Connectivity matrices were submitted to graph theoretical analyses of clustering, node strength, and path length and examined in relation to girls' depressive symptoms.

Results: Depressive symptoms at ages 16 and 18 were positively associated ($p < .0001$), consistent with past work. Depressive symptoms were positively correlated with fiber-weighted average path length ($p = .032$). Additionally, higher levels of depressive symptoms were predictive of lower overall node strength ($p = .02$) and decreased clustering ($p = .0025$).

Conclusions: Girls who experience depression at age 16 remain vulnerable to experiencing depression later in early adulthood. One mechanism underlying the continuity of depressive symptoms in girls may relate to specific aspects of their neural structural connectivity. Future work should consider the interplay of depressive symptoms and neural network characteristics at multiple time points in development.

Keywords: Adolescent Depression, Diffusion Tensor Imaging (DTI), Women's Mental Health

Disclosure: Nothing to Disclose.

T89. Neurobiologically Derived Clusters Differentiate Youth Based on Internalizing Symptom Load

Anne Wheeler*, Rebecca Trossman, Sonja Stojanovski, Grace Jacobs, Laura Stefanik, Joseph Viviano, Aristotle Voineskos

SickKids Research Institute, Toronto, Canada

Background: The structure and function of the default mode network (DMN) has been associated with internalizing

disorders. Behavioural and neurobiological heterogeneity as well as the presence of nonlinear brain behaviour relationships likely contribute to inconsistent results in this literature.

Methods: Here we examine internalizing symptoms in relation to patterns of 1) DMN resting state functional connectivity, and 2) DMN region volumes, in a large community sample of children and youth (Philadelphia Neurodevelopmental Cohort; $N=713$). A data-driven clustering approach based on participant similarity network fusion is used to identify groups of individuals with similar DMN structure and function. Group differences in the number of symptoms and patterns of DMN region volumes and functional connectivity are described.

Results: Grouping similar participants based on DMN structure and function revealed three clusters that differed in mean number of internalizing symptoms. Functional connectivity was highest in the group with the highest symptom load, intermediate in the group with the lowest symptom load and lowest in the group with an intermediate symptom load. This pattern was particularly pronounced for functional connection between anterior and posterior DMN regions. DMN region volumes were largest in the group with the lowest symptom load. Linear relationships between number of symptoms and DMN structure and function were not significant, additionally, clustering participants based on a single imaging modality did not reveal any group differences in the mean number of internalizing symptoms.

Conclusions: By combining information about DMN structure and function from different imaging modalities we identify groups of individuals with similar DMN patterns that are associated with differing levels of symptoms of depression and anxiety. These relationships were not apparent when examining linear brain behaviour relationships or when clustering participants based on functional or structural DMN information alone. These groups may reflect distinct etiopathology related to internalizing symptoms.

Keywords: Internalizing Disorders, Default Mode Network, Clustering

Disclosure: Nothing to Disclose.

T90. Investigating Convergent Neural Pathways in Zebrafish Mutants of Autism Risk Genes

Catalina Sakai, Sundas Ijaz, Marcus Ghosh, Matthew State, Antonio Giraldez, Jason Rihel, Ellen Hoffman*

Yale University, New Haven, Connecticut, United States

Background: The goal of this research is to elucidate basic neurobiological mechanisms underlying autism spectrum disorders (ASD). By targeting 10 ASD-associated genes in zebrafish using CRISPR/Cas9, our objective is to identify convergent neural pathways across risk genes. Using high-throughput pharmaco-behavioral profiling, we aim to identify pharmacological candidates that selectively rescue mutant behavioral phenotypes, revealing neural pathways with relevance to ASD.

Methods: We generated zebrafish mutants of 10 autism risk genes, including Contactin Associated Protein-2 (CNTNAP2), which is linked to a syndrome of autism and epilepsy, as well as "high confidence" ASD risk genes ascertained by whole-exome sequencing, including Sodium channel, voltage gated, type II alpha subunit (SCN2A) and Chromodomain helicase DNA binding protein 8 (CHD8). We investigated the simultaneous development of excitatory and inhibitory neuronal populations in mutants. To identify neurochemical pathways that are disrupted in mutants, we used quantitative behavioral profiling to compare the behavioral fingerprints of each mutant to the profiles of wild-type fish exposed to >500 psychoactive agents. This approach allows us to identify neurochemical pathways that are disrupted in mutants and potential suppressors of mutant behavioral phenotype(s).

Results: We previously identified forebrain GABAergic deficits, increased susceptibility to drug-induced seizures, and nighttime hyperactivity in zebrafish mutants of cntnap2. Using pharmaco-behavioral profiling, we found that estrogens are significantly enriched in the top ranks of compounds that anti-correlate with the cntnap2 mutant behavioral fingerprint, and that the plant-derived estrogen, biochanin A, selectively rescues the cntnap2 mutant behavioral phenotype. Next, we identified points of phenotypic convergence and divergence across the behavioral profiles of mutants of the ASD risk genes: CHD8, CUL3, DYRK1A, GRIN2B, KATNAL2, POGZ, and TBR1. These mutants display rest-wake cycle behavioral phenotypes compared to background-matched wild-type larvae. First, we found that scn1lab mutants display forebrain GABAergic deficits, nighttime hyperactivity, and increased susceptibility to drug-induced seizures, similar to cntnap2, suggesting common pathways. scn1lab mutants also display decreased daytime activity. Second, we identified a robust and reproducible behavioral fingerprint in chd8 mutants consisting of decreased rest bout length. Third, we found that grin2b mutants display nighttime hyperactivity, while tbr1 mutants exhibit decreased activity at night and increased daytime activity. Finally, we identified the top psychoactive compounds that correlate and anti-correlate with each mutant behavioral fingerprint. We are currently conducting pharmaco-behavioral screens to identify compounds that selectively rescue these mutant behavioral phenotypes.

Conclusions: These studies highlight the potential of studying zebrafish mutants of multiple ASD risk genes to identify points of convergence resulting from gene disruption and pharmacological pathways with relevance to ASD.

Keywords: Autism Spectrum Disorder, Developmental Neuroscience, Genetics, Zebrafish, Pharmacology

Disclosure: Nothing to Disclose.

T91. Regional and Cell-Type Specific Reconfiguration of DNA Methylation Patterns During Normal Brain Development and in a Mouse Model of Mental Disorders

Margarita Behrens*, ChiYu Lai, Yupeng He, Junhao Li, Chongyuan Luo, Eran Mukamel, Joseph Ecker

The Salk Institute for Biological Studies, La Jolla, California, United States

Background: The essential role of the frontal cortex and hippocampus in behavior and cognition requires the coordinated interaction, via electrical and chemical signaling,

of multiple neuronal cell types and a diverse population of glial cells. Individual brain cells have unique roles within circuits that are defined both by their location and pattern of connections, as well as by their molecular identity. We previously showed that DNA methylation patterns in frontal-cortex neurons are highly dynamic during brain maturation, and that neurons specifically accumulate a substantial amount of non-CG methylation through early childhood and adolescence. This unique feature of neurons, which distinguishes them from other differentiated cell types including astrocytes, shows that the period between late gestation and early postnatal neural circuit maturation is accompanied within each neuron-type by a parallel process of large-scale molecular identity reconfiguration. Alteration in the formation of these patterns by changes in the maternal environment during gestation, such as that occurring during maternal immune activation, could lead to circuit rearrangements resembling those found in psychiatric disorders with a neurodevelopmental origin, such as autism and schizophrenia.

Methods: We have used nuclei isolation and whole genome bisulfite sequencing, as well as RNA-seq in cell-type specific nuclei to analyze DNA methylation patterns and transcriptional regulation throughout development on both frontal cortex and hippocampus, and compared these results to those obtained in the offspring of animals subjected to maternal immune activation (MIA) produced by exposure of pregnant dams to 20 mg/kg PolyI(:C) on E12.5.

Results: Our data suggests large changes in transcriptional patterns as neurons mature, and that these are affected in the adult offspring of Poly(I:C) treated females. We have found highly specific changes in DNA methylation patterns in the two brain regions during development, with pyramidal neurons from cortex and hippocampus proper showing high overall similarities, but striking differences in large-hypo methylated regions coinciding with region specific enhancers. Correlation analyses showed enrichment in developmental differentially-methylated regions in the offspring from Poly (I:C) treated pregnant dams, suggesting that maternal immune activation produces an alteration in the normal methylation program in neurons.

Conclusions: The period of synaptogenesis and neural circuit maturation is accompanied within each neuron-type by a parallel process of large-scale molecular identity reconfiguration. This process differs in two brain regions highly involved in cognition, the frontal cortex and hippocampus, during the maturation period encompassing late embryonic to early postnatal at the neuron-type level. Alterations in the program of DNA methylation during late gestation/early postnatal development, through maternal immune activation may lead to profound changes in transcription and brain development, potentially linking disruption of the epigenome with neurodevelopmental disorders such as autism and schizophrenia.

Keywords: Brain Development, Neuronal Epigenome, Maternal Immune Activation

Disclosure: Nothing to Disclose.

T92. Postnatal Exposure of Valproic Acid as one of Potential Models of Autism Spectrum Disorders

Konstantin Yenkoyan*, Katarine Fereshetyan

Yerevan State Medical University, Yerevan, Armenia

Background: Autism Spectrum Disorders (ASD) are complex neurodevelopmental disorders characterized by repetitive behavior impaired verbal and nonverbal communication. Valproic acid (VPA) is known as an antiepileptic drug with teratogenic effect and is used for animal models of ASD. Recent studies show that VPA exposure causes increasing of neonate's glutamate decarboxylase and decreasing of GABA transaminase activities. Both of these effects lead to the elevation of GABA concentration in the brain. GABA demonstrates excitatory effect at the late embryonic and early postnatal stages of life, thus regulates a variety of different developmental processes from cell proliferation, migration, differentiation, synapse maturation and cell death. Thus, dysfunction of the GABAergic signaling on perinatal period may leads to an excitatory/inhibitory unbalance in neuronal circuits, which can manifest through behavioral alterations like observed in ASD patients. The aim of the present study was to observe the neurobehavioral development of newborn rats treated with VPA.

Methods: Rats received intraperitoneal (i.p.) injections of NaVPA 200 mg/kg and saline vehicle at postnatal days 5-12 (P5-12). Dams were housed individually and allowed to raise their litters until weaning on postnatal day 23. Rats' body mass were monitored during P5-25. On P5-9 the pups were tested for surface righting reflex activity. Negative geotropism was tested on postnatal days 13-19. On P25 the elevated plus maze test was carried out. Pups were placed in the center of the maze, facing an open arm. The total entries into open arms and the time spent in the open arms were measured. On P26 pain sensitivity was assessed using hot-plate test. The hot plate was set to a temperature of $50 \pm 0.5°$ C and the latency to lick hind paw was measured.

Results: To examine pup's normal physical development we monitored eye opening time and body weight between control and VPA treated groups during the first 20 days of life. There are no significant differences in eye opening days (on day P13-16, $p < 0.05$, $n = 20$ and $n = 15$ for CNR and VPA treated groups respectively). Analysis of pups' body weight data revealed a significant increase, VPA treated pups have higher body weights than controls during the development period P5-20, on P21,22 the body weight of both of the groups was equal ($p < 0.001$).

An elevated plus-maze test was performed to assess anxiety-related behavior. Each session lasted for five minutes. The number of entries into the open arm and the time spent in the open arm were both increase in the VPA exposed animals than the control group ($M = 76.6$, $SD \pm 36.9$, $M = 133$, $SD \pm 30.2$, $p < 0.001$ for CNR and VPA treated groups respectively). These results suggest that early postnatal VPA exposures show an anxiolytic effect.

VPA treated pups exhibited significantly higher latency to lick hind paw than control ($M = 21.7$, $SD \pm 9.3$, $M = 30.2$, $SD \pm 7.7$, $p < 0.05$).

Conclusions: The goal of the present study was to identify several behavioral characteristics in rats postnatally treated with valproate. We conclude that this model does mimic many of the features seen in individuals with ASD.

Keywords: Autistic Spectrum Disorders, Valproic Acid, Behavior
Disclosure: Nothing to Disclose.

T93. Adolescent Exuberant Local Inputs Onto a Frontal Top-Down Cortical Projection Drive Adult Attentional Behavior

Elisa Nabel, Yury Garkun, Giulia Taccheri, Sarah Lopez, Michael Demars, Hirofumi Morishita*

Icahn School of Medicine at Mount Sinai, New York, New York, United States

Background: Long-range connectivity between distal cortical regions develops across adolescence into adulthood to enable complex cognitive processes such as attention. Disruptions in long-range connectivity from frontal cortex increasingly identified in psychiatric and neurodevelopmental disorders emerge following adolescence, but how adolescent development contributes to long-range connectivity remains unknown. In the visual system, frontal top-down cortical neurons in the anterior cingulate cortex (ACA) directly project back to the visual cortex (VIS) to modulate visual processing, a hallmark component of visual attention. Here we aim to establish adolescent contributions to long-range top-down cortical circuit maturation in murine frontal cortex and its impact on cognitive behavior later in life.
Methods: To assess changes in synaptic drive onto top-down ACA->VIS neurons occur between adolescence and adulthood, we performed projection-specific whole-cell patch clamp recordings from top-down projection neurons visualized by fluorescence retrobeads injected in VIS in slice. To examine developmental structural changes of top-down ACA->VIS neurons, we analyzed dendritic spine density and morphology from fluorescently visualized dendritic spines by injecting a GFP encoding retrograde adeno-associated virus (AAV) vector into VIS. To detect brain regions contributing inputs to ACA->VIS projection neurons, we employed rabies mediated monosynaptic input mapping, and quantified if input numbers from these brain regions changed across development. To examine the causal contribution of top-down projection neuron activity during development on attentional behavior, we introduced inhibitory designer receptor exclusively activated by a designer drug (DREADD) into top-down circuits using an intersectional Cre-dependent viral approach, and suppressed top-down neuron activity through Clozapine-N-oxide administration only during adolescence, followed by adult testing of visual attention by the five-choice serial reaction time task, which assays sustained visual attention by requiring mice to maintain divided attention across 5 touchscreens for the presentation of a brief light stimulus that must be nose-poked to release a reward. Structural consequence of adolescent chemogenetic manipulation was assessed by examining adult dendritic spine.
Results: Patch-clamp recordings from frontal top-down projection neurons to visual cortex showed significantly greater mEPSC frequency in adolescents compared to adults with no significant changes in mIPSC, suggesting greater excitatory synaptic drive during adolescence. Consistently, at the structure level, dendritic spine analysis revealed greater total dendritic spine density of apical dendrites during adolescence driven by an adult decrease of thin spines, without an adult increase in mature stubby of mushroom spines. Rabies mediated mapping of monosynaptic input onto ACA->VIS projection neurons further demonstrated that top-down ACA->VIS neurons are more extensively connected to local networks in frontal cortex during adolescence than adulthood. Further, loss of this local network connectivity shifts top-down circuits toward greater distal connectivity into adulthood as are pruned and the relative structural weight of distal inputs increases. Strikingly, chemogenetic perturbation of top-down ACA->VIS projection neuron activity selectively during adolescent window of heightened local excitatory drive produced visual attentional behavior deficits accompanied by reduced dendritic spines in adulthood. Collectively, these data propose that adolescent top-down ACA->VIS projection activity is required to accrue an adequate pool of dendritic spines to maintain circuitry required to support adult visual attentional behavior.
Conclusions: Our study identified adolescence as a key developmental stage of exuberant local excitatory drive onto a top-down frontal cortex projection neurons. Transient perturbation of top-down neuron activity at this critical period of local connectivity revealed long-lasting structural and behavioral deficits in adulthood. Collectively, our data suggest that activity-dependent refinement of local inputs during adolescence is essential for a frontal top-down projection to establish attention in adulthood. This developmental process may be essential to fine-tune local computations in frontal cortex to effectively control distal sensory cortical region. Long-range functional deficits observed in humans may relate to aberrant development during this essential period of heightened excitatory drive. Heterogeneous high-risk gene variation or environmental insults at this critical developmental stage could converge on failures of proper circuit rewiring and thus produce common functional deficits—as is observed across attention deficits in autism, schizophrenia, and ADHD. Clinical approaches that protect and elicit excitatory drive during this adolescent critical period could ameliorate visual attention deficits in neurodevelopmental disorders, further, interventions promoting activity-dependent spine formation may have therapeutic value for adult deficits. Interrogating these possibilities in mouse models carrying these circuit deficits would provide valuable pre-clinical insight.
Keywords: Attention, Anterior Cingulate Cortex, Top-Down Control, Adolescence, Dendritic Spine
Disclosure: Nothing to Disclose.

T94. Deficits in Docosahexaenoic Acid Accrual During Peri-Adolescent Development Reduce Rat Forebrain White Matter Microstructural Integrity: Dissociation From Neuroinflammation

Robert McNamara*, Ruth Asch, Jennifer Schurdak, Bart Peters, Diana Lindquist

University of Cincinnati College of Medicine, Cincinnati, Ohio, United States

Background: Mood and psychotic disorders are associated with deficits in the omega-3 polyunsaturated fatty acid

docosahexaenoic acid (DHA), elevated pro-inflammatory signaling, and reductions in forebrain white matter integrity (WMI) measured by diffusion tensor imaging (DTI). The present study determined the effects of altering brain DHA accrual during adolescence on WMI in rat brain by DTI. To assess a potential mediating role of inflammation, we investigated pro-inflammatory markers in plasma (IL-1beta, IL-6, C-reactive protein, CRP), and cortical expression of IL-1beta and CD11b, a marker of microglial activation. We additionally examined myelin basic protein (MBP) expression as a central measure of myelination. Based on extant evidence, our prediction was that DHA levels would be positively associated with WMI, and that this effect would be associated with peripheral and central markers of pro-inflammatory signaling.

Methods: During peri-adolescent development male rats were fed a diet deficient in n-3 fatty acids (DEF, $n = 20$), a fish oil-fortified diet containing preformed DHA (FO, $n = 20$), or a control diet (CON, $n = 20$). In adulthood (P90), DTI scans were performed using a 7T Bruker Biospec system (Bruker BioSpin, Ettlingen, Germany). Indices of brain WMI were assessed using voxelwise tract-based spatial statistics and corrected for multiple comparisons ($p \leq 0.05$ corrected). Postmortem fatty acid composition of the forebrain was determined by gas chromatography, and peripheral (IL-1beta, IL-6, CRP) and forebrain (IL-1beta, CD11b) pro-inflammatory markers as well as MBP expression were determined.

Results: Compared with CON rats, forebrain DHA levels were lower in DEF rats (-30%, $p \leq 0.0001$) and higher in FO rats (+8%, $p = 0.01$). Compared with CON rats, DEF rats exhibited greater radial diffusivity (RD) and mean diffusivity (MD) in the right external capsule, and greater axial diffusivity (AD) in the corpus callosum genu and left external capsule. DEF rats also exhibited greater RD in the right external capsule compared with FO rats. Forebrain MBP mRNA expression did not differ between groups. Central (IL-1beta) and peripheral (IL-1beta, IL-6) pro-inflammatory markers were lower in FO rats, but were not different in DEF rats, and both FO and DEF rats exhibited lower CRP levels. CD11b, a marker of microglial activation, did not differ between groups.

Conclusions: Rat brain DHA accrual during adolescence impacts WMI in the adult rat forebrain. Specifically, deficits in brain DHA accrual were associated with right-lateralized reductions in RD and MD in external capsule WM, a pattern that is consistent with demyelination or dysmyelination, and increased AD in the left corpus callosum and external capsule which may reflect a perturbation in axonal pruning. Abnormalities in WMI in DHA-deficient rats could not be attributed to elevated peripheral or central pro-inflammatory signaling. These preclinical findings add to a growing body of translational evidence implicating omega-3 polyunsaturated fatty acids in WMI, and suggest that the DHA deficits and reduced WMI observed in patients with mood and psychotic disorders may be inter-related phenomena.

Keywords: Diffusion Tensor Imaging, Inflammation, Omega-3 Fatty Acids

Disclosure: Nothing to Disclose.

T95. Reduced Suicidality and Enhanced Neuronal Integrity With High Frequency Repetitive Transcranial Magnetic Stimulation Treatment in Adolescents

Paul Croarkin*, Charles Lewis, Hang Joon Jo, Deniz Doruk Camsari, Jennifer Vande Voort, Kathryn Schak, John Port

Mayo Clinic, Rochester, Minnesota, United States

Background: Suicide is a leading cause of death in adolescents and young adults worldwide. Despite considerable research efforts, targeted treatments for suicidality in adolescents are lacking. Recent work with accelerated repetitive transcranial magnetic stimulation (rTMS) protocols in adults support the safety, tolerability, and potential effects on decreasing measures of suicidality. However, very little is known about the effect of standard high frequency rTMS on suicidality in adults and no prior studies have examined this in adolescents. This project sought to examine the possible effect of rTMS on adolescent suicidality in an acute course of high frequency rTMS.

Methods: Data were pooled from 3 prior protocols providing 6 weeks of open-label rTMS treatment for adolescents (aged 13-19 years) with treatment resistant depression. All participants ($n = 20$) were outpatients, taking antidepressant medication, and rTMS was provided as adjunctive treatment. The rTMS sessions consisted of 10 Hz, 120% motor threshold treatment delivered to the left dorsolateral prefrontal cortex (LDLPFC), with 4 second stimulus trains, 26 second intertrain intervals, and 3,000 magnetic pulses per session. Participants received 30 sessions of rTMS over 6-8 weeks. Suicidality was assessed at baseline, after 10 treatments, after 20 treatments, posttreatment, and at a 6-months follow-up visit. Outcome measures of suicidality included the Columbia Suicide Severity Rating Scale (C-SSRS), item 13 "Suicidality" on the Children's Depression Rating Scale, Revised (CDRS-R), and item 13 "Suicidal Ideation" on the Quick Inventory of Depressive Symptomatology-Adolescent 17 item Self Report Form (QIDS-A17-SR). As depression symptom severity and suicidality may be colinear, we examined the potential correlations among changes in depressive symptom severity and suicidality over the course of 6 weeks of acute rTMS. A subset of participants ($n = 7$) underwent proton magnetic resonance spectroscopy of the LDPFC and anterior cingulate cortex (ACC) at baseline and posttreatment to examine possible clinical correlates with changes in glutamate metabolites or N-acetylaspartate (NAA) that were corrected to cerebrospinal fluid.

Results: Three participants dropped out of treatment early. One participant became suicidal during the first week of treatment and was hospitalized. This suicidal ideation was characterized as unrelated to investigational rTMS. One participant did not tolerate the first session of rTMS and another withdrew from treatment based on the inconvenience of daily rTMS sessions. The remainder or participants ($n = 17$) completed at least 25 sessions of rTMS. There were no suicide attempts during the acute treatment course. Suicidality as assessed by the C-SSRS improved from baseline to posttreatment ($p = 0.02$, Wilcoxon Signed Rank Test) but not from baseline to 6-month follow-up ($p = 0.1$, Wilcoxon

Signed Rank Test). Suicidality as assessed by item 13 of the CDRS-R demonstrated improvement from baseline to posttreatment ($p = 0.004$) and at 6-month follow-up ($p = 0.01$). Changes in baseline to posttreatment depression severity as assessed with the CDRS-R and QIDS-A17-SR did not demonstrate significant correlations with changes in suicidality as assessed with individual suicidality items and the C-SSRS (all $p > 0.25$). There were no associations among changes in objective measures of suicidality and changes in glutamate levels in the DLPFC or ACC. However, group level change in item 13 of the CDRS-R from baseline to posttreatment demonstrated a significant negative correlation with group level change of [NAA] in the anterior cingulate cortex (Spearman's Rho = -0.83, $p = 0.04$).

Conclusions: Suicidality in adolescents and young adults is a pervasive public health challenge with few targeted treatments. To our knowledge this is the first examination of the clinical impact of high frequency rTMS on adolescent suicidality. These preliminary data suggest that improvements in suicidality with high frequency rTMS were associated with enhanced neuronal integrity. Interpretation of the present findings must be placed in the context of the limitations of an open trial of high frequency rTMS and a relatively small sample size. Although rTMS appears to be safe and tolerable in adolescents, future work should focus on decreasing the time burden for suicidal participants and further development of target engagement biomarkers.

Keywords: Repetitive Transcranial Magnetic Stimulation, Suicidality, Adolescents, N-acetylaspartate, Magnetic Resonance Spectroscopy

Disclosure: Part 4: Neuronetics, Grant, Assurex, Grant, Pfizer, Inc., Grant.

T96. An in Vivo Study to Understand CNS Fluid Distribution and Target Engagement Following IV Administration of BIIB076, an Anti-Tau Therapeutic, in Cynomolgus Monkeys

Danielle Graham*, Weiping Chen, Julie Czerkowicz, Anne Cheung, Carrie Wager, Paul Weinreb, Phillipp von Rosenstiel, Kumar Kandadi Muralidharan

Biogen, Cambridge, Massachusetts, United States

Background: BIIB076 (6C5 huIgG1/λ) is a human monoclonal antibody with the potential to be a disease-modifying treatment for various tauopathies including Alzheimer's disease (AD). BIIB076 binds with subnanomolar affinity to human and cynomolgus monkey recombinant tau. In addition, BIIB076 is known to bind monomeric and preformed fibrillar tau, human brain tau isolated from both healthy individuals and AD patients, as well as cerebrospinal fluid (CSF) tau in healthy individuals and AD subjections. The primary objective of this experiment was to measure antibody concentrations at the desired site of action (interstitial fluid, ISF). A secondary objective of this experiment was to understand the PKPD relationship observed in ISF relative to ventricular CSF (LV CSF) and lumbar CSF (L-CSF).

Methods: Cynomolgus monkeys 2-4 years of age underwent cranial surgery for placement of guide cannula in the hippocampus region. Following a two-week recovery period, animals received a high dose intravenous infusion of BIIB076 on study day 0. Blood, CSF, and interstitial fluid (ISF) samples were collected at pre-specified time points to measure drug and tau levels. Serum, CSF, and ISF drug concentrations were measured with an anti-idiotype immunoassay (ELISA) incorporating anti-human antibodies. A highly sensitive, novel bead-based immunoassay was used to measure plasma, CSF, and ISF tau levels. Both free tau (not bound to BIIB076) and total tau (bound and unbound) were measured.

Results: Pharmacokinetic data and analysis indicates an IV infusion of BIIB076 produces proportional drug concentrations in serum and CSF. Once steady state concentrations are achieved the ratio of AUC over 8 hours between CSF and serum is estimated to be 0.1%. The concentration of drug measured in the ISF is greater than CSF and enters ISF more rapidly than ventricular CSF. Pharmacodynamic analysis indicates a single high dose of BIIB076 will reduce free tau in all three compartments. A single high dose administration of BIIB076 reduced free tau in the LV CSF and hippocampal ISF by roughly 60% and 90%, respectively. Total tau levels remained unchanged in the lumbar CSF, but were roughly reduced by 85% in the ISF and 50% in LV CSF.

Conclusions: These data confirm BIIB076 demonstrates a measurable PKPD profile in CSF. More importantly, this is the first study to report that an antibody therapeutic administered IV can reach the brain interstitial fluid at significant concentrations. In addition, these demonstrate target engagement (reduction in free tau) at the site of action (ISF) and confirm that lumbar CSF tau will provide a useful endpoint to demonstrate biological activity in clinical trials.

Keywords: Tau, CNS Antibody Delivery, Cerebrospinal Fluid, CSF Biomarkers, Therapeutics

Disclosure: Part 1: Biogen, Employee, Cerecor, Consultant, Spouse, **Part 2:** Biogen, Employee, **Part 3:** Biogen, Employee, **Part 5:** Biogen, Employee.

T97. Clathrin Nanoparticles Efficiently Deliver BDNF to the Hippocampus, Enhance Neurogenesis and Synaptogenesis and Reverse Memory Deficits in a Mouse Model of HIV-Tat Neurotoxicity

Gordana Vitaliano*, Jae Kim, Christopher Adam, Jay McLaughlin, Marc Kaufman, Franco Vitaliano

Harvard Medical School, Belmont, Massachusetts, United States

Background: Advances in treatment of neurodegenerative disorders have been made by administering brain derived neurotrophic factor (BDNF) directly to the brain, or by using drugs that increase BDNF indirectly. BDNF promotes neuroregeneration and restores brain functions, but cannot easily cross an intact blood brain barrier (BBB) or diffuse within the brain, and it is unstable in the blood or when given orally. The goal of this effort was to use our clathrin nanotechnology (Vitaliano at al. 2012) to produce stable BDNF-nanoparticles (NPs) that can efficiently target brain tropomyosin receptor kinase B (TrkB) receptors, and reverse neurotoxic effects of HIV transactivator of transcription (tat) protein implicated in HIV-Associated Neurocognitive Disorder (HAND).

Methods: A nanoparticle was constructed by conjugating one molecule of BDNF to the clathrin protein heavy chain via polyethylene glycol (PEG). Nanoparticle size, uniformity and purity, were determined by Dynamic Light Scattering (DLS), electron microscopy and SDS-PAGE respectively. GT-tg bigenic mice were treated daily for 7 days with saline (tat-) or doxycycline (100 mg/kg/d i.p.) that induces tat expression (tat+). Concurrently, animals received daily intranasally either NPs (0.3 mg/kg of BDNF with 2.4 mg/kg of clathrin); BDNF; clathrin; or saline (40µl). Subsequently, NP or saline treated tat+ mice were tested with Barnes maze and Novel Object Recognition (NOR) tests. For immunohistochemistry, mice also received Bromodeoxyuridine (BrdU 50mg/kg, Q12h i.p.) on days 1 and 2, and were sacrificed on days 7 or 14 of dox/saline administration. Neurogenesis and newborn cell proliferation and survival were determined with Doublecortin (DCX), Ki67 and BrdU antibodies respectively. For Western Blot (WB) analyses, hippocampi were removed and processed on the 4th day of dox/saline administration, and BDNF concentrations and signaling were analyzed.
Results: WB and microscopic analyses of hippocampal brain regions confirmed delivery of BDNF-clathrin NPs. First, NPs reversed BDNF signaling deficits in tat + animals by significantly increasing hippocampal levels of mature-BDNF ($p<0.007$), pro-BDNF ($p<0.01$), pAKT ($p<0.01$), and Akt ($p<0.002$) in NP treated mice compared to mice that didn't receive NPs. Second, NPs enhanced cell survival and proliferation, and doubled the density of young neurons in the hippocampus. BrdU+ ($p<0.0002$), Ki67+ ($p<0.002$) and DCX+ ($p<0.001$) cell densities significantly increased in the granule cell layer of dentate gyrus in NP treated tat+ mice, compared to saline treated tat+ or tat- mice. Third, NPs improved synaptogenesis in the hippocampus by significantly increasing synaptophysin levels ($p<0.006$) in NP treated tat+ mice vs. saline treated tat+ or tat- mice. Finally, hippocampal-based memory acquisition ($p<0.04$) and flexibility ($p<0.004$), and novel object recognition ($p<0.016$) significantly improved in NP vs. saline treated tat+ mice.
Conclusions: NPs bypassed the BBB, doubled BDNF levels, enhanced hippocampal cell proliferation, survival, neurogenesis and synaptogenesis, improved memory, and reversed neurotoxic effects of tat protein in GT-tg mice. Hence, clathrin provides a highly efficient nanoplatform for delivery of BDNF to the brain. This noninvasive nanotechnology may be able to enhance neuronal regeneration and plasticity, and restore brain functions more quickly and completely than existing treatment methods. Therefore, it has the potential to become a powerful tool in regenerative medicine and in the future, may lead to the development of diagnostic imaging tools, targeted delivery systems, and repair platforms (Vitaliano, 2016).
Keywords: Neurotechnology, BDNF Delivery, Clathrin Nanoparticles, HIV-Associated Neurocognitive Disorder, GT-tg Mouse Model
Disclosure: Part 1: EXQOR Technology Inc., Stock / Equity, Self/Spouse, **Part 4:** NIH and EXQOR Tech. Inc., Grant.

T98. Synaptic Changes in the OFC With Long-Term Obesogenic Diet

Benjamin Lau, Brittany Ambrose, Stephanie Borgland*

Hotchkiss Brain Institute, University of Calgary, Calgary, Canada

Background: The orbitofrontal cortex (OFC) receives sensory information about food and integrates these signals with expected outcomes. Thus, the OFC registers the current value of foods and updates actions based on this information. OFC lesions in animals show a lack of food devaluation. Interestingly, obese humans and rats fed a cafeteria diet have impaired devaluation of food rewards, implicating a potential obesity-induced dysfunction the OFC.
Methods: Rats were given restricted (1h /day), extended (23h/day) or no (chow only) access to a cafeteria diet. Whole cell patch clamp electrophysiology was used to assess alterations in local inhibitory synaptic transmission onto pyramidal neurons.
Results: Rats became obese after 40 days of extended, but not restricted access to a cafeteria diet. OFC pyramidal neurons from rats with extended access to a cafeteria diet had decreased inhibitory input partially due to an increase in endocannabinoid signaling at inhibitory synapses onto pyramidal neurons. Furthermore, rats with extended access to a cafeteria diet exhibited increased endocannabinoid tone due to altered group 1 mGluR signaling.
Conclusions: Taken together, these data suggest that cellular adaptations in the lateral OFC are associated with extended but not restricted access to a cafeteria diet. Thus, obesity can decrease inhibitory input to OFC pyramidal neurons possibly underlying impairments in food devaluation.
Keywords: Lateral Orbitofrontal Cortex, Diet Induced Obesity, Synaptic Plasticity, Endocannabinoid
Disclosure: Nothing to Disclose.

T99. Endocannabinoid Control of Gut-Brain Satiation Signaling

Donovan Argueta, Nicholas DiPatrizio*

University of California, Riverside, Riverside, California, United States

Background: Studies over the past several decades – including those from our laboratory – highlight the brain's ability to dynamically integrate peripheral signals to maintain energy balance. Our work suggests a novel gut-brain signaling pathway, mediated by the endocannabinoid (eCB) system, that controls food intake and becomes dysregulated in a rodent model of western diet-induced obesity (WDIO). We propose that gut-derived eCBs promote feeding and body weight gain in WDIO by a mechanism that includes inhibiting release of gut-derived satiation signals, which communicate with the brain via the vagus nerve to control feeding.
Methods: Separate groups of mice were fed a standard low-fat and sugar chow or high-fat and sugar "western diet" for 60 days. Behavioral analysis of feeding patterns was assessed for 24h following treatment with the peripherally-restricted neutral cannabinoid type 1 receptor (CB1R) antagonist,

AM6545, the cholecystokinin A receptor (CCK-A) antagonist, devazepide, or in combination. Levels of eCBs were quantified in intestinal tissues and blood via ultra performance liquid chromatography coupled to tandem mass spectrometry (UPLC/MS/MS) in standard and western diet fed mice. To evaluate whether eCB signaling at intestinal CB1Rs control the release of the gut-derived satiation peptide, cholecystokinin (CCK), levels of CCK-8 were quantified in blood plasma by standard ELISA following treatment with AM6545.

Results: When compared to standard diet-fed mice, WDIO mice displayed increases in daily caloric intake that resulted from larger meal sizes and rates of feeding, an effect met with increased levels of the eCB, 2-AG, in intestinal tissues and blood. Inhibition of this signaling with AM6545 completely normalized feeding patterns to levels found in control mice, and led to increased circulating levels of CCK-8. Furthermore, inhibiting CCK-A receptors with devazepide completely blocked the effects of AM6545 treatment on feeding.

Conclusions: Our studies suggest that hyperphagia associated with WDIO is driven by an enhancement in eCB signaling at CB1Rs in the gut, which is proposed to delay the release of CCK from enteroendocrine cells in the upper intestine during a meal and promote increased meal size and rate of feeding. Inhibition of peripheral CB1Rs may reduce feeding in WDIO by a mechanism that includes disinhibiting the release of CCK-8, which increases gut-brain vagal satiation signaling. Our studies suggest that the eCB system in the gut may be a novel pharmacological target for the safe treatment of obesity.

Keywords: Endocannabinoids, Gut-Brain Axis, Diet Induced Obesity, Vagus Nerve

Disclosure: Nothing to Disclose.

T100. A Dimensional Investigation of Emotional and Self-Regulatory Functioning in Relation to Eating Disorder Symptoms in a Transdiagnostic Sample

Jason Lavender*

University of California, San Diego, San Diego, California, United States

Background: Eating disorders (EDs) are serious and often protracted psychiatric illnesses. Despite the distinct symptoms of each ED, several core symptoms are found across multiple diagnoses (e.g., binge eating, compensatory behaviors). Theoretical models suggest that a number of shared biobehavioral mechanisms underlie the ED spectrum. Notably, disturbances in emotional and behavioral regulation are present across the range of EDs, and characterizing functioning in theoretically salient underlying domains may provide insights into onset, maintenance, and treatment. The current data are drawn from an NIMH-funded K23 that is focused on affective and cognitive dimensions of functioning in relation to ED symptoms across the ED spectrum, consistent with research initiatives focusing on shared processes across diagnoses (i.e., RDoC). Data collection for this project is currently ongoing. The purpose of this abstract was to examine dimensional relationships of affective and self-regulatory constructs with core ED symptoms (i.e., binge eating, purging, compulsive exercise).

Methods: Participants were 40 primarily Caucasian (95%) adult females (Age = 37.2 +/- 14.4; BMI = 29.5 +/- 8.9, range = 15.3-47.8). Inclusion criteria required clinically significant ED symptoms and related impairment (scoring more than 1 SD above community norms on the Eating Disorder Examination and above the established clinical cutoff of the Clinical Impairment Assessment). The non-disorder specific approach was chosen because many ED patients display subthreshold but clinically significant ED symptoms, and diagnostic shifts between EDs are common. This approach is also consistent with the dimensional focus of the K23 from which these data are drawn, as well as transdiagnostic research approaches to examining underlying domains of functioning. Participants completed dimensional ED measures, including the Eating Pathology Symptom Inventory (binge eating and purging severity) and the Compulsive Exercise Test (tendency to exercise in a driven/compelled manner). Affect-related measures were subscales from the Difficulties with Emotion Regulation Scale (difficulties with goal-directed behavior and impulse control when distressed, lack of adaptive emotion regulation skills, and nonacceptance of emotions) and the Eating Expectancy Inventory (belief that eating will provide emotional relief). Finally, self-regulatory measures included the Monetary Choice Questionnaire (index [k] of delay discounting) and the UPPS-P Impulsive Behaviors scale (tendency for disinhibition in response to aversive emotions [negative urgency] and pleasant emotions [positive urgency], sensation seeking, lack of perseverance, and lack of premeditation). Bivariate correlations were computed to characterize the dimensional associations of the affect and self-regulatory variables with ED symptoms. Given the preliminary nature of the present findings and the ongoing data collection, an alpha value of .05 was utilized for determining significance.

Results: Several positive associations were found for binge eating severity, including expectancy that eating will relieve negative affect ($r = .55$, $p < .001$), negative urgency ($r = .32$, $p = .045$), and lack of perseverance ($r = .35$, $p = .029$). Multiple variables were also positively related to purging severity, including nonacceptance of emotions ($r = .38$, $p = .015$), difficulty with behavioral control when distressed ($r = .38$, $p = .015$), positive urgency ($r = .45$, $p = .003$), sensation seeking ($r = .36$, $p = .021$), and lack of premeditation ($r = .36$, $p = .021$). Finally, for compulsive exercise severity, nonacceptance of emotions ($r = .36$, $p = .022$) was positively associated, whereas lack of perseverance ($r = -.39$, $p = .014$) and delay discounting ($r = -.38$, $p = .021$) were negatively associated. All significant effects were in the medium-to-large range.

Conclusions: Increasing behavioral, neurocognitive, and neurobiological findings suggest altered affective and self-regulatory functioning in EDs. The dimensional and transdiagnostic approach of this study (data collection ongoing) overcomes the limitations of investigations that focus only on a specific ED or on diagnostic comparisons, despite the overlapping nature of symptoms across EDs and the common occurrence of diagnostic shifts. Although preliminary, these findings demonstrate a unique pattern of associations of the affect- and self-regulatory-related variables with ED symptoms. Notably, more affect-related variables were associated with binge eating and purging, particularly the latter, these variables were less implicated in

the non-purging compensatory behavior of compulsive exercise. Further, the distinct pattern of associations across ED variables with respect to impulsivity domains suggests the possibility of different impulsive-compulsive processes underlying the various ED symptoms. These results from an ongoing research study suggest the need to investigate whether specific treatment elements may have utility for certain ED symptoms (e.g., pharmacologic or psychotherapeutic approaches to addressing affective disturbances, neurocognitive training for addressing self-regulatory control difficulties). Finally, future research investigating the neural activation underlying these processes in a transdiagnostic ED sample will be critical, and the present results suggest the importance of selecting tasks that will assess these distinct processes to best capture clinically salient relationships with specific ED symptoms.

Keywords: Eating Disorders, Emotion Regulation, Impulsivity, Self-Regulation

Disclosure: Nothing to Disclose.

T101. Calcium-Permeable AMPA Receptors in the Nucleus Accumbens Mediate Cue-Triggered Food-Seeking in Obesity-Prone Rats: Implications for Food Addiction

Carrie Ferrario*, Rifka Derman

University of Michigan Medical School, Ann Arbor, Michigan, United States

Background: Studies in humans suggest that obesity-susceptible individuals are more sensitive to the incentive motivational impact of sights, sounds, and smells that are associated with food (i.e., food cues). However, distinguishing cause from effect is difficult in human studies, and very little is known about the underlying mechanisms. In addition, although there is vibrant discussion about the concept of "food addiction", the degree to which similar vs. different neurobehavioral mechanisms underlie overconsumption of food vs. drug is poorly understood.

Methods: Here, we used selectively bred obesity-prone and obesity-resistant rats to examine basal differences in cue-triggered food-seeking using established Pavlovian-to-instrumental transfer (PIT) methods. This classic measure of the incentive motivational impact of food cues relies in part on activity in the Nucleus Accumbens (NAc), and there is indirect evidence for the involvement of AMPAR-mediated transmission in the expression of PIT. Therefore, we also determined whether AMPAR up-regulation is associated with PIT using BS3 crosslinking and Western blotting approaches, and determined how NAc AMPAR blockade affects the expression of PIT in obesity-prone and obesity-resistant groups using intra-NAc infusion of AMPAR selective antagonists, naspm and CNQX. All procedures were approved by the UM Institutional Animal Care and Use Committee, and were conducted in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals.

Results: We found that obesity-prone rats exhibit robust cue-triggered food-seeking (i.e., PIT) that was selectively mediated by CP-AMPARs in the NAc core. Additional biochemical data suggest that this is due in part to training-induced increases in CP-AMPAR surface expression in the NAc of obesity-prone rats. In contrast, in obesity-resistant rats the expression of PIT was weak and unreliable, and training did not increase NAc AMPAR surface expression.

Conclusions: Collectively, these data show that basal incentive motivational responses to food cues are stronger in obesity-susceptible populations prior to the development of obesity. This provides support to the idea that enhanced incentive-motivation may be a cause, rather than a consequence of obesity. In addition, they demonstrate a novel role for experience induced up-regulation of NAc CP-AMPARs in PIT, and suggest addiction-like enhancement of incentive motivation may contribute to the obesity epidemic, particularly in susceptible individuals.

Keywords: Striatum, AMPA Glutamate Receptors, Neuroplasticity, Reward, Pavlovian Conditioning

Disclosure: Nothing to Disclose.

T102. Increased Interoceptive Anticipatory Brain Response to Soft Touch in Women Remitted From Bulimia Nervosa

Christina Wierenga*, Amanda Bischoff-Grethe, Laura Berner, Alan Simmons, Miki Ogasawara, Laura Greathouse, Ursula Bailer, Martin Paulus, Walter Kaye

University of California, San Diego, San Diego, California, United States

Background: Bulimia nervosa (BN) is characterized by cyclical episodes of fasting, loss-of-control eating past fullness, and compensatory behaviors, like self-induced vomiting. Although the etiology of BN is poorly understood, altered fronto-striatal circuits associated with reduced self-regulatory control and reward prediction error have been reported in BN. Deficient interoception, or the sensing and integration of bodily state signals, may also contribute to BN pathophysiology, given that the hallmark dysregulated eating reflects a disruption of bodily homeostasis. Findings of altered neural response to anticipation of palatable food and to chemosensory (taste) and physical sensations (hunger/satiety) in BN support this notion. To determine whether BN is associated with a generalized (non-food-related) deficit in interoception, we examined brain response during anticipation and experience of pleasant affective touch. We predicted individuals with BN would demonstrate a mismatch between response to touch anticipation and experience in striatal and insula reward and interoception regions, consistent with an interoceptive prediction error.

Methods: Women remitted from BN (RBN; $N = 23$) and control women (CW; $N = 25$) performed a well-validated soft touch task during fMRI. Participants received slow brush strokes on the forearm and palm. Visual cues signaled upcoming stimulation to permit examination of both touch anticipation and experience. Women remitted from BN were studied to avoid the physical effects of dysregulated eating on brain response. To examine whether RBN and CW differ in brain response to anticipation or experience of pleasant touch, voxel-wise Group (RBN, CW) x Condition (anticipation, touch) x Location (palm, forearm) linear mixed effects analyses were computed in R within regions of interest associated with reward and interoception: the insula and striatum. Intrinsic smoothness was estimated using the

spatial autocorrelation function (acf) option in AFNI's 3dFWHMx. Minimum cluster sizes were calculated with AFNI's 3dClustSim in order to guard against false positives; a peak voxel of $p < 0.01$ with a cluster threshold of $\alpha < 0.05$ was required for significance. Post-hoc pairwise comparisons were false discovery rate corrected for multiple comparisons. Voxel-wise within-group Huber robust regressions were conducted to examine the relationship of harm avoidance and subjective visual analog scale (VAS) stroke pleasantness ratings with BOLD percent signal change for each condition and location. Only statistically significant clusters identified in the robust regression that overlapped with the significant cluster resulting from the Group x Condition interaction were retained, as overlapping clusters suggest the interaction may have been driven by the regression findings.

Results: Groups did not differ on VAS pleasantness ratings of touch. A Group (RBN, CW) x Condition (anticipation, touch) interaction was found in the right dorsal caudate. Post-hoc analyses showed both CW and RBN had increased activation during touch compared to anticipation, with RBN demonstrating greater anticipatory response than CW. For RBN, greater dorsal caudate palm anticipatory response was associated with lower pre- and post-scan pleasantness ratings, and increased harm avoidance. When only anticipation trials were included, a main effect of group (RBN > CW) was found in the left putamen and anterior and posterior insula. No group differences were detected for touch trials.

Conclusions: Elevated interoceptive anticipatory response in the striatum and insula in RBN, in the context of similar response to experienced touch between groups, suggests difficulty integrating environmental context (cues) with actual body-brain signals. This aberrant signaling might indicate altered afferent/efferent feedback processing that could disrupt learning from experience, serving to maintain disordered behavior. The association of greater anticipatory caudate response with reduced pleasantness ratings and elevated harm avoidance in RBN adds to a growing body of evidence linking negative experience with increased anticipatory reward response in BN, perhaps providing a mechanism for behavioral findings that bulimic episodes are often planned and preceded by negative affect. Altered interoception may have broader clinical relevance to BN as body-brain signaling guides decision-making, self-regulation, body image, and emotional experience, factors commonly disturbed in BN. Elucidating the neurocircuitry contributing to BN symptoms may directly inform new therapeutics, and results lend support to the development of interventions that address altered interoceptive expectancy in BN.

Keywords: Bulimia Nervosa, Interoception, Functional MRI (fMRI)

Disclosure: Nothing to Disclose.

T103. Dasotraline for Treatment of Adults With Moderate to Severe Binge-Eating Disorder: Effect on Behavioral Outcomes

Bradford Navia, James Hudson, Susan McElroy, Ling Deng, Seth Hopkins, Kenneth Koblan, Robert Goldman, Antony Loebel*

Sunovion Pharmaceuticals, Inc., Fort Lee, New Jersey, United States

Background: Binge-eating disorder (BED), the most common eating disorder in the US (lifetime prevalence, 2.8%), is associated with impairment in quality of life and functioning, high rates of medical comorbidity (e.g., obesity, metabolic syndrome), and increased healthcare utilization and costs. BED is typically associated with obsessive and dysphoric thoughts and compulsive behaviors relating to a range of eating and body image concerns. Few drugs are currently approved for BED. Dasotraline, a potent inhibitor of dopamine and norepinephrine transporters (IC50, 3 nM and 4 nM, respectively), and a weaker serotonin transporter inhibitor (IC50 15 nM), does not directly stimulate dopamine release, and has a PK profile characterized by slow absorption (tmax, 10-12 hrs), and a long elimination half-life (t½, 47-77 hrs) resulting in stable plasma concentrations over 24 hours with once-daily dosing. Its low abuse potential has been confirmed in a placebo- and methylphenidate-controlled abuse liability study in recreational stimulant users. In a recent study, dasotraline has demonstrated significant efficacy in treating adults with moderate-to-severe BED. We report here the results of secondary behavioral and psychological outcomes from this study.

Methods: Patients with moderate to severe BED, based on DSM-5 criteria, were randomized, double-blind, to 12 weeks of treatment with flexible doses of dasotraline (4, 6, and 8 mg/d), or placebo. The primary efficacy endpoint was number of binge eating days/week, assessed using a mixed model for repeated measures analysis. Secondary behavioral and functional outcome measures included the Yale-Brown Obsessive-Compulsive Scale Modified for Binge Eating (Y-BOCS-BE); and the Eating Disorder Examination Questionnaire Brief Version (EDE-Q7), which consists of a global score, and 3 subscale scores (dietary restraint, shape concern, and weight concern).

Results: 317 patients were randomized and received at least 1 dose of study drug (female, 84%; mean age, 38.2 years). LS mean (SE) reduction from baseline in the number of BE days per week was significantly greater for dasotraline vs. placebo at week 12 (-3.74 [0.12] vs. -2.75 [0.12]; $P < 0.0001$; effect size = 0.74). LS mean [SE] change from baseline to week 12 was significantly greater for the dasotraline vs. placebo on the Y-BOCS-BE total score (-17.05 [0.68] vs. -9.88 [0.65]; $P < 0.0001$; ES, 0.96), obsession subscale score (-8.32 [0.36] vs. -4.58 [0.34]; $P < 0.0001$; ES, 0.95), and compulsion subscale score (-8.69 [0.36] vs. -5.35 [0.34]; $P < 0.0001$; ES, 0.87); and was significantly greater on the EDE-Q7 global score (-0.85 [0.18] vs. -0.23 [0.11]; $P < 0.001$; ES, 0.49), dietary restraint subscale score (-0.55 [0.15] vs. +0.15 [0.14]; $P < 0.001$; ES, 0.44), shape concern subscale score (-0.93 [0.14] vs. -0.43 [0.13]; $P = 0.011$; ES, 0.33), and weight concern subscale score (-1.03 [0.14] vs. -0.44 [0.14]; $P < 0.01$; ES, 0.38).

Conclusions: In this double-blind, 12-week, placebo-controlled study, dasotraline, in doses of 4-8 mg/d, demonstrated significant improvement in secondary behavioral and psychological outcomes, including Y-BOCS measures of obsessions and compulsions associated with BED, EDE-Q7 measures of dietary restraint, and weight and shape concerns.

Clinicaltrials.gov number: NCT02564588

Keywords: Binge Eating Disorder, Dasotraline, Reuptake Inhibitor, CNS Clinical Trials
Disclosure: Part 1: Sunovion Pharmaceuticals, Inc., Employee, **Part 2:** Sunovion Pharmaceuticals, Inc., Employee, **Part 3:** Sunovion Pharmaceuticals, Inc., Employee, **Part 4:** Sunovion Pharmaceuticals, Inc., Employee, **Part 5:** Sunovion Pharmaceuticals, Inc., Employee.

T104. Abnormal Brain Activation and Connectivity in Body Dysmorphic Disorder and Anorexia Nervosa When Viewing Bodies

Francesca Morfini, Teena Moody, Gigi Cheng, Michael Strober, Jamie Feusner*

Semel Institute for Neuroscience & Human Behavior, Los Angeles, California, United States

Background: Individuals with anorexia nervosa (AN) and those with body dysmorphic disorder (BDD) experience distorted perception of their appearance and may experience abnormal perception of others' appearance. Clinical experience suggests that bodies might be more salient in AN than in BDD. However, both groups misestimate the size of others' bodies and rate them as lower in attractiveness than healthy controls (CON), and both groups have similar abnormal brain activation in visual systems when viewing others' faces, suggesting that similar abnormalities in visual processing may underlie their perceptual experiences. In addition, in AN studies have revealed abnormalities in brain regions including intraparietal lobule and amygdala during body processing and other studies have shown decreased resting state connectivity involving visual networks. However, no studies have directly compared brain activation or connectivity patterns in AN and BDD when viewing bodies. Our aim was to determine brain activation and connectivity patterns in AN and BDD compared with CON when viewing bodies. We used others' bodies rather than own bodies to reduce the confound of high emotional arousal, and to determine if general abnormalities in body processing are present. We examined the dorsal visual network and investigated associations with symptom severity. We hypothesized that both AN and BDD would have reduced activation, as seen previously in both groups for low-detail face stimuli, and reduced network connectivity in the dorsal visual network which is involved in global image processing.
Methods: We acquired functional magnetic resonance imaging data in 69 participants: 21 weight-restore AN (mean age 23.4 ± 3.4; 20F/1M), 24 BDD (mean age 23.8 ± 4.9; 23F/1M), and 24 CON (mean age 22.0 ± 4.6; 22F/2M). All were unmedicated, right-handed, and of equivalent age, sex, and level of education. While being scanned, participants matched photos of other's bodies and shapes as a control task. The stimuli were of different types: low spatial frequency (LSF), normal spatial frequency, or high spatial frequency.
Whole-brain BOLD fMRI and T1 structural MRI data were acquired with a 3-T Siemens TRIO scanner. BOLD data were analyzed with FSL. Image preprocessing included motion-artifact correction by an ICA denoising. Group level analysis was performed with FSL's MELODIC to obtain network components correlated with canonical networks (Smith et al., 2009). The component most correlated with the dorsal visual network was used in connectivity analyses. Data analysis used a block design and contrasted each stimulus type to a control task using the general linear model to assess activation patterns among groups and to extract eigenvalues. For connectivity analyses, we compared network coherence values from the ICA among groups using dual regression to create component-specific networks from subject-level spatial maps. We used F-tests ($p < 0.05$, corrected) to identify regions that differed among groups, and group pairwise t-tests thresholding at $p < 0.05$. We also explored correlations between activity/connectivity with EDE, BABS, BDD Y-BOCS scores.
Results: Activation:
BDD had lower activation than CON in the dorsal visual network as hypothesized (left superior parietal lobule, right precuneus, right lateral occipital cortex) when viewing LSF images. No significant differences between AN and CON were found in the dorsal visual network.
Connectivity:
Significant differences among groups were found in the dorsal visual network for BDD and AN compared to CON when viewing LSF images. As hypothesized, both BDD and AN had lower connectivity than CON. Specifically, BDD demonstrated lower connectivity in bilateral superior lateral occipital cortex, while AN showed lower activation predominantly in right lateral occipital cortex. BDD showed greater connectivity in the right cuneus.
Symptomatology:
There was a trend for a negative correlation between BDD Y-BOCS and brain activation eigenvalues in the dorsal visual network for LSF images (Pearson's $r = -0.36$, $p < 0.08$), with lower activity corresponding to greater symptom severity.
Conclusions: This is the first study to assess brain activation and connectivity patterns when viewing images of bodies in AN and BDD. Both groups exhibit similar patterns of abnormally reduced connectivity in the dorsal visual network when viewing images of bodies that convey low level of detail (LSF), although distinct patterns of activation.
Considering that the dorsal visual network plays a role in LSF processing, the hypo-activation and low connectivity found in BDD suggest a failure to properly engage and exchange inputs within the brain network responsible for encoding global features of visual stimuli. This may result in failure to contextualize visual details into an integrated global "template." From a phenomenological perspective, this could relate to the tendency to perceive small imperfections as more prominent than they are. Similarly, reduced connectivity in the dorsal visual network for AN suggests an overlapping but not identical phenotype since hypoactivation was not observed. Study results (using body, face and house stimuli) of a general tendency in BDD of insufficient activation for LSF information, with a similar pattern in AN, is consistent with a model of imbalance in global/local processing. Future studies are needed to probe relationships between abnormal activation/connectivity patterns and direct measures of body perception disturbances in these populations. Our results have clinical implications for identifying potential targets for novel treatment methods such as specific behavioral or perceptual retraining or brain stimulation to ameliorate abnormalities that may underlie perceptual distortions.

Keywords: Visual Information Processing, Anorexia Nervosa, Body Dysmorphic Disorder, Bodies, Connectivity
Disclosure: Nothing to Disclose.

T105. Altered Functioning of Cognitive Control Circuits Across Development in Obsessive-Compulsive Disorder

David Pagliaccio*, Marilyn Cyr, Sophie Schiff, Emily Sternberg, Martine Fontaine, Helen Simpson, Moira Rynn, Rachel Marsh

New York State Psychiatric Institute, New York, New York, United States

Background: Obsessive-compulsive disorder (OCD) is characterized by intrusive thoughts, images, or impulses and repetitive actions performed to reduce distress. These key features of OCD are thought to be due to a failure to inhibit one's thoughts or behaviors and prior work has implicated corresponding alterations in fronto-striatal control circuits. OCD tends to show a bimodal distribution in onsets with peaks in childhood and then again in early adulthood. While some findings suggest similarities in the pathophysiology of childhood vs. adult onset OCD, other work suggests that these may be phenomenologically distinct. For example, childhood onset OCD tends to show higher familial loading of OCD, to be more prevalent among boys, and to be more related to tic disorders than adult onset OCD. Yet, neuroimaging work has yet to examine whether the functioning of cognitive control circuits differs across children and adults with OCD, particularly as a function of age of onset.
Methods: Treatment-seeking children/adolescents (mean age = 12.15, SD = 3.04) and adults (mean age = 28.78, SD = 6.62) with OCD and matched healthy volunteers completed psychiatric and neuropsychological evaluation and a magnetic resonance imaging (MRI) session. OCD participants were free of Tic disorders and comorbid anxiety disorders. The MRI scan included high-resolution structural imaging and three runs of multi-band acquisition (TR = 850ms) during the Simon Incompatibility task. Acquisition and image preprocessing mirrored that of the Human Connectome Project. The Simon task requires ignoring a task-irrelevant stimulus feature when that conflicts with the task-relevant dimension. Reaction time and brain activity was examined comparing conflict trials and congruent trials during which the task-relevant and task-irrelevant features did not conflict.
Results: Preliminary analyses were conducted to examine group differences in task performance as a function of diagnostic status and age. Examining 20 healthy adults, 28 adults with OCD, 12 healthy children, and 25 children with OCD indicated a main effect of diagnostic status on the conflict effect (mean RT conflict – mean RT congruent, $F = 3.91$, $p = .05$) and a stronger group difference when examining this conflict effect on trials that follow a prior congruent trial, which has previously been showing to elicit greater conflict response ($F = 6.38$, $p = .01$). All participants responded slower to conflict trials, but this conflict effect was greater among OCD patients relative to controls. No significant main effects of age or age x diagnosis interactions were detected and age of OCD onset did not significantly predict the conflict effect in the adult OCD participants. Preliminary imaging findings suggested greater conflict-related activations in fronto-striatal and cingulo-opercular regions in OCD compared to control participants. On-going fMRI analyses will further examine these group differences and age effects on conflict-related activity within these circuits.
Conclusions: Preliminary results suggest that OCD patients experience more conflict on the Simon task than their healthy counterparts but that these group differences are unassociated with current age or age of OCD onset. These findings suggest that the ability to respond to conflict may mark the pathophysiology of OCD but is unrelated to differences between child- and adult-onset OCD, though this will be explored more thoroughly in on-going neuroimaging analyses.
Keywords: OCD, Obsessive-Compulsive Disorder (OCD), Functional MRI (fMRI), Brain Development, Cognitive Control
Disclosure: Nothing to Disclose.

T106. Disentangling the Role of Specific Orbitofrontal Circuits in Decision-Making

Stephanie Groman*, Colby Keistler, Jane Taylor

Yale University, New Haven, Connecticut, United States

Background: The orbitofrontal cortex (OFC) is critically involved in adaptive decision-making and OFC dysfunction has been linked to inflexible behavior in several mental disorders. The OFC sends dense projections to multiple subcortical targets and it is unknown whether distinct OFC circuits mediate different aspects of decision-making processes that may be involved in the pathophysiology of schizophrenia and addiction.
Methods: To determine the role of specific OFC circuits in decision-making, we used a novel viral approach combining a floxed diphtheria toxin receptor and a retrograde Cre-expressing virus to target OFC neurons that project selectively to the nucleus accumbens ($N = 20$) or the amygdala ($N = 20$). Rats were trained on a three-choice, probabilistic reversal-learning (PRL) task and then received an injection of saline or diphtheria toxin (DT; 30 ug/kg). Since rats do not express DT receptors, administration of DT causes selective ablation in those OFC projection neurons that express DT receptors. Decision-making was then reassessed in the PRL task.
Results: Ablation of OFC neurons projecting to the nucleus accumbens impaired the ability of rats to flexibly adjust their decision-making in the PRL by reducing the influence of negative outcomes on decision-making. In contrast, ablation of OFC neurons projecting to the amygdala improved PRL performance by increasing the influence of positive outcomes on future decision-making.
Conclusions: These data indicate that distinct OFC circuits mediate dissociable components of decision-making. By combining sophisticated viral approaches with translationally analogous behavioral assessments, these data suggest that decision-making may be a useful biomarker for understanding OFC pathophysiology in mental disorders.
Keywords: Reinforcement-based Decision-Making, Lateral Orbitofrontal Cortex, Computational Modeling, Cognitive Control, Reversal Learning
Disclosure: Nothing to Disclose.

T107. Corticostriatal Networks Reveal Multiple Levels of Cortical Output Integration

Wei Tang*, Suzanne Haber

Harvard Medical School, McLean Hospital, Belmont, Massachusetts, United States

Background: The dorsal anterior cingulate cortex (dACC) and the striatum are key structures that mediate higher brain functions, including emotion, motivation, higher cognition and executive control. Corticostriatal projections from the dorsolateral and ventromedial prefrontal cortex (dlPFC) & vmPFC) converge with those from the orbitofrontal cortex (OFC) in specific zones of the striatum, indicating crosstalk between these pathways. Recently we found a similar convergent zone in the dACC but with more extensive input patterns, including those from the dlPFC, the OFC, the vmPFC, the ventrolateral PFC (vlPFC) and the premotor cortex. Based on network theory, these convergent zones may serve as hubs to facilitate communication between functional modules via extensive connections. In this study, we investigate how the dACC hub is connected with the striatal hubs. The results have implications for how reward-driven and goal-directed functions may be integrated at different levels of the dACC-striatum network.
Methods: To identify the dACC hub, seven dACC subregions received bidirectional tracer injections. Labeled cells throughout the PFC areas were quantified using stereology. A dACC subregion received inputs from significantly more PFC areas compared to other dACC regions. The PFC-striatal projection areas were identified and hubs were defined by those areas receiving dense terminal fields from more than 5 PFC areas. We then compared the dACC projections to the striatum between those from the hub and non-hub regions of the dACC. We also compared how the hub and non-hub regions of the striatum receive projections from the dACC.
Results: The results showed a three-way connectivity pattern that links the PFC, the dACC and the striatum: (1) PFC projections to the striatum form scattered convergent zones in the mid-to-rostral caudate, putamen and ventral striatum. (2) The dACC hub projects to both the ventral striatum and the dorsomedial caudate, while the non-hub dACC have more focal terminal fields, terminating in either the dorsal or ventral striatum, but not both. (3) The terminal fields from each dACC subregion in the striatum, including those from the hub, overlap with the PFC terminals that also project to that part of the dACC. Importantly, the dACC hub projects to more PFC convergent zones in the striatum than do the non-hub dACC subregions.
Conclusions: These results showed different levels of functional integration across cortical areas. One level of integration is carried out by the striatum, where the output of different PFC areas crosstalk with each other in the convergent zones. Another level of integration is carried out by the dACC, where the PFC projections first converge onto the dACC hub, and then the hub carries the integrated information further to the striatum. This multi-level mechanism enables flexible integration of reward processing, learning and cognitive functions that are carried out by different subdivisions of the PFC. Thus, damage to their projection convergent zones in the striatum and/or the dACC may affect the degree of integration, leading to unbalanced behavior and psychiatric disorders.
Keywords: dACC, Striatum, Corticostriatal Networks
Disclosure: Nothing to Disclose.

T108. Maternal Immune Activation With a TLR7 Agonist Results in Sex-Specific Behavioral Alterations in Mice: Relevance to Neuropsychiatric Disorders

Galen Missig*, James Robbins, Emery Mokler, Christopher McDougle, William Carlezon

Harvard Medical School, Belmont, Massachusetts, United States

Background: Epidemiological evidence suggests that immune activation during pregnancy via infection or autoimmune disease is a risk factor for neuropsychiatric illness. Work from our lab and others has demonstrated that prenatal immune stimulation results in offspring that exhibit a behavioral phenotype characterized by decreased social behavior, increased anxiety-like behavior, and increased repetitive behavior. Previous maternal immune activation (MIA) protocols have primarily involved administration of agents to mimic infection that target subtypes 3 (i.e., poly I:C) and 4 (i.e., lipopolysaccharide) of the toll-like receptor (TLR) family, a class of receptor proteins that regulate innate immune responses. In this study, we examined the role of TLR7 in MIA, considering evidence that this receptor subtype is implicated in autoimmune-related inflammation and follows a developmentally regulated expression pattern in the brain.
Methods: We administered subcutaneous injections of the selective TLR-7 agonist imiquimod (IMQ, 5.0 mg/kg) or vehicle to timed pregnant dams (C57BL/6J mice) on embryonic days (E) 12, 14, and 16. In the first experiment, we assessed the offspring on a battery of behavioral tests over the course of 13 weeks that included pup ultrasonic vocalizations (USVs), open field, elevated plus maze, social approach, reciprocal social interaction, marble burying, self-grooming, and spontaneous alternations. In a second (parallel) experiment, we collected fetal brains 3 hours following E16 injections and examined changes in expression of several immune response-related transcripts.
Results: Mice treated with prenatal IMQ treatment exhibited overt behavioral phenotypes with sex-specific elements. In male mice only, IMQ treatment produced decreases in both anxiety-like behavior and in the number USVs emitted, but increases in self-grooming and a "conditional" hyperactivity that was evident only during social interaction. In female mice only, IMQ treatment produced increases in marble burying. In both sexes, IMQ treatment produced fragmented patterns of social interaction, with increases in the total number of interactions and decreases in the average length per interaction in both social approach and reciprocal interaction tests. In addition, IMQ-induced increases in interleukin-6 (IL-6) levels in maternal serum, as well as increases in expression of the chemokine receptor CXCR4 and glycogen synthase kinase 3-beta (GSK-3β) in brain, were observed in both sexes on E16.
Conclusions: Maternal immune activation with a TLR7 agonist produces a behavioral phenotype that is distinct from —and often inverse to—the phenotype seen with MIA

procedures that target other TLR subtypes. Several of the behavioral signs resemble the core features of bipolar disorder (BP) and attention deficit hyperactivity disorder (ADHD). Considered with the existing literature, our work suggests that selective activation of TLR subtypes can produce phenotypes that differ in important domains and, as such, this approach may enable new insights on the pathophysiology of neuropsychiatric illnesses.
Keywords: Maternal Immune Activation, Toll-Like Receptors (TLRs), Bipolar Disorder, Attention Deficit Hyperactivity Disorder
Disclosure: Nothing to Disclose.

T109. Role of Dopamine D2 Receptors in Impulsive Behavior

Sehyoun Yoon, Bokyeong Kim, Ryuichi Nakajima, Hyo Jin Lee, Yeon-Kyung Lee, June-Seek Choi, Bong-June Yoon, George J. Augustine, Ja-Hyun Baik*

Korea University, Seoul, Korea, Republic of (South Korea)

Background: 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. Dysfunctional dopaminergic neurotransmission, especially involving dopamine D2 receptor (D2R), has been proposed to be a mechanism underlying impulsivity.
Methods: We examined impulsive behavior in wild-type (WT) and D2R knockout (D2R -/-) mice by using a five-choice serial reaction time task (5-CSRTT).
Results: We observed that the absence of D2R increases impulsive behavior but also a deficit in attentional performance in mice.
Conclusions: We will present further 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.
Keywords: Impulsivity, Optogenetics, Dopaminergic System
Disclosure: Nothing to Disclose.

T110. Evaluating Translational Neurophysiological Measures to Improve Efficacy of Preclinical Therapeutic Target Discovery

David Gregg, James Cavanagh, Jared Young, Jonathan Brigman*

University of New Mexico Health Sciences Center, Albuquerque, New Mexico, United States

Background: There is a pressing need for translational tests of cognitive functioning that demonstrated consistent neurophysiological biomarkers across species. While preclinical studies provide numerous therapeutic targets for neuropsychiatric disorders, the failure to convert these to clinical treatments highlight that behavioral similarity alone without biomarkers of brain function is insufficient. As part of an ongoing project, we integrated touch-screen analog of tasks measuring specific Research Domain Criteria (RDoC) aspects with electroencephalography (EEG) in the mouse to determine neural activity directly comparable to human data being generated by our collaborators.
Methods: C57BL/6J mice were first trained to initiate and respond for reward to visual stimuli in a touch-sensitive screen. Following pre-training, mice were fitted with fixed EEG leads resting on the dura. After recovery, mice were trained on a series of behavioral tasks to RDoC-specific domains. These included the Five-Choice Continual Performance Task (5C-CPT) of cognitive control, Progressive Ratio Break-Point Task (PRBP) of effortful motivation, and a Probabilistic Discrimination Task (PrDT) of reward learning, all while EEG signal was recorded.
Results: Mice were able to successfully perform touch-screen variants of the tasks adapted for tethered EEG recording. During 5C-CPT performance mice successfully differentiated between target and non-target trials with EEG exhibiting similarly differential frontal event-related components corresponding to trial type. Mice tested on the PRBP exhibited break-points similar to those in lever-based tasks and frontal EEG signal was linked to variability in willingness to exert effort to obtain reward. When tested on different probabilities of reward by stimulus-set, cortical EEG signal exhibited differential responses to reward by reward difficulty level.
Conclusions: Here, we demonstrated the capability of utilizing identical methods for investigating rodent neuronal activity performing the same behavioral tasks that are available in rats and humans. By utilizing clinical EEG methodology we can parse aspects of cognitive control, effortful motivation, and reward learning. These techniques also provide important information regarding the validity rodent models to diseases in addition to the likelihood of translating drug-induced changes in performance across species.
Keywords: EEG Biomarkers, Touchscreen, Research Domain Criteria (RDoC)
Disclosure: Nothing to Disclose.

T111. Sex Differences in Time to Depression Remission Following Repeated Ketamine Infusions

Cristina Albott*, Paulo Shiroma, Christopher Erbes, Paul Thuras, Joseph Wels, Kelvin Lim

University of Minnesota, Minneapolis, Minnesota, United States

Background: Recent preclinical reports have identified sex differences in sensitivity to ketamine as well as differential efficacy of ketamine in rescuing stress-induced reduction of spine density in the medial prefrontal cortex. Literature in humans has noted sex differences in the prevalence and symptoms of depressive disorders and well as in antidepressant response rates. We present a post hoc analysis of an original study investigating sex differences in response and remission rates of depression and PTSD symptoms following serial ketamine infusions.

Methods: Fifteen individuals with treatment-resistant depression (TRD) and posttraumatic stress disorder (PTSD) received six IV infusions of 0.5 mg/kg ketamine over 40 minutes on a Monday-Wednesday-Friday schedule during a 12-day period. Primary outcome measures included the Montgomery-Asberg Depression Rating Scale (MADRS) and the PTSD-symptom Checklist (PCL-5). Secondary measures included response and remission criteria for depression symptoms as well as remission criterion for PTSD symptoms. Depression response was defined as $\geq 50\%$ improvement in pre-infusion/baseline depressive symptoms as measured by the Montgomery–Åsberg Depression Rating Scale (MADRS), and remission was defined as a MADRS score ≤ 9. PTSD remission was defined as a PCL-5 score < 33. Outcomes measures were collected prior to each infusion and 24-hours after each infusion. Subjects were followed weekly for 8 weeks after the sixth and final infusion.
Results: Of the fifteen individuals who completed all six infusions, five were women and ten were men. Fourteen achieved depression response criterion (93%), nine achieve remission of depression (60%), and twelve (80%) met remission criterion for PTSD. The study cohort consisted of five women (33%) and ten men (67%). Post-hoc analyses demonstrated no statistically significant difference in MADRS score or PCL5 score for women compared to men after completion of the 6 infusions. While the rates of remission for men and women were not significantly different, the time to remission differed significantly between sexes: 12.4 days for women compared to 6.9 days for men ($F = 5.552$, $df = 14$, $p = .035$). The difference in time to depression symptom response was not statistically significant between sexes (7.0 days for women and 5.1 for men). Additionally, there was no difference between men and women in time to remission of PTSD symptoms (2.4 days for women and 2.6 days for men).
Conclusions: This post-hoc analysis provides the first clinical evidence suggesting sex differences in remission rates for depression symptoms following repeated ketamine infusions. Interestingly, the current analysis suggests a trend inconsistent with preclinical literature insofar as women appear to be less sensitive to the antidepressant effects of serial ketamine infusions whereas female rats were more sensitive to the antidepressant effects of a single ketamine infusion. This post-hoc analysis warrants further study in a larger cohort in which putative biomarkers of processes mediating this difference (i.e. reproductive hormone levels) can also be assayed.
Keywords: Ketamine, Antidepressant, Sex Differences
Disclosure: Nothing to Disclose.

T112. A Longitudinal MRI-Study of the Effects of Lithium on Cortical Thickness and Brain Volume and its Association With Clinical Response in Bipolar Disorder

Sabrina Correa da Costa*, Marcus V. Zanetti, Rafael T. Souza, Maria C. Otaduy, Claudia C. Leite, Geraldo F. Busatto, Joao L. Quevedo, Wagner F. Gattaz, Jair C. Soares, Rodrigo Machado-Vieira

University of Texas Health Science Center at Houston, Houston, Texas, United States

Background: Background: Lifetime prevalence of bipolar disorder (BD) is 2.1% worldwide, with subthreshold forms affecting another 2.4%. Findings from structural magnetic resonance imaging studies suggest that some brain abnormalities may exist early in the course of illness and potentially predate illness onset, while others appear to develop with recurrent affective episodes. Lithium has demonstrated effects on modulating expression of neurotrophic factors, with potential to reverse dysfunctional cellular repair, reduction in brain volume, and cell atrophy and death. Recent data in animal models found that chronic lithium therapy induced significant increases in total brain volume and cortical gray matter, with long-lasting effects even after termination of therapy. Therefore, the present study aims to assess the effects of lithium therapy on cortical thickness and brain volume and its association with clinical response in medication-free BD subjects during acute depressive episode.
Methods: Methods: Twenty-six medication-free BD subjects during an acute depressive episode were included in this study. Patients were treated for 6 weeks with lithium monotherapy at therapeutic doses in an open-label trial. Measures of cortical thickness and brain volume using region of interest (ROI) MRI were performed pre- and post-treatment and analyzed using FreeSurfer.
Results: Results: significant cortical volume and thickness changes in specific brain areas were found after 6 weeks of lithium monotherapy. Positive association between anatomical changes in limbic areas with clinical response were also observed. More specifically, cortical volume and thickness were increased in the R cortex hemisphere (p 0.015), R cerebellum white matter (p 0.0457), R thalamus (p 0.0250), left inferior lateral ventricular area (p 0.024), Left hemisphere cortex (p 0.0059), CC anterior (p 0.0083), total cortical volume (p 0.004), total gray volume (p 0.003), and supra tentorial volume (p 0.026). Reduction of size of lateral, third and fourth ventricles were also found.
Conclusions: Conclusion: The specific neuroanatomical changes found in our study, mostly in the limbic areas, and its association with positive clinical outcomes highlight the potential role of neurotrophic and neuroprotective effects underlying the therapeutic actions of lithium in BD. It also highlights the relevance of these specific limbic areas as putative targets for novel therapies.
Keywords: Lithium, MRI, Bipolar Disorder
Disclosure: Nothing to Disclose.

T113. Association Between Molecular Markers of Aging With Early Life Stress and Psychiatric Disorders

Kathryn Ridout*, Stephanie Parade, Ronald Seifer, H-T Kao, Barbara Porton, Stevie Magnan, Lawrence Price, Audrey Tyrka

Brown University, Providence, Rhode Island, United States

Background: Maltreatment is a major public health problem impacting over 19.4 million children through abuse, poverty, or neglect that increases risk for a number of psychiatric disorders, including depression, anxiety, post-traumatic stress, and internalizing behaviors. Studies of adults with a history of childhood maltreatment provide evidence that biological markers of aging, telomere length and

mitochondrial DNA copy number (mtDNAcn) are impacted by this exposure and may be involved in the mechanisms linking maltreatment with increased disorder risk. There are a few studies in children suggesting that telomere length is shortened with maltreatment exposures, however these studies temporally examine telomeres 3-15 years after the maltreatment exposure. There have been no studies of maltreatment exposure and mtDNAcn in children. This study aimed to examine the impact of childhood maltreatment in children aged 3-5 on telomere length and mtDNAcn collected 6 months after the exposure and again 6 months after the initial collection.

Methods: Children aged 3-5 were identified through either the local child protective services agency ($n = 133$, maltreated) or a local pediatric medical clinic ($n = 117$, control). Structured record review was used to confirm maltreatment status. Home interviews were conducted to assess maltreatment history, other traumas, and life stressors at baseline. A composite adversity variable for total number exposures was calculated. The CBCL and DIPA were administered for caregiver report of psychiatric symptoms. Saliva was collected for DNA isolation 6 months after maltreatment exposure and at 6-month follow-up. Telomere length and mtDNAcn was measured using qPCR. Age and ethnicity were included as covariates given prior literature regarding their effects on telomere length and mtDNAcn. Student's t-test, ANOVA, and regression models were used to examine the relationship between biomarkers of aging and maltreatment cross-sectionally and over time.

Results: Maltreatment status was significantly associated with telomere length at baseline and follow-up ($p = .002$, .004). The adversity composite was significantly associated with telomere length at baseline ($p = .001$) and follow-up ($p < .0001$). Similarly, maltreatment status was associated with a change in mtDNAcn that approached significance at baseline and was significantly associated at follow-up ($p = .070$, .048). The adversity composite was significantly associated with mtDNAcn at follow-up ($p = .002$). Telomere length at both baseline and follow-up were associated with internalizing behaviors at both timepoints ($p < .05$ for all). Telomere length at follow-up was significantly associated with PTSD symptoms at baseline ($p = .033$) and approached significance with PTSD symptoms at follow-up ($p = .105$). Telomere length at baseline or follow-up was not associated with MDD at either timepoint ($p > .05$). Baseline mtDNAcn was significantly associated with internalizing behaviors at baseline ($p = .014$), while follow-up mtDNAcn was significantly associated with internalizing behaviors at baseline and follow-up ($p = .003$, $< .0001$). Baseline mtDNAcn was significantly associated with baseline MDD symptoms, but not follow-up MDD symptoms ($p = .004$, .594). mtDNAcn at baseline or follow-up was not associated with PTSD symptoms at either timepoint ($p > .05$). Maltreatment did not predict the change in telomere length or mtDNAcn between baseline and follow-up when controlling for age and ethnicity ($p = .986$, .900, respectively). Telomere length did not predict MDD, PTSD symptoms, nor internalizing behaviors ($p > .05$). mtDNAcn predicted MDD symptoms over time ($p = .046$), but did not predict internalizing or externalizing behaviors or PTSD symptoms ($p > .05$).

Conclusions: This study examined associations between telomeres and maltreatment in a cross-sectional and longitudinal manner that was temporally closer to the exposure than the previous literature. Unlike studies examining telomere length at timepoints further removed, we observed telomere lengthening 6 months after the exposure. Additionally, this is the first study to show that mtDNAcn, a marker of biologic aging, is impacted by maltreatment exposure and predicts psychiatric symptoms. Both of these biologic markers of aging may be related to the underlying molecular mechanisms linking maltreatment and psychiatric disorder risk.

Keywords: Childhood Maltreatment, Telomere, Mitochondria

Disclosure: Nothing to Disclose.

T114. Electroconvulsive Treatment of Patients With Major Depressive Episodes may Normalize Dopamine D2 Receptor Binding Potential in the Executive and Limbic Striatum: A Pilot PET Study

Jonas Svensson, Mikael Tiger, Benny Liberg, Christer Halldin, Martin Schain, Lars Farde, Johan Lundberg*

Karolinska Institutet, Stockholm, Sweden

Background: Mood disorders are the leading cause of disability worldwide1. The most effective established treatment of major depressive episodes, both within unipolar and bipolar disorder, is electroconvulsive treatment (ECT). Side effects, eg memory loss, limit the application of ECT to only acute treatment of the most severe cases. A better understanding of the mechanism of action of ECT could pave the way for better treatment options.

Both lower concentration of the dopamine (DA) precursor tyrosine in plasma and CSF and reduced levels of the dopamine metabolite homovanillic acid (HVA) in the CSF has been described in depression2,3. Motor retardation is a feature of both severe depression and the DA neurodegenerative Parkinson's disease (PD). Notably, ECT has been reported to attenuate the motor symptoms of patients with PD4.

Positron Emission Tomography (PET) enables estimation of protein expression in the living human brain. [11C] raclopride is an established PET-radioligand selective for DA D2 type receptors and suitable for quantification in D2-receptor high density regions6.

The aim of the study was to use PET to examine A) if there is a difference between patients with a severe depressive episode and control subjects in D2-receptor binding in the striatum, and B) if ECT affects the D2-receptor binding in patients.

Methods: Nine patients with a major depressive episode that was to be treated with ECT were recruited. Matched control subjects were recruited. All subjects were examined twice: the patients directly before initiation and after conclusion of the ECT-series; the control subjects with a time interval matching that of the patients.

All subjects were examined with MRI to rule out pathology and for co-registering with the PET-image. ECAT HRRT (Siemens Molecular Imaging) and the radioligand [11C] raclopride was used to quantify D2-like receptors in the brain. Processing of the MR-images was done in FSL (FMRIB Software Library v5.0), where individual MR-images was transformed to MNI-space. Regions of interest

(ROIs) were extracted using an atlas by Tziortzi7 where subregions of striatum (executive, limbic and sensorimotor) are functionally defined using striato-cortical anatomical connectivity data from control subjects. After back translation, the ROIs were applied to individual parametric PET data. To reduce noise, parametric binding potential (BPND) data was created using WAPI8.

For significance testing, Welch dependent t-test was used for both cross-sectional (matched) data and for the longitudinal analysis. Effect sizes were quantified using Cohen's d.

Results: At inclusion, the patients had a moderate to severe depressive episode (MADRS 34.08.0). Two patients were excluded from the final analysis for technical reasons. All patients included in the final analysis responded to ECT (7.21.7 treatments) with a post ECT MADRS of 12.33.1 ($p<0.001$). ANalysis of [11C]raclopride binding showed (A) significantly lower BPND in patients compared to controls. Executive: (Mdiff = -0.63, $p<0.05$, 95% CI [-1.07, -0.18], Cohen's d = -1.15). Limbic: (Mdiff = -0.65, $p<0.05$, 95% CI [-1.09, -0.21], Cohen's d = -1.65). Sensorimotor: (Mdiff = -0.89, $p<0.05$, 95% CI [-1.32,-0.44], Cohen's d = -1.43); and (B) the mean level of BPND in the patients was shown to increase numerically between PET1 and 2. Executive: (Mdiff = -0.4, $p=0.15$, 95% CI [-0.98, 0.19], Cohen's d = -0.70). Limbic: (Mdiff = -0.28, $p=0.30$, 95% CI [-0.87, 0.31], Cohen's d = -0.58). Sensorimotor: (Mdiff = -0.49, $p=0.10$, 95% CI [-1.11, 0.12], Cohen's d = -0.76).

Conclusions: Previous case control studies on [11C]raclopride binding in depression have shown ambiguous results9. Here, in a limited sample of severely depressed subjects we show a large effect in all subregions of the striatum. The finding indicates that D2-binding may be a biomarker for severe depression.

There is limited data on D2 receptor binding and treatment effects. Saijo et al. (2010) reported that D2/3 receptor binding decreased in the anterior cingulate cortex of depressed patients after ECT10. In this pilot study on a small number of depressed patients, we saw a numerical increase in binding after ECT treatment. Although not statistically significant, the effect sizes (Cohen's D 0.58-0.76) indicate substantial effects. If verified in a larger sample the data support D2 receptor binding as a biomarker for ECT effect.

Limitations: [11C]raclopride is sensitive to the concentration of endogenous DA. It is thus not clear whether the findings are related to changes in DA or D2-receptor concentration. This is a pilot study in a limited sample that should be replicated in a larger group.

Keywords: Depression, Dopamine (D2, D3) Receptors, ECT

Disclosure: Nothing to Disclose.

T115. Baseline Speech and Voice Parameters and Residual Symptoms as Predictors of Relapse in Subjects With Recurrent Major Depressive Disorder

Randall Morrison*, Srini Vairavan, Andreas Tsiartas, Jennifer Smith, Dimitra Vergyri, Kimberly Cooper, Jaskaran B. Singh, Ella Daly, Jim O'Neill, Maggie Fedgchin, Seth Ness, Vaibhav Narayan, Madhukar Trivedi, Wayne Drevets

Janssen Research & Development, LLC, Titusville, New Jersey, United States

Background: Residual symptoms of Major Depressive Disorder (MDD) may convey risk for relapse, although relationships have been modest (e.g., Nierenberg et al., 2010). The OBSERVEMDD trial is an ongoing, naturalistic observational trial of recurrent MDD subjects, intended to identify predictors of relapse. Subjects receive clinician-directed oral antidepressant treatment and are followed for at least 1-year including frequent assessment of symptoms/behavior, biomarkers, function/Quality of Life (QoL). Montgomery-Asberg Depression Rating Scale (MADRS) ratings are obtained at study visits at 8-week intervals; the primary definition of relapse is MADRS $>=22$. In addition, the 16-item Quick Inventory of Depressive Symptomatology-Self-Report (QIDS) is obtained at 1-week intervals.

There is increasing interest in acoustic speech and voice parameters as markers of depression severity, and/or relapse risk (e.g., Mundt et al., 2012). The present analysis was primarily intended to evaluate the contribution of speech/voice parameters as markers of relapse risk within "residual symptom" models of relapse prediction (e.g., Nierenberg et al., 2010; Sakurai et al., 2017).

Methods: Subjects: $N=330$ subjects with recurrent MDD who recently (within 3 months of trial enrollment) responded to oral antidepressant treatment for a Major Depressive Episode, currently clinically stable with MADRS $<=14$ at study screening, and are receiving ongoing, clinician-directed treatment as usual with an oral antidepressant. The total sample is further divided into two subsamples based on MADRS, and also QIDS relapse criteria. Inclusion criteria for the analysis includes relapse event (defined by MADRS or QIDS) to have occurred at 9 weeks or more in the study and data available for the self-report measures and speech/voice parameters at baseline. The total number of subjects with data availability of self-report measures and speech/voice parameters at baseline are 201 (MADR Relapse = 49, Non-Relapse = 152) and 213 (QIDS Relapse = 60, Non-Relapse = 153) respectively.

Measures: The self-report measures considered in this analysis include Depression Implicit Association Test (DIAT), EuroQol Group, 5 dimension, 5 level (EQ5D), Generalized Anxiety Disorder Questionnaire (GAD-7), MOS Sleep-R Questionnaire, Patient Adherence to Antidepressant Medication Questionnaire (PAQ), Pain Frequency, Intensity and Burden Scale (PFIBS), Perceived Stress Scale (PSS), QIDS, Recent Life Changes Stress Test (RLCST), Snaith Hamilton Pleasure Scale (SHAPS), World Health Organization Disability Assessment Scale, Health Resource Use Questionnaire (HRUQ) and the speech/voice parameters include energy, spectral features (Mel frequency cepstral coefficient, Mel-scaled filter banks), temporal evolution of speech rate and pauses, and features quantifying the tone and intonation of voice. Speech/voice parameters were scored using automated software from brief speech recordings at baseline.

Analyses: The analysis discussed is based on an interim data from the ongoing trial. Modeled on reports by Nierenberg et al. (2010) and Sakurai et al. (2017), predictive models of QIDS-defined relapse (QIDS $>=11$) were explored, using baseline "residual symptom" features. In addition, similar models were developed regarding MADRS-defined relapse (MADRS $>=22$).

Results: The predictive models are created using an ensemble prediction approach using nested cross validation technique. The modelling technique used in this analysis is Gradient Boosted Regression Trees (GBRT) and only the top 25 predictors are used in the models. The predictive models based on QIDS data had a balanced accuracy (BAC) and positive predictive value (PPV) of 0.61 and 0.79 respectively. In addition, the Fisher exact test is also significant (p-value = 0.005) with odd-ratio of 2.45. The top 25 predictors include GAD total score, WHODAS total score, EQ5D visual analytic score, QIDS total score, spectral features of voice, speech rate and pauses. However, the predictive model results based on MADRS data were not significant with BAC, PPV and p-value from Fisher exact test as 0.52, 0.77, $>$ 0.05 respectively.

Conclusions: The findings highlight the importance of speech/voice parameters for further study as markers of relapse risk, and need for models for risk and timing of relapse that improve upon information from residual symptoms. Although the model based on MADRS defined relapse was not predictive, these results may have been impacted by the lower number of MADRS-defined relapses. Regardless, it is interesting that only speech and voice features entered the model to predict MADRS-defined research; no patient-reported measures were entered. Patient-reported residual symptoms may be less predictive of clinician-defined relapse than patient-reported "relapse." As clinician-rated relapse is gold standard, including for pharmacological trials to reduce/prevent relapse (Sim et al., 2016), development of better predictive models of such relapses remains a critical need.

Keywords: Major Depressive Disorder, Relapse, Residual Symptoms

Disclosure: Part 5: Janssen R&D, Employee.

T116. An Approach to Profiling Mood and Anxiety Disorders Based on Functional Brain Circuits, Behavior and Symptoms

Zoi Samara*, Tali Ball, Andrea N. Goldstein-Piekarski, Katherine A Grizansio, Carlos Correa, Leanne M. Williams

Stanford University School of Medicine, Palo Alto, California, United States

Background: In developing precision medicine for psychiatry, one approach is to conceptualize disorders as disruptions in underlying brain circuits that may cut across more than one diagnostic category. We are developing and testing a theoretically-motivated taxonomy for conceptualizing mood and anxiety disorders in terms of multiple different ways that functional brain circuits become disrupted (Williams et al., 2016). Clinical features of stress and fear, anhedonia and poor cognitive function implicate dysregulation of stimulus-evoked circuits involved in threat regulation, reward and cognitive control. Although our current knowledge has necessarily focused on diagnostically based case:control studies, these features and circuit dysfunctions are likely expressed in specific subtypes of people within multiple mood and anxiety diagnoses. Here, our objective was to explore the brain circuit-symptom-behavior relations that characterize these subtypes in a representative sample.

Methods: We recruited an un-medicated sample of 114 participants, 80 who met traditional diagnostic criteria for major depression, and one-third also met criteria for an anxiety disorder. We also recruited a second community sample of 160 un-medicated adults spanning a wider range of mood and anxiety disorders. Participants were tested with a standardized battery of functional brain imaging sequences and stimulus paradigms. A facial emotion viewing task (conscious and non-conscious conditions) was used to elicit threat and reward circuits and a Go/NoGo paradigm was used to elicit the cognitive control circuit (Korgaonkar et al., 2013). We also assessed intrinsic (i.e., task-independent) functional connectivity. Finally, we assessed participants on a comprehensive set of symptom scales (including the Mood and Anxiety Symptom Questionnaire, the Beck Anxiety Inventory, the Quick Inventory of Depressive Symptomology, and the Penn State Worry Questionnaire), and cognitive and emotional behavioral performance battery assessing executive functioning, attention, working memory, and reaction time to emotional face stimuli (Silverstein et al., 2007).

To operationalize our taxonomy, we identified brain-symptom-circuit profiles according to the following steps. First, we systematically defined the regions comprising each circuit of interest, and the connections between them, based on the comprehensive synthesis of knowledge underlying our theoretical taxonomy (Williams, 2016). Second, activation for each of region and their connectivity were quantified for the contrast of interest for each paradigm. Third, we computed specific subtypes according to the definitions in our model-driven taxonomy. Following this model-driven approach with general linear models, we tested if subgroups defined by extreme scores on each type of circuit disruption were also distinguished by symptom and behavioral features implicated in the same construct; for example, symptoms of fear and anxious arousal related to disruptions to threat circuitry.

Results: Several distinctive profiles suggest that circuit-defined subtypes might be expressed in specific symptom and behavioral features. Using the hypothesis-driven alpha threshold of .05, we observed that amygdala-ACC hypo-connectivity relevant to threat dysregulation was related to symptoms of anxious arousal ($F(16, 135) = 2.1$, $p = .01$; $t(136) = -2$, $p = .05$) and to longer dwell times when identifying threat (Fig 1). Striatal-vMPFC hypo-connectivity relevant to reward circuit dysfunction and anhedonia was related to reaction time insensitivity to socially rewarding stimuli (Fig 1), and DLPFC-ACC hyper-connectivity during a cognitive control task was related to poorer executive function (Fig. 1). The intrinsic salience circuit was associated with greater symptoms of anxious apprehension ($p = .02$).

Conclusions: Using a model-driven approach we can begin to characterize the clinically-relevant way in which brain circuit disruptions are expressed in specific symptoms and behaviors involved in core functions such as regulating emotion and cognition. This approach lays a foundation for further data-driven approaches, quantification of circuit interactions and probing of circuit types using targeted interventions.

Keywords: MRI, Anxiety, Depression, Precision Medicine

Disclosure: Nothing to Disclose.

T117. Stress-Induced Changes in the Translatome of Serotonergic Neurons are Sex Specific

John Neumaier*, Atom Lesiak, Kevin Coffey, Joshua Cohen, Charles Chavkin

University of Washington, Seattle, Washington, United States

Background: Serotonin is a primary mediator of stress, anxiety, and depression and drugs that regulate serotonin reuptake and metabolism have been critical for treating anxiety and depressive disorders. However, currently available medications target rather few mechanisms within these neurons and can induce numerous side effects, highlighting the importance of identifying new molecular targets within the serotonergic neurons. A major challenge in identifying novel molecular targets in serotonergic neurons is the diversity of other cell types that are also located in the midbrain.
Methods: In this study, we conducted a cell-specific gene expression analysis of serotonergic neurons in the dorsal raphe utilizing the translating ribosome affinity purification (TRAP) method. To express HA-tagged RPL22 protein (RiboTag) specifically within serotonergic neurons, we crossed Cre-dependent floxed RiboTag mice with serotonin-specific Cre driver mice (Pet1-CRE). The resulting cross allowed for isolation and next-gen sequencing of ribosome-associated mRNA transcripts specifically from serotonergic neurons. RNA was collected from mice 4 hours after the last swim of a two-day repeated swim stress and were compared to that of unstressed mice. RiboTag-associated RNA was used to make RNA libraries that were than subjected to RNAseq and downstream bioinformatics analysis. Normalized gene counts were analyzed by Gene Set Enrichment Analysis and differentially expressed genes were identified with DESeq2.
Results: RNA was collected from mice 4 hours after the last swim of a two-day repeated swim stress and were compared to that of unstressed mice. RiboTag-associated RNA was used to make RNA libraries that were than subjected to RNAseq and downstream bioinformatics analysis. Normalized gene counts were analyzed by Gene Set Enrichment Analysis and differentially expressed genes were identified with DESeq2. The differentially expressed genes were analyzed for upstream and downstream motifs and for coregulation using unbiased cluster analyses. There were substantially more differentially expressed RNAs identified in stressed female mice relative to unstressed than in males. Several patterns emerged in the females, including evidence for co-regulation of neuroinflammatory pathways as well coordinated regulation of membrane channels within serotonin neurons of female mice. Validation of differentially expressed genes are currently being validated in new sets of mice.
Conclusions: These data point to a number of novel targets within serotonin neurons that may be important new targets for the development of antidepressant treatments.
Keywords: RNAseq, RiboTag, Sex Differences, Bioinformatics
Disclosure: Nothing to Disclose.

T118. Deficits in Frontoparietal Activation and Anterior Insula Functional Connectivity During Regulation of Cognitive-Affective Interference in Bipolar Disorder

Kristen Ellard*, Julia Felicione, Aishwarya Gosai, Amy Peters, Conor Shea, Andrew Nierenberg, Darin Dougherty, Thilo Deckersbach

Harvard Medical School, Boston, Massachusetts, United States

Background: The simultaneous regulation and integration of affective and cognitive information is critical to adaptive functioning. Bipolar disorders (BD) are characterized by affective lability and emotion and cognitive dysregulation, key factors of pathology-related dysfunction. Mapping deficits in cognitive-affective regulation and integration at the level of neurocircuitry allows for the potential identification of intervention targets to improve cognitive and emotion dysregulation in BD. In the current study, we collected functional MRI data in a sample of BD patients and healthy controls during performance on a modified version of the Multi Source Interference Task (MSIT). Specifically, MSIT trial presentations were superimposed on positive, negative and neutral images from the International Affective Picture Scale (IAPS), probing patients' neurocircuitry during the regulation of simultaneous cognitive and affective interference.
Methods: Functional MRI data were collected from 36 BD I patients and 36 healthy controls during performance on the MSIT-IAPS. MSIT incorporates aspects of well-established measures of two different types of cognitive interference (spatial and flanker) to measure cognitive control (Bush et al., 2003; 2006). For the modified version, each trial of the MSIT is overlaid on a positive, negative or neutral IAPS picture in order to introduce affective interference in addition to cognitive interference. Following standard preprocessing procedures, task-based fMRI data were analyzed using SPM8. Contrasts were modeled in a flexible factorial GLM comparing BD patients versus controls during interference > non-interference, negative/positive > neutral, and negative/positive interference > negative/positive non-interference trials. Generalized PPI (gPPI) analyses were conducted using right anterior insula, a key node implicated in cognitive-affective integration (e.g. Sridharan et al., 2008) as the seed region.
Results: Bipolar patients evidenced significantly weaker activation relative to healthy controls during Interference > Non-Interference contrasts in anterior insula ($t(365) = 4.00$, $p = .03$ FWE), dorsolateral prefrontal cortex ($t(365) = 5.16$, $p = .001$ FWE), and inferior parietal lobule ($t(365) = 4.47$, $p = .006$ FWE), regardless of IAPS valence. No other contrasts survived FWE corrections. Significantly stronger functional connectivity was found in BD patients relative to controls between the right anterior insula seed region and right amygdala ($t(365) = 3.87$, $p = .006$ FWE), hippocampus ($t(365) = 6.21$, $p < .000$ FWE), ventromedial PFC ($t(365) = 4.70$, $p = .002$ FWE), dorsal anterior cingulate ($t(365) = 4.44$, $p = .006$ FWE), and posterior cingulate ($t(365) = 5.20$, $p < .000$ FWE) in Interference > Non-Interference trials, but only during trials superimposed on a

negatively valenced image. Significantly weaker functional connectivity was found in BD patients relative to controls during negatively valenced Interference > Non-Interference trials between right anterior insula and left inferior parietal lobule ($t(365) = 2.90$, $p = .002$), right ventrolateral PFC ($t(365) = 3.49$, $p < .000$), and right DLPFC ($t(365) = 2.53$, $p = .006$), but these differences did not survive FWE correction.

Conclusions: During regulation and integration of both affective and cognitive information, BD patients evidenced significantly decreased recruitment of key regions implicated in cognitive control (DLPFC, IPL) as well as reduced recruitment of the anterior insula, a key node implicated in integrating affective and cognitive information. Additionally, when simultaneously regulating cognitive interference and negatively valenced stimuli, BD patients evidenced weaker functional connectivity between anterior insula and cognitive control regions, and increased functional connectivity between anterior insula and limbic and affective processing regions. Results of this study suggest one pathway to dysregulation in BD is through over-reliance upon a distributed network facilitating affective processing during cognitively demanding tasks, and inefficient integration of affective and cognitive information. Results also suggest the importance of developing interventions that target not only cognitive control-related regions but also key nodes implicated in the integration of cognitive and affective information (anterior insula, IPL).

Keywords: Bipolar Disorder, Cognitive Control, Emotion Regulation, Emotion Cognition Integration, fMRI

Disclosure: Nothing to Disclose.

T119. Genomic Estimation of Metabolite Scores (GEMS) Reveals Potential Association of Choline Levels With Bipolar Disorder

Joanna Biernacka*, Beth Larrabee, Mark Frye

Mayo Clinic, Rochester, Minnesota, United States

Background: The metabolome is the collection of small molecules (metabolites) involved in biochemical reactions in a biological sample. The metabolome is influenced by both genetic and environmental factors. Particular perturbations in the metabolome may be associated with risk of disease; thus, investigation of metabolite differences between cases of a disease and healthy controls may be useful in revealing disease etiology. Here we propose a new approach, Genomic Estimation of Metabolite Scores (GEMS), which involves using genome-wide single nucleotide polymorphism (SNP) data to estimate the genetically-determined deviations from population averages in an individual's metabolite levels, followed by testing for association between these score and phenotypic traits. This approach attempts to investigate the endogenous (i.e. genetic) effects on metabolite levels to identify metabolites that impact disease risk. To demonstrate this new approach, we have applied it to search for metabolites that may contribute to risk of bipolar disorder.

Methods: We used summary statistics from a large published metabolomics genome-wide association study of almost 8,000 individuals (Shin et al. Nat Genet 2014), as well as genome-wide SNP data from a study of bipolar disorder, to estimate genetically-determined blood metabolite scores for 249 metabolites in bipolar disorder cases and controls. The target dataset consisted of 1001 bipolar cases and 1034 controls from the Genetic Association Information Network (GAIN) dataset. Metabolite scores were estimated using the polygenic risk score approach implemented in the software PRSice. In particular, we used estimated effect sizes for SNP associations with blood metabolites based on the results of Shin et al., to calculate polygenic scores for all 249 metabolites in the bipolar cases and controls from the GAIN samples. We then used logistic regression to test for association of the metabolite polygenic scores with bipolar disorder. P-values < 2*10-4 were considered statistically significant.

Results: Choline was the only estimated metabolite that demonstrated significant association with bipolar disorder after correction for multiple testing ($p = 3.2E-05$), with bipolar cases having on average higher genetically-determined choline levels than controls. The metabolite with the second strongest evidence of association was myo-inositol ($p = 0.002$), with cases having marginally higher myo-inositol on average, although this difference is not statistically significant after Bonferroni correction. Other nominally significant findings for metabolites that have been implicated in bipolar disorder include glutamate ($p = 0.036$).

Conclusions: Our findings are intriguing given the previously-reported altered levels of choline and choline-containing compounds, as well as myo-inositol, in bipolar disorder. Much of the prior research of these compounds in bipolar disorder was based on brain metabolite levels evaluated using magnetic resonance spectroscopy. To our knowledge, our study provides the first evidence that the altered metabolite levels may at least partly represent systemic differences driven by genomic variants. While tissue-specific metabolomics offers the possibility to study associations that more directly represent the underlying biological mechanisms of neuropsychiatric disease etiology, blood metabolites can also serve as important biomarkers for predicting and understanding psychiatric diseases. Our results motivate further investigation of the role of choline and myo-inositol in bipolar disorder, as well as further application of the proposed GEMS approach to the study of complex human traits.

Keywords: Metabolomics, Genomics, Bipolar Disorder

Disclosure: Nothing to Disclose.

T120. The Antidepressant Effect of TMS: Resting State Functional Connectivity Predictors and Mechanisms of Treatment Response

Tracy Barbour*, Erik Lee, Ellard Kristen, Joan Camprodon

Massachusetts General Hospital, Boston, Massachusetts, United States

Background: Repetitive transcranial magnetic stimulation (rTMS) is a painless, focal, and noninvasive tool for inducing long-term neuromodulatory effects, that is approved by the Food and Drug Administration (FDA) for the treatment of major depressive disorder (MDD). This treatment utilizes targeted stimulation over the dorsolateral prefrontal cortex

(DLPFC), taking place over the course of weeks, and resulting in an upregulation of functional activity at the site of stimulation. rTMS has emerged as one of the most effective treatments for MDD, but our understanding of how rTMS alters connectivity between functional networks, and the mechanisms of associated antidepressant response remain unclear. In this work, the we explore measures of functional connectivity before and after rTMS treatment, and evaluate how both baseline levels of connectivity, and changes in connectivity that occur throughout the course of TMS, are related to antidepressant response.
Methods: A total of 17 patients were stimulated with rTMS 10 Hz frequency stimulation over the left prefrontal cortex for the treatment of MDD. Resting state fMRI was employed before and after the full course of rTMS to measure functional connectivity. The location of the stimulation site used for treatment was functionally localized to the area of the left prefrontal cortex with the greatest functional anti-correlations to the subgenual cingulate cortex (SCC). Measures of functional connectivity were calculated before and after treatment and related to clinical response, as measured through the Hamilton Depression Rating Scale (HAMD-17). Relationships were tested for significance between the SCC, the target of stimulation, and 7 predefined resting-state networks representative of brain function. All inferences were corrected for multiple comparisons using permutation tests (for network comparisons), and considered significant for $p < .05$.
Results: Of the 17 subjects, 8 responded (50 % or more improvement on HAMD-17) after completing rTMS, and 7 remitted. There was significant overlap between the sites of stimulation of the responders and non-responders. Changes in connectivity (post – pre), between the target of stimulation and the SCC were significant predictors of antidepressant response ($r = 0.56$, $p = 0.02$, two-tailed). Additionally, baseline functional connectivity between the SCC and default mode network (DMN) predicted antidepressant response ($r = 0.69$, $p = 0.02$, two-tailed) to rTMS, and changes in connectivity between the SCC and DMN observed after rTMS also predict the magnitude of antidepressant response ($r = -0.62$, $p = 0.024$ one-tailed).
Conclusions: Our results show that baseline levels of functional connectivity can be predictive of antidepressant response, and also, changes in functional connectivity can explain the mechanisms of action of TMS in MDD. Specifically, the antidepressant response to rTMS is associated with increased anti-correlation between the target of stimulation in the left frontal cortex and the SCC, as well as increases in connectivity between the SCC and DMN. The default mode network, a system involved in self-inspection and rumination, was shown to be strongly related to antidepressant response. Interestingly though, our results show that increases in connectivity between the SCC and the DMN are related to antidepressant response, which is contrary to what has been previously reported. Our predictors analyses further show that individuals with weaker baseline connectivity between the SCC and DMN also see the greatest antidepressant response, suggesting that it is important for patients with depression to achieve strong connectivity between the SCC and the DMN, and that individuals with already high baseline connectivity may have less to gain from TMS.

Keywords: rTMS, Depression, Functional MRI (fMRI)
Disclosure: Nothing to Disclose.

T121. Clustering Identifies Symptom-Brain-Behavior Subtypes That cut Across Mood, Anxiety, and Trauma Disorders

Katherine A. Grisanzio, Andrea N. Goldstein-Piekarski, Michelle Wang, Abdullah Rashed Ahmed, Zoe Samara, Leanne Williams*

Stanford University School of Medicine, Stanford, California, United States

Background: Although the symptoms that define mood, anxiety and post-traumatic stress disorders are highly overlapping across disorders and highly heterogeneous within disorders, we do not yet know if there are coherent subtypes that span multiple diagnoses and are also expressed functionally in underlying cognition and brain function, as well as clinically in daily function measures. Identification of cohesive subtypes would help disentangle the symptom overlap in our current diagnoses and serve as a tool for predicting and tailoring treatment choices. Here we propose and demonstrate one data-driven approach for identifying subtypes within a trans-diagnostic sample.
Methods: A data-driven analysis was undertaken for data acquired from 2006 to 2010 from the BRAINnet Foundation Database. Data was collected from five medical or clinical research sites. The medical research sites were located in universities with teaching hospital outpatient clinics focused on the disorders of interest, and based in the same geographical communities as the controls. 420 individuals (mean age 39.8 +/- 14.1 SD, 61.0% female), comprising patients with a primary diagnosis of Major Depressive Disorder ($n = 100$), Panic Disorder ($n = 53$) or Post-traumatic Stress Disorder ($n = 47$) and case-wise matched healthy controls ($n = 220$) were assessed with a standardized protocol. We followed a step wise data-driven approach to achieve the primary study outcome of identifying trans-diagnostic subtypes. First, machine-learning with a hierarchical clustering algorithm was implemented to classify participants based on self-reported negative mood, anxiety and stress symptoms. Second, we tested the robustness and generalizability of the resulting subtypes in an independent sample of 381 adult participants, age 18 to 86 (mean: 36.65 +/- 14.57 SD, 54.3% female), also recruited from community sources and tested at an academic center. This validation sample encompassed an equivalent trans-diagnostic scope, including Major Depressive Disorder, Post-traumatic Stress Disorder, Panic Disorder, Generalized Anxiety Disorder, Social Anxiety Disorder, Obsessive Compulsive Disorder, Simple Phobia, Bipolar II disorder, as well as healthy controls. Third, we assessed whether these symptom subtypes were expressed at behavioral and physiological levels of functioning by examining if groups differentiated on neurocognitive performance and neurophysiological brain activation. Fourth, we assessed the clinical meaningfulness of the subtypes in relation to measures of daily functional capacity. These findings were interpreted relative to a complementary diagnostic frame of reference, by mapping the multi-scale subtypes onto DSM-defined diagnostic classifications.

Results: We identified six distinct subtypes characterized by "Normative mood", "Tension", "Anxious Arousal", "General Anxiety", "Anhedonia", and "Melancholia" and these subtypes were replicated in an independent sample. Subtypes were functionally expressed through differences in neurocognitive performance on domains of cognitive control and memory and in neurophysiological brain activation assessed by EEG-recorded alpha and beta power. The subtypes also differed in regard to social functional capacity and emotional resilience. These multi-scale subtypes cut across DSM-defined diagnoses of MDD, panic disorder, and PTSD.
Conclusions: These findings offer a data-driven framework for identifying robust subtypes that are meaningful in terms of specific, coherent relations between symptoms, behavior, brain function and observable real-world function.
Keywords: Depression, Anxiety, Posttraumatic Stress Disorder, Machine Learning Clustering, Precision Psychiatry
Disclosure: Part 1: Psyberguide, Advisory Board.

T122. Assessing Anhedonia With Quantitative Tasks, Digital and Patient Reported Measures in a Multi-Center, Double-Blind Trial With BTRX-246040 for the Treatment of Major Depressive Disorder

Annette Madrid, Daniel Smith, Sarah Alvarez-Horine, Kerensa Saljooqi, Paul Dagum, Atul Mahableshwarkar*

BlackThorn Therapeutics Inc., San Francisco, California, United States

Background: Anhedonia, a core feature of major depressive disorder (MDD), is present in approximately 37% of patients with MDD and may represent a physiologically distinct subgroup of patients with motivational and reward processing deficits1. An inadequate response to treatment is also seen in many patients with anhedonia. Understanding and characterizing anhedonia may help in developing treatments, as current approaches do not sufficiently target the depression/anhedonia related motivational and reward-processing deficits2. BTRX-246040 is a potent and selective antagonist of the human nociceptin receptor (NOPR), in development for the treatment of patients with neurobehavioral disorders characterized by these deficits.
Methods: NEP-MDD-201 is a randomized, multi-center, double-blind placebo-controlled study evaluating the efficacy of BTRX-246040 as compared to placebo in adult patients with MDD over an 8-week period. Patients eligible after screening will be randomized to treatment with either placebo or 80mg of BTRX-246060. Efficacy will be assessed using the Montgomery-Asberg Depression Rating Scale (MADRS), the Clinical Global Impression of Severity (CGI-S) and Improvement (CGI-I), and the Facial Expression Recognition Task (FERT). Anhedonia will be characterized with both patient-reported outcome measures (Dimensional Anhedonia Rating Scale [DARS] and Snaith Hamilton Pleasure Scale [SHAPS]) and computerized tasks (Probabilistic Reward Task [PRT] and Effort Expenditure for Reward Task [EEfRT]). Additionally, vocal and passive-behavioral digital measurements will be collected via applications downloaded on patients' smartphones. Safety will be assessed using adverse event (AE) monitoring, the Columbia Suicidality Severity Rating Scale (C-SSRS), physical examinations, vital signs, laboratory assessments, and electro-cardiograms (ECGs).
Results: The trial has recently commenced and blinded baseline data will be available for presentation. Baseline demographics, disease severity and results from the clinician-rated, patient-rated, and computerized active and passive assessments will be summarized. Descriptive statistics will be calculated. Tabulated summaries of baseline data from approximately 20 patients with and without anhedonia will be presented.
Conclusions: Anhedonia can be characterized utilizing clinician assessment, patient report, computerized and smartphone based evaluations. Descriptive statistics for blinded preliminary data will be presented to compare patients with and without anhedonia.
Keywords: Anhedonia, Reward Processing, Motivational, Digital Assessment
Disclosure: Part 1: BlackThorn Therapeutics, Employee, GlaxoSmithKline, Stock / Equity, Johnson & Johnson, Stock / Equity, Pfizer, Stock / Equity, **Part 2:** BlackThorn Therapeutics, Employee, GlaxoSmithKline, Stock / Equity, Johnson & Johnson, Stock / Equity, Pfizer, Stock / Equity, **Part 3:** BlackThorn Therapeutics, Employee, **Part 5:** BlackThorn Therapeutics, Employee.

T123. MRSI in Bipolar: Neurometabolite Patterns and Alterations With Lamotrigine Treatment

Shariful Syed*, Jeremy Coplan, Chadi Abdallah

Miller School of Medicine, University of Miami, Miami, Florida, United States

Background: Bipolar disorder is associated with alterations in brain metabolic states. Recent studies of bipolar disorder using magnetic resonance spectroscopy imaging (MRSI) have demonstrated alterations of certain neurometabolite patterns occurring in the anterior cingulate cortex (ACC) and other brain regions such as the dorsolateral prefrontal cortex (dLPFC). We wished to further explore how neurometabolites such as GABA and Choline are altered with current standard of care treatment in those with Bipolar I (BPI) disorder.
Methods: 10 patients with a diagnosis of bipolar I, currently depressed were enrolled, and the right dorsolateral prefrontal cortex (DLPFC) and anterior cingulate cortex (ACC) was investigated before and after 8 weeks of Lamotrigine monotherapy by 1 H-MRS. The results were compared with 10 age- and gender-matched controls.
Results: In patient diagnosed with Bipolar I disorder compared with healthy controls, those with BPI diagnosis demonstrated reduced Choline and Creatinine levels in Anterior Cingulate Cortex pretreatment with Lamotrigine monotherapy (Choline_t = -2.38, p = .027; GABA_t = 4.3 $p = .0004$). After treatment, there was significant increase in GABA in right dorsolateral prefrontal cortex (t = 3.12, $p = .03$). Neurometabolite ratios with creatine as a denominator showed Glutamate/Glutamine and NAA in ACC/dLPFC when comparing BPI patients to healthy controls.

Conclusions: Our results indicate that Bipolar I disorder is accompanied by state dependent neurometabolic alterations in GABA, Glutamate, and NAA, some of which may be modulated by treatment with Lamotrigine.
Keywords: Magnetic Resonance Spectroscopy, Bipolar Disorder, Lamotrigine
Disclosure: Nothing to Disclose.

T124. Hormonal Regulation of G Protein-Coupled Receptor Kinase 2 (GRK2) Biases Kappa Opioid Receptor Signaling to Produce Sexually Dimorphic Analgesic Responses

Antony Abraham*, Selena Schattauer, Joshua Cohen, Juliana Chase, Salina Johnson, Benjamin Land, Charles Chavkin

University of Washington, Seattle, Washington, United States

Background: Chronic stress activates the kappa opioid receptor (KOR) to produce aversive and analgesic responses via distinct intracellular signaling pathways. The aversive components of KOR activation requires the G protein-receptor kinase 3 (GRK3)/β-arrestin/p38 MAPK signaling pathway, whereas analgesic effects of KOR activation requires the release of the G beta-gamma subunit from the KOR-associated inhibitory Gαi protein. G-biased KOR agonists have been in therapeutic development for their analgesic and antipruritic effects, but studies have indicated that males and females show dissimilar behavioral and molecular responses to KOR activation. Understanding how KOR activation leads to sexually dimorphic responses is necessary to advance the development and implementation of KOR agonists in humans.
Methods: Female and male C57BL/6 mice (8-12 weeks) were used in these studies. Mice received either repeated forced swim stress (rFSS; 15 min swim followed by 4x6 min swims separated by 6 min) or the KOR agonist U50,488 (0-10 mg/kg) prior to behavioral and molecular experiments. Western blot analysis was used to assess phosphorylation of p38 MAPK and ERK. Analgesia was assessed using either the warm-water tail withdrawal test or the hot plate test. Place preference or aversion was conditioned using contextual visual and tactile cues. Female mice were ovariectomized at least two weeks prior to tissue collection or behavioral experiments. CMPD 101, a GRK2/3 inhibitor, was administered 30 min prior to behavioral experiments or tissue collection.
Results: Western blots of striatal homogenates from female and male mice showed similar patterns of p38 MAPK phosphorylation following KOR agonism. Phosphorylation of the p38 MAPK is required for KOR-mediated aversion and potentiation of cocaine conditioned place preference, and these behaviors were observed in both female and male mice. In contrast, KOR agonism also consistently increased ERK phosphorylation in the striatal homogenates from males, but not females. Additionally, KOR agonism did not produce analgesia in either a warm-water tail withdrawal test or a hot plate test in females. These effects could not be attributed to preferential activation of arrestin-dependent signaling in female mice, as systemic deletion of GRK3 did not increase KOR-mediated analgesic responses. An alteration in basal dynorphin tone to produce receptor desensitization is also unlikely to explain sexually dimorphic analgesic responses, as female mice with a global knockout of dynorphin did not show a KOR-mediated analgesic response. Instead, we observed that KOR-mediated analgesia was present during low estrogen phases of the estrous cycle and suppressed by high estrogen phases of the estrous cycle. KOR activation in ovariectomized females increased ERK phosphorylation and increased KOR-mediated analgesic responses, demonstrating the necessity of gonadal hormones for the altered bias in intracellular signaling. We then hypothesized that GRK2, a kinase regulated by estradiol and that has been shown to sequester G beta-gamma signaling, could be responsible for the lack of KOR-mediated analgesia observed in females. Although there are no drugs selective specifically for GRK2, administration of a GRK2/3 inhibitor (15 mg/kg CMPD 101, Tocris) to female mice prior to KOR activation increased KOR-mediated analgesia and ERK phosphorylation. Current experiments aim to determine the substrates for GRK2 activity and the generalizability of these effects to other Gαi-coupled receptor systems.
Conclusions: We found that in female mice, KOR-mediated ERK phosphorylation and analgesia were absent, but p38 MAPK activation and aversion was retained. KOR-mediated ERK phosphorylation and analgesia were observed in female mice during low estrogen phases of the estrous cycle or following ovariectomy, indicating that these effects were dependent on circulating hormone levels. These behavioral and molecular effects of KOR activation could also be observed in females following inhibition of GRK2, a kinase regulated by estradiol. Together, these findings implicate a GRK2-dependent mechanism underlying sexually dimorphic responses to KOR activation. Our results suggest that G-biased KOR agonists in clinical development may have dissimilar effects in male and female populations, and provides a molecular target to mitigate these differential effects of KOR activation. Future studies will examine whether estradiol modulation of GRK2 underlies a variety of other sexually dimorphic responses mediated by G beta-gamma signaling.
Supported by NIH grants P50-MH106428, R01-DA030074, T32-DA07278, and NARSAD, Young Investigator Award (BBL).
Keywords: Kappa Opioid Receptor, Sexual Dimorphism, Analgesia
Disclosure: Nothing to Disclose.

T125. Electrophysiological Evidence of Enhanced Early and Later Emotional Information Processing in Youth With Pediatric Bipolar Disorder

Alessio Simonetti, Marijn Lijffijt, Ramandeep Kahlon, Kellen Gandy, Ruchir Arvind, Pooja Amin, David Arciniegas, Jair Soares, Kirti Saxena, Alan Swann*

Baylor College of Medicine, Houston, Texas, United States

Background: Impaired emotion regulation could affect cognition and behavior, and is an aspect of bipolar disorder. To our knowledge, information about changes in the

temporal dynamics of emotional information processing in pediatric affective disorders is limited to pediatric major depression, and in bipolar disorder is limited to adults. We studied the temporal dynamics of emotional information processing in children/adolescents with Pediatric Bipolar Disorder (PBD) and healthy controls (HC) using the occipital P100 and the occipital Late Positive Potential (LPP) evoked by faces with emotional expressions.
Methods: We studied 15 children/adolescents with PBD and 14 age and sex-matched HC's. Participants passively viewed youth and adult faces with a neutral, happy or fearful expression while EEG was measured. We examined occipital P100 (70-150 ms) and LPP (400-1000 ms) components as correlates of sensory and attentional emotional processing. General Linear Models were used to examine group differences.
Results: P100 ($F[1,28] = 8.64$, $P = .007$) and LPP ($F[1,28] = 9.31$, $P = .005$) amplitudes were more positive in PBD than HC. The enhanced LPP amplitude was accounted for largely by the enhanced P100 amplitude ($F[1,27] = 17.59$, $P < .001$), suggesting a common mechanism underlying amplification of both components.
Conclusions: Outcomes suggest increased early sensory (P100) and late attentional (LPP) reactivity to emotional information in PBD than HC, perhaps associated with early stimulus-induced hyperactivation of the amygdala relative to prefrontal cortical involved in emotion regulation. Our outcomes may indicate that youth with bipolar disorder have impaired disengagement from emotional information. Other studies suggest that this may be absent in adults with bipolar disorder.
Keywords: Childhood Onset Bipolar Disorder, Neurophysiology, Impulse Control, Amygdala
Disclosure: Nothing to Disclose.

T126. Depression is Associated With Pregenual ACC Activation During Unpredictable Threat

Namik Kirlic*, Robin Aupperle, Ruben Alvarez

Laureate Institute for Brain Research, Tulsa, Oklahoma, United States

Background: The emotion context-insensitivity (ECI) hypothesis (Rottenberg et al., 2005) suggests that depression is associated with diminished emotional reactivity to both positive and negative stimuli (i.e., emotional blunting). However, there is also evidence of increased reactivity to anticipation of physically aversive stimuli (i.e., electric shock) in depressed individuals (Grillon et al., 2013). We examined how major depressive disorder (MDD) related to neural and psychophysiological responses during anticipation of predictable and unpredictable threat of electric shock.
Methods: Twenty healthy control (HC) and 17 participants with MDD were scanned while virtually navigating two contexts. In the predictable threat context (PCXT), participants could receive an electrical stimulation on the ankle only during a visual cue (PCUE). In the unpredictable threat context (UCXT), the cue (UCUE) had no signal value as participants could receive stimulation at any time. The Hamilton Depression Rating Scale (HAM-D) was used to assess depressive symptoms. Mixed effects multilevel regression analysis was used to identify group differences in neural activation to unpredictable threat (UCXT-PCXT) and the relationship between percent signal change (PSC) and MDD illness severity. Analyses were conducted voxel-wise across the whole brain and for PSC within regions of interest (amygdala, bed nucleus of stria terminalis [BNST]).
Results: Relative to HC, the MDD group had significantly greater activation in the right pregenual anterior cingulate cortex (pgACC) during unpredictable threat ($pcorr < .005$). Region of interest analysis within amygdala and BNST did not reveal significant group differences. During unpredictable threat, pgACC activity was not associated with a reduction in activity in the BNST and amygdala ($p > .05$). Severity of illness in the MDD group was inversely associated with pgACC activation ($r(15) = -.50$, $p < .05$).
Conclusions: The present study suggests that the pgACC plays an important role in how individuals with MDD process threat. While MDD in general may relate to greater pgACC activation, this did not seem to translate into effective inhibition of limbic areas (i.e., BNST). Further, results suggest that pgACC activation in MDD may be playing a compensatory role for those with less severe symptoms and/or become blunted for those with more severe symptoms.
Keywords: Depression, BOLD imaging, Threat of Shock
Disclosure: Nothing to Disclose.

T127. Chronic Stress-Induced Complement Component 3 Signaling Promotes Depressive-Like Behavior

Amanda Crider, Tami Feng, Talisha Davis, Babak Baban, Anilkumar Pillai*

Medical College of Georgia, Augusta, Georgia, United States

Background: Increasing evidence suggests a potential role of the immune system in the pathophysiology of MDD. The complement system represents one of the major effector mechanisms of the innate immune system, and plays a critical role in inflammation. However, the role of complement components in chronic stress-induced depressive behavior is not understood.
Methods: Adult male mice were exposed to chronic unpredictable stress (CUS) to examine the role of complement system in depressive-like behaviors. Complement component 3 receptor (C3aR) knockout mice was used. Viral mediated overexpression of C3 was performed in mouse PFC.
Results: We found that increased C3 expression in PFC as a result of chronic stress causes depressive-like behavior. Conversely, mice lacking C3aR were resilient to stress. Moreover, selective overexpression of C3 in PFC was sufficient to cause depressive-like behavior in mice. Also, we found C3aR-dependent changes in infiltrating macrophages into CNS following CUS.
Conclusions: These results studies identify C3 signaling as a key factor in MDD pathophysiology and as a possible new therapeutic target for MDD and other stress-related neuropsychiatric disorders.
Keywords: Immune, Depression, Stress Models
Disclosure: Nothing to Disclose.

T128. Deep Brain Stimulation to the Super-Lateral Part of the Medial Forebrain Bundle (slMFB) is Associated With Sustained Efficacy in Major Depression for More Than Five Years

Thomas Schlaepfer*, Bettina Bewernick, Hannah Kilian, Volker Coenen

University of Freiburg & Johns Hopkins University, Freiburg, Germany

Background: Studies of Deep Brain Stimulation (DBS) to three different targets (Brodmans Area cg25, anterior limb of the capsula interna and Nucleus Accumbens) in patients suffering from treatment resistant major depression (TRD) showed promising effects in comparable patient populations, 50-60% of patients responded with decreases in depression severity ratings by at least 50%. Stimulation intensities used in these studies ranged from 4-10V, and first clinical response was seen after about four weeks in all studies. It has been demonstrated that slMFB DBS is associated with rapidly developing antidepressant effects in patients with TRD. For none of the stimulation targets long-term outcomes have been reported.
Methods: Eight TRD patients were treated with DBS bilateral to the slMFB. Primary outcome measure was a 50% reduction in Montgomery-Åsberg Depression Rating Scale (MADRS) (response) and remission (MADRS <10) at 12 months compared to base-line. Secondary measures were anxiety, general functioning, quality of life, safety and cognition assessed for five years.
Results: Eight TRD patients were treated with DBS bilateral to the slMFB. Primary outcome measure was a 50% reduction in Montgomery-Åsberg Depression Rating Scale (MADRS) (response) and remission (MADRS <10) at 12 months compared to base-line. Secondary measures were anxiety, general functioning, quality of life, safety and cognition assessed for five years.
Conclusions: Long-term results of slMFB-DBS suggest acute and for five years sustained antidepressant effect;
Being able to induce a rapid and robust antidepressant effect even in a small, sample of TRD patients without significant psychiatric comorbidity, render the slMFB an attractive target for future studies.
Keywords: Deep Brain Stimulation, Major Depression, Neuromodulation, Treatment Resistance
Disclosure: Part 1: Medtronic, Inc., Grant, Boston Scientific, Inc., Grant, **Part 4:** Medtronic, Inc., Grant, Boston Scientific, Inc., Grant.

T129. Sphingomyelins as Predictors and Biologic Correlates of Antidepressant Response in the Co-Med Trial

Andrew Czysz*, Charles South, Bharathi Gadad, Erland Arning, Abigail Soyombo, Teodoro Bottiglieri Bottiglieri, Madhukar Trivedi

University of Texas Southwestern Medical Center, Dallas, Texas, United States

Background: Metabolomics is a developing and promising tool for exploring the molecular pathways that underlie the symptoms of depression and predicting antidepressant response. In this study, we used the AbsoluteIDQ™ p180 kit to investigate whether plasma metabolites (sphingomyelins, lysophosphatidylcholines, phosphatidylcholines, and acylcarnitines) from a sub-set of participants in the Combining Medications to Enhance Depression Outcomes (CO-MED) trial could act as predictors or biologic correlates of antidepressant response.
Methods: Participants in this trial were assigned to one of three pharmacological treatment arms: escitalopram monotherapy, bupropion-escitalopram combination, or venlafaxine-mirtazapine combination. Plasma was collected at baseline in 159 participants and again 12 weeks later at study exit in 83 of these participants. Metabolite concentrations were measured and combined with various clinical and sociodemographic variables using the hierarchal lasso to dissect whether specific metabolites are particularly informative of drug response in certain populations.
Results: Increased concentrations of sphingomyelin C26.1 or a higher monosaturated-to-saturated fatty acid ratio in participants with prior suicide attempt modeled a worse outcome for participant's change in QIDS. In contrast, an increased ratio of hydroxylated sphingomyelins relative to non-hydroxylated sphingomyelins in those with melancholic features suggested an improved QIDS score. Changes in total hydroxylated sphingomyelins, and particularly hydroxysphingomyelin C22.2, after treatment modeled improved QIDS in participants with baseline suicidal ideation. The predictive power of metabolites was not medication-specific.
Conclusions: Together, these data identify sphingomyelins, particularly hydroxylated species, as potential predictors of antidepressant treatment response, and further suggest their role in mediating treatment effect, regardless of drug choice.
Keywords: Metabolites, Depression Treatment Response, Translational Biomarker Development
Disclosure: Nothing to Disclose.

T130. Circuit Mechanisms and Predictors of Electroconvulsive Therapy (ECT) Anti-Suicidal Properties: A Dimensional fcMRI Study

Erik Lee, Tracy Barbour, Kristen Ellard, Michael Henry, Gordana Vitaliano, Joan Camprodon*

Massachusetts General Hospital, Harvard Medical School, Boston, Massachusetts, United States

Background: Electroconvulsive Therapy (ECT) is the most effective treatment in psychiatry, and among the most effective in medicine. In addition to its efficacy in depression, mania, psychosis and catatonia, it has primary anti-suicidal properties independent from its effects on the primary disorders/syndromes. Understanding the mechanisms of action of this life-saving treatment may be a strategy to identify novel and much needed targets for anti-suicidal therapies.
Methods: We studied 16 patients presenting with a major depressive episode treated with ECT. Before and after the acute course of treatment, we obtained resting state functional MRI measures, quantified depression severity with the Quick Inventory of Depressive Symptomatology

(QUIDS-SR) and suicide risk with the Concise Health Risk Tracking (CHRT).

After standard data preprocessing steps, whole-brain connectivity analyses were conducted using anterior cingulate cortex (ACC) seeds. The ACC has been previously linked to suicide risk and severity using multiple research methodologies (postmortem pathology, PET, MRI, etc.). Pearson correlations coefficients were computed between individual seed regions and all remaining brain voxels. To examine the relationship between functional connectivity and clinical outcomes, whole-brain regressions were conducted using changes in CHRT and QUIDS scores.

Results: ECT led to a significant decrease in depression severity and clinical suicide risk, confirming the expected antidepressant and antisuicidal properties of this intervention.

We identified changes in connectivity in two overlapping nodes in the right Inferior Parietal Lobule, encompassed in the fronto-parietal Central Executive Network, that both explained and predicted the improvement in suicide risk specifically (CHRT changes), but not the syndromal changes in depression severity captured with the QUIDS.

Conclusions: These results suggest a putative mechanism for the anti-suicidal properties of ECT. Changes occur across regions known to be relevant for affective regulation, self-processing and top-down executive control, all dimensions relevant to the "suicidal syndrome". These circuit-based effects represent a possible target for treatment, potentially using alternative and less invasive therapeutic modalities like transcranial magnetic stimulation (TMS). In addition, patterns predicting positive response may be used as biomarkers to support clinical decision-making and ECT treatment selection and planning.

Keywords: Electroconvulsive Therapy, Suicide, Depression, Connectivity, Novel Targets

Disclosure: Nothing to Disclose.

T131. What Symptoms Trigger Symptom-Onset Serotonin Reuptake Inhibitor Treatment in Premenstrual Dysphoric Disorder? A Prospective Evaluation

Ariadna Forray*, Kathryn Gilstad-Hayden, Kimberly Yonkers

Yale University School of Medicine, New Haven, Connecticut, United States

Background: Premenstrual dysphoric disorder (PMDD) is a severe, sometimes disabling form of premenstrual syndrome that affects from 1.2%-6.4% women globally. There is strong evidence supporting the efficacy of serotonin reuptake inhibitors (SRIs) for the treatment of PMDD, including when administered only during the second half of the menstrual cycle. While the utility of symptom-onset treatment clear, the symptoms that trigger medication initiation is not. In this study, we evaluated the symptoms that led to the initiation of symptom-onset SRI treatment.

Methods: This is a secondary analysis from a double-blind, placebo-controlled, multi-site, parallel group trial evaluating the efficacy of symptom-onset dosing with the sertraline. Women were eligible if they were between 18–48 years, had menstrual cycles of 21–35 days and met criteria for PMDD. Women with an active major depressive episode or a history of a manic or psychotic disorder, current treatment with a psychotropic medication or oral contraceptive interfering in normal menses were not eligible. Women were randomized to either sertraline or placebo in a 1:1 ratio, instructed to start taking pills when they perceived symptoms and continue taking pills until menses. Treatment included six menstrual cycles. We sought to understand whether any particular symptoms "triggered" pill taking. To do this we utilized descriptive statistics to evaluate the frequency of moderate to extreme symptoms (scores 4, 5 and 6) from the Daily Record of Severity of Problems (DRSP) on day they started medication (day 0) and two preceding days (day -1 and -2, respectively).

Results: A total of 252 were randomized (125 to sertraline and 127 to placebo). Women were predominantly white 69% ($n = 175$), college educated 91% ($n = 229$) and living with a partner or spouse 50% ($n = 127$) with a mean age (SD) of 34.2 (6.1). The most commonly reported symptoms on day 0 were: fatigue 31.4% (95% CI: 25.3%, 37.5%), marked anxiety 27.4% (95% CI: 21.5%, 33.2%), irritability 26.0% (95% CI: 20.3%, 32.3%), mood lability 24.2% (95% CI: 18.8%, 29.8%), and sleep disturbance 22.4% (95% CI: 17.0%, 27.9%). Sensitivity to rejection was the symptom with the greatest increase in prevalence from day -2 to day 0, 6.6% to 14.2% to 22.4%, respectively. This was followed by: anhedonia, 4.6% to 9.1% to 12.6%; headache 5.1% to 9.6% to 17.5%; breast tenderness, 5.6% to 9.1% to 17.0%; and irritability 11.7% to 20.3% to 26.0%; from day -2 to day 0 respectively. There was also a marked increase in the mean (SD) number of women experiencing "extreme" severity of symptoms on day 0 compared to days -1 and -2: 5.1 (3.1), 1.7 (1.6) and 2.1 (1.3), respectively.

Conclusions: This is the first prospective study of this size to evaluate the symptoms related to symptom-onset dosing of an SRI for PMDD. Symptoms related to irritability, mood lability and anxiety, which comprise three of the core symptoms of a PMDD diagnosis, were among the most commonly experienced when women decided to begin medication administration. The other two symptoms commonly experienced were fatigue and sleep disturbance. These symptoms would all contribute to substantial functional impairment and thus likely influenced the decision to begin medication treatment.

Keywords: Premenstrual Dysphoric Disorder, Symptom-Onset Treatment, Serotonin Reuptake Inhibitors

Disclosure: Nothing to Disclose.

T132. A Multidimensional Analysis of Mood Disorder Interactions With Anticipatory Cortisol Reactivity in Resting State and Memory-Based fMRI

Amy Peters, Leah Kling, Kelly Ryan, Anne Weldon, Monica Starkman, Melvin McInnis, Jon-Kar Zubieta, Robert Welsh, Sara Weisenbach, Scott Langenecker*

University of Illinois at Chicago, Chicago, Illinois, United States

Background: HPA axis dysregulation plays a critical role in understanding of risk for mood disorders. However, HPA axis dysregulation is not a uniform abnormality in mood

disorders, nor does it uniformly predict symptom severity, although some links to comorbidity and treatment resistance have been reported. In addition to circadian disruptions reported in mood disorders, there is also evidence for elevated reactive or anticipatory cortisol responses to stressors. In our own work, we have attempted to understand the role of between group differences in mood disorder as well as individual differences in anticipatory stress reactivity. We have reported that salivary cortisol prior to and after functional MRI may reflect both the anticipatory stress response to the expectation of stress, as well as the experience of the confining and loud scanner environment. Thus, pre-scan salivary cortisol was related to degree of reactivity of amygdala and subgenual anterior cingulate to faces (Weldon et al., 2015). Furthermore, both dorsal and mid cingulate activation was elevated in a proportional relationship to pre-scan cortisol levels. Furthermore, in those with mood disorders, there was an attenuation of activation with increasing pre-scan cortisol levels, an interaction between disease and pre-scan cortisol on activation to emotional faces (Peters et al., 2016). These effects were most prominent for all faces in dorsal anterior cingulate, middle temporal gyrus, precuneus, and both caudate and putamen. Additional regions showed specific interactions between cortisol and activation to fearful and angry faces for those with mood disorders. The present study intended to extend the prior findings by evaluating whether the cortisol by mood disorder interactions in activation were related to declarative memory. We also investigated whether pre-scan cortisol was related to resting state connectivity of default mode and salience and emotion networks, respectively.

Methods: Two independent samples were collected. The first sample include 62 adults (age 18-65) with current major depressive or bipolar disorder in comparison to 39 age, sex, and education matched healthy comparison (HC) participants. The individuals provided pre-scan cortisol and the sample is the same as reported in Peters et al., 2016. We now investigated how performance on and activation during the Semantic-cue List Learning Test (SLLT) was predicted by pre-scan cortisol levels. We also investigated the interaction between disease (combined mood disorder group vs HC) and anticipatory cortisol in activation. We used both block (encoding vs silent rehearsal) and event-related (later recalled vs not recalled) analysis strategies to build first level models. Second level models included predictors of pre-scan cortisol, depression symptoms, movement parameters, sex, time of scan, diagnostic group, and diagnostic group by cortisol interaction term in a regression model in SPM8.

The second sample included young adults (aged 18-30) with a history of mood disorder, currently in the full or partially remitted state ($N=73$) compared to age, sex, and education matched HC group ($N=47$). This sample completed an eight-minute, eyes-open resting state scan and contributed pre-scan salivary cortisol samples. Resting state models included use of seed-based connectivity analyses, with 2 seeds from the salience and emotion network (SEN, amygdala and subgenual anterior cingulate) and 2 seeds from the default mode network (DMN, posterior cingulate and hippocampus). Rs-fMRI analyses were conducted with homebuilt scripts that include movement, white matter, CSF regressors within a principal components model. Note that we do not censor data or use global signal regression (Jacobs et al., 2014, Jo et al., 2013, Saad et al., 2017).

Results: SLLT: Overall, pre-scan cortisol level was positively associated with greater engagement of fronto-limbic activation for encoding - silent rehearsal contrast. However, pre-scan cortisol in MD was associated with attenuated activation of activation in a number of regions, including the right medial frontal, bilateral superior and right middle temporal gyri, right insula, left lingual gyrus, and left claustrum relative to HCs. The same pattern of MD-associated attenuation with increasing pre-scan cortisol was also observed for the words recalled –not recalled contrast in the right ventral anterior cingulate, left hypothalamus, and left middle temporal gyrus. Activation in right middle frontal gyrus was related to performance, but only for MD group.

Rs-fMRI: Large whole-scale, decreased within-and cross-network relationships were observed in relation to pre-scan cortisol, for DMN and SEN seeds. However, this pattern was inverted in those with remitted MDD. For both DMN and SEN, this increased resting state connectivity was observed for dorsal anterior cingulate and medial prefrontal cortex (all seeds), precuneus (DMN seeds), brain stem and cerebellum (all seeds), and bilateral DLPFC (all seeds, with some laterality effects).

Conclusions: Anticipatory salivary cortisol levels reflect prominent individual differences for healthy subjects and interact with disease status in both task-based results (emotion, memory) and resting state analyses with key networks. The effects of cortisol on activation and connectivity are widespread, but tend to coalescence around some key regions. Precuneus, dorsal cingulate, and dorsomedial prefrontal cortices were implicated in prior emotion, current memory, and current resting state analyses. Temporal and medial and lateral prefrontal cortical areas were not uniformly involved, but more so in disease by cortisol interactions for rs-fMRI analyses. Together, these results implicate the importance of HPA axis functioning as a primary and interactive driver of activation in fMRI studies.

Keywords: Cortisol, Memory, Resting State Functional Connectivity, Mood Disorders

Disclosure: Nothing to Disclose.

T133. Preliminary in Vivo Evidence for mGluR5 as a Potential Biomarker to Differentiate Major Depressive Disorder From Bipolar Disorder

Sophie Holmes*, Maggie Davis, Nicole DellaGioia, Jean-Dominique Gallezot, Nabeel Nabulsi, Gustavo Angarita, Richard Carson, Hilary Blumberg, Irina Esterlis

Yale University, New Haven, Connecticut, United States

Background: Major depressive episodes are common to both major depressive disorder (MDD) and bipolar disorder (BD). In BD, depressive episodes are the most common episodes presenting to clinicians, with patients spending three times as much time depressed as manic or hypomanic1. Furthermore, the depressed episode is associated with a high rate of suicide2, 3. However, current treatments for bipolar depression have limitations in their efficacy and may contribute to

worsening of manic symptoms and cycling in some individuals4. At present, differentiating BD from MDD relies solely on the presence or absence of manic/hypomanic episodes, which can lead to misdiagnosis and improper treatment. Little is known about the neurobiological differences between depressive episodes of the two disorders. Elucidating these distinct mechanisms is key in accurate diagnosis and in developing critically-needed, targeted treatments for bipolar depression. Glutamate dysfunction is thought to be involved in depression of both BD and MDD5, but whether there are specific differences in the glutamate system between the two disorders is unclear. The metabotropic glutamate receptor 5 (mGluR5) has been implicated in the symptoms of mood disorders and represents a promising treatment target6. Using positron emission tomography (PET) and [18F]FPEB, a radiotracer that binds to the mGluR5 with high specificity, we aimed to investigate mGluR5 availability in the prefrontal cortex (PFC) in BD depression and MDD for the first time.

Methods: Eighteen unmedicated individuals meeting DSM criteria for a current depressive episode [9 with BD depression (age = 42 ± 14years, MADRS = 18 ± 6), 9 with MDD (age = 40 ± 12years, MADRS = 21 ± 4)], and 18 healthy comparison (HC) individuals (41 ± 4years) participated in one [18F]FPEB PET scan to quantify mGluR5 in-vivo, as well as psychiatric symptom assessment and cognitive testing. Cognitive functioning was assessed using the CogState cognitive battery. BD and MDD groups were matched for age, sex and smoking status. The outcome measure was volume of distribution (VT: the ratio of activity in tissue relative to that in blood), computed using a venous input function. The radiotracer was injected as bolus plus constant infusion and subjects were scanned during steady state (90-120mins post-injection). VT values were compared across groups in a composite PFC region consisting of orbitofrontal cortex, ventromedial and dorsolateral PFC.

Results: A univariate ANOVA with mean mGluR5 availability (VT) as dependent variable and diagnosis as independent variable revealed a main effect of diagnosis ($F2,33 = 3.583$, $p = 0.039$). Post-hoc independent-samples t-tests indicated significantly lower mean mGluR5 availability in BD compared to HC individuals by an average of 17% ($p = 0.02$, Cohen's $d = 1.01$), but no difference between MDD and HC individuals ($p = 0.24$). mGluR5 availability was not significantly different between BD and MDD individuals ($p = 0.17$), though there was a 10% lower mGluR5 availability in BD vs MDD individuals and a moderate effect size ($d = 0.67$). Within the BD group, there was a significant negative correlation between dlPFC mGluR5 availability and scores on a psychomotor function task ($r = -0.77$, $p = 0.016$).

Conclusions: Our findings indicate lower PFC mGluR5 availability in BD compared to HC individuals, but no difference in mGluR5 availability between MDD and HC groups. We also observed a difference in mGluR5 availability between BD and MDD groups, although this did not reach statistical significance. The observed differences in mGluR5 availability could reflect differences in glutamate neurotransmission between depressive episodes in BD and MDD. Specifically, lower mGluR5 in BD may be associated with excessive glutamate signaling. Indeed, we previously showed that an increase in glutamate neurotransmission is associated with a downregulation of mGluR57, and magnetic resonance spectroscopy (MRS) studies indicate higher levels of glutamate in BD individuals in a depressive episode8. A larger cohort of MDD and BD individuals is required to determine whether mGluR5 might be a biomarker to distinguish the two disorders during a depressive episode. However, current data point to the potential role of mGluR5 dysfunction in BD during a depressive episode, which is in line with preclinical and postmortem evidence.

Keywords: PET Imaging, Bipolar and Unipolar Depression, Glutamate Receptor Activity

Disclosure: Nothing to Disclose.

T134. Chemogenetic Stimulation of Retina Activates Locus Coeruleus Neurons and Prevents the Development of Light Deprivation-Induced Depression-Like Behavior

Hannah Bowrey*, Morgan James, Mohsen Omrani, Aida Mohammadkhani, Gary Kane, Gary Aston-Jones

Rutgers University, Piscataway, New Jersey, United States

Background: Chronic light deprivation induces a depressive-like phenotype, which is due to a locus coeruleus noradrenaline (LC-NA)-dependent mechanism (Gonzalez and Aston-Jones, PNAS 2008). Suprachiasmatic nucleus (SCN) provides an indirect circadian input onto LC via a relay in dorsomedial hypothalamus (DMH) (Aston-Jones et al., Nat. Neurosci. 2001). SCN is therefore in a key position to integrate light information with LC, via the pathway: retina → SCN → DMH → LC. We refer to this pathway as the Photic Regulation of Arousal and Mood (PRAM) pathway (Bowrey et al. Depress. Anxiety 2017). Here we test the hypothesis that increasing PRAM pathway activity by chemogenetic manipulation of the retina will activate LC and prevent darkness-induced depression-like behavior.

Methods: Experiment 1. Sprague Dawley rats received intraocular injections of an AAV encoding a Gq-linked designer receptor exclusively activated by designer drugs (DREADD: AAV2-hSyn-hM3D(Gq)-mCherry; n = 12), control virus (AAV2-hSyn-EGFP; n = 10) or no virus (n = 10). Rats were placed in continuous darkness for 8 wk, and those that received virus were concurrently subjected to daily ip injections of the DREADD agonist clozapine-N-oxide (CNO). Rats were then subjected to assays of mood (saccharin preference test, elevated plus maze and forced swim test) and vision (electroretinagram: ERG). LC tissue was stained for Poly ADP ribose polymerase (PARP) and tyrosine hydroxylase (TH, a marker of NA neurons).

Experiment 2. To determine if the PRAM pathway is functional, we tested whether the LC responds to chemogenetic activation of the retina. We dark-adapted two groups of rats (either AAV2-hSyn-hM3D(Gq)-mCherry, n = 6 or AAV2-hSyn-EGFP, n = 10) for 12 hours before recording activity of LC neurons in anesthetized rats that had masked eyes to block light input. We also conducted immunohistochemistry for the immediate early gene protein cFOS, a marker of neuronal activity in SCN, DMH and LC, to determine whether key structures of the PRAM pathway were stimulated during chemogenetic activation of the retina.

Experiment 3. To determine the retinal cell-type responsible for depression-like behavior, intrinsically photosensitive

retinal ganglion cells (ipRGCs) of animals raised in 12:12 light:dark conditions were ablated using a saporin toxin that selectively eliminates melanopsin-expressing cells (Mel-SAP; n = 10). Two control groups received intraocular injections of vehicle (0.01 M PBS) and were kept in either continuous darkness (n = 10) or in 12:12 light:dark conditions (n = 10). Ten weeks later, rats were subjected to identical analyses as those in Experiment 1.

Results: Experiment 1. ERG analysis showed that DREADD stimulation increased RGC activity. Constant darkness (8 wk) induced a depression-like phenotype in control animals, which was prevented by daily activation of retinal DREADDs. Experiment 2. DREADD stimulation of retina increased the tonic firing rate of LC by 60% and was associated with increased FOS expression in SCN, DMH and LC. Experiment 3. Mel-SAP lesions of melanopsin RGCs induced a depression-like phenotype. This was also associated with increased apoptosis in LC-NA cells as seen with increased PARP staining.

Conclusions: Dysregulation of the PRAM pathway may induce neural damage in LC-NA neurons, and this damage is associated with a depressive behavioral phenotype, which can be prevented by PRAM-induced activation of LC. The PRAM pathway presents a novel circuit for the regulation of mood, and thus a possible new direction for the treatment of depression in humans.

Keywords: Depression, Locus Coeruelus, Retina, Novel Therapeutics, Light Therapy

Disclosure: Nothing to Disclose.

T135. The Tryptophan-Kynurenine Pathway in Bipolar Depression: Effects of Inflammation Modulation

Stephen Murata*, Monica Feliz Castillo, Markus Schwarz, Brendan Martin, Angelos Halaris

Loyola University Chicago Stritch School of Medicine, Maywood, Illinois, United States

Background: In connection with the emerging link between treatment resistance and neuroprogression in bipolar disorder, it is increasingly apparent that clinical remission is achieved in part through pharmacologically induced arrest of neuroprogression. Literature increasingly suggests that bipolar and unipolar depression are chronic inflammatory states that drive underlying abnormalities in the tryptophan-kynurenine pathway (TRYCATs). Specific pro-inflammatory cytokines are potent upregulators of indoleamine-2,3-dioxygenase (IDO) activity. IDO is a rate-limiting enzyme that shunts tryptophan from 5-HT synthesis towards neurotoxic and excitotoxic TRYCATs including the NMDA receptor agonist, quinolinic acid (QUIN). Since the cyclooxygenase type 2 (COX-2) selective inhibitor, Celecoxib (CXB), can reduce the pro-inflammatory drivers of IDO, and because an accepted indirect measure of IDO activity is the kynurenine/tryptophan ratio, we hypothesized that the pattern of this ratio (and its constitutive components) over the treatment course may reflect differences in tryptophan handling that contribute (at least in part) to the 4.13-fold ($p = 0.04$) and 14.34-fold ($p < 0.001$) increase in response and remission rates, respectively, we have observed. The purpose of this study was to characterize potential predictive and/or therapeutic biomarkers in the TRYCATs pathway to stratify patients for CXB augmentation.

Methods: This was a randomized, double-blind, two-arm, placebo-controlled study consisting of a screening visit, a 2-week washout, and a 1-week placebo run-in phase. Subjects who met study criteria and were not placebo-responders underwent a physical exam, medical history, routine laboratory tests, and completed a number of rating instruments. Subjects at baseline were rated in a blinded manner, and those who persisted in scoring ≥ 18 on the 17-item Hamilton Depression scale (HAM-D) were randomized to receive escitalopram (ESC) + CBX, or ESC + placebo. The entire cohort of 70 patients (35 in each arm) received 8 weeks of treatment to qualify as study completers. A database of normative (untreated) data was already collected in a previous study. Blood levels of inflammation biomarkers, TRYCATS, and CBX and ESC were determined at baseline, weeks 4 and 8. A Wilcoxon Signed Rank Sum test was used to assess for intra-group differences in TRYCAT metrics between baseline and week 8 of treatment. A Wilcoxon Rank Sum test was then used to examine inter-group differences in TRYCAT metrics from baseline to week 8, with respect to response or remission status in the ESC + CBX arm.

Results: Kynurenine, tryptophan, and kynurenine/tryptophan ratio level change scores did not vary by treatment arm. Kynurenine and kynurenine/tryptophan also showed no significant difference in change scores or inter-group levels with respect to treatment response or remission status, within the ESC + CBX arm. Tryptophan was significantly reduced within the ESC + CBX arm with respect to both treatment response ($p = 0.05$) and treatment remission ($p = 0.03$), confirmed by a significant reduction in tryptophan change scores according to treatment response ($p = 0.02$) and remission ($p = 0.02$).

Conclusions: The robust clinical response to CXB (in preparation) supports the concept that inflammation modulation strategies can help overcome refractoriness to antidepressant treatment in bipolar depression. Comparable findings have been reported in the literature for unipolar depression. Here we sought to determine whether the benefit of CXB augmentation is predicted and/or mediated by mechanisms involving the kynurenine pathway. The fact that baseline IDO activity (controlled for baseline tryptophan and kynurenine levels) did not predict response to CXB suggests that pre-existing differences in TRYCAT regulation, at least at the IDO level, is insufficient to identify potential responders. It further suggests the benefits of CXB may become manifest downstream of IDO, or in a manner dependent on CXB but independent of TRYCAT regulation altogether.

Our findings in unipolar depression showed that ESC monotherapy increased tryptophan and decreased QUIN in a manner largely independent of inflammatory modulation. This was consistent with the working hypothesis that CBX reverses the shunting into TRYCAT pathway, and promotes pooling of tryptophan for 5-HT synthesis. However, in bipolar depression, we obtained findings (both negative and positive) in the opposite direction: tryptophan increased and QUIN remained constant (consistent with unchanged IDO activity) as a result of inflammatory modulation. Is this reversal a function of CBX or bipolarity? Emerging reports suggest that CBX may reduce constitutive IDO expression,

which can plausibly mask a net decrease in IDO output behind a stable kynurenine/tryptophan ratio. But when considered in relation to unchanged kynurenine and decreased tryptophan, this unchanged ratio may be a clue for a separate immune-related tryptophan utilization, including accelerated tryptophan shunting into 5-HT synthesis, a possible overriding of ESC-mediated tryptophan rescue by a COX-2 dependent mechanism, or some other functional pathway unique to the neuroprogressive signature in bipolar depression.

Keywords: Bipolar Disorder, Kynurenine Pathway, Celecoxib, Inflammation, Indoleamine 2,3-Dioxygenase

Disclosure: Nothing to Disclose.

T136. Fractal Dimension of Subcortical Brain Structures May Predict the Risk for Depression

Zhishun Wang*, Ardesheer Talati, Zhiyong Huo, Connie Svob, Myrna Weissman, Jonathan Posner

Columbia University, New York, New York, United States

Background: Familial history of depression increases its risk in biological offspring. We previously found familial depression to be associated with reduced cortical thickness in frontal and parietal regions [1], with effects remaining stable across two time points 8 years apart [2]. Here, we used fractal dimension (a complexity measure of a spatial or temporal pattern with self-similarity) to investigate longitudinally the impact of familial depression on the morphometry of subcortical brain structures, including amygdala/hippocampus complex and basal ganglia nuclei.

Methods: Data derive from a 30-year three-generation study with continuing clinical assessments ($N=200$) [3]. Multimodal magnetic resonance imaging (MRI) was performed with high-resolution anatomical MRI scans during Waves 5 and 6 (8 years apart). Wave 5 comprised 143 biological offspring, 83 at high familial risk for major depression disorder (33.12 ± 12.69 yrs; 37 males), 60 at low risk (26.72 ± 13.22 yrs; 26 males). Wave 6 comprised 82 biological offspring,43 at high risk (39.42 ± 12.68 yrs; 20 males), 39 at low risk (31.24 ± 12.79 yrs; 17 males). At Wave 5, T1-weighted anatomical images were acquired on a Siemens Sonata 1.5T scanner using a 3D MPRAGE pulse sequence with spatial resolution of 1.17x1.17x1.2 mm. At Wave 6, 3D FSPGR anatomical images were acquired on a GE Signa 3T scanner with a spatial resolution of 0.98x0.98x1.0mm. We segmented each anatomical image into 15 subcortical regions (brain stem, left & right nucleus accumbens, left & right putamen, left & right caudate, left & right palladium, left & right thalamus, left & right hippocampus, left & right amygdala) using a Bayesian model-based segmentation algorithm implemented in FSL FIRST toolbox[4]. We used a method we developed previously [5] to calculate the fractal dimension of each of the 15 segmented subcortical structures. We performed group comparisons of the fractal dimension features using general linear model (GLM) on each of the 15 structures between risk groups within each wave, covarying for age and sex and correcting p-value for multiple comparisons using Bonferroni correction ($p<0.05/15$ to reach a corrected $p<0.05$). We tested whether these comparisons were reproducible at Waves 5 and 6 and calculated Pearson correlations of the fractal dimension features between the two waves.

Results: Wave 5: Compared with the low risk group, the high-risk group (full sample: $n=143$) had a reduced fractal dimension in the right nucleus accumbens, left and right pallidum, right thalamus, and an increased fractal dimension in the left amygdala. When restricted to the participants who also had scanning data during W6 ($n=82$), the high-risk group had a reduced fractal dimension in the right caudate, left pallidum, right hippocampus, left and right thalamus, and an increased fractal dimension in the left amygdala. Wave 6: Compared with the low risk group, the high-risk group had reduced fractal dimension in the left and right caudate, right pallidum, and left and right hippocampus. Stability of Fractal Dimension between Waves 5 and 6: The fractal dimension features of 15 subcortical brain structures were significantly correlated ($p<0.05/15$, Bonferroni correction) between Wave 5 and Wave 6 in the bilateral caudate, bilateral hippocampus, and bilateral pallidum.

Conclusions: We find that familial risk for depression is associated with significant reductions in complexity of subcortical morphology in the caudate, pallidum, and hippocampus. Findings were stable across time (two MRI scans, different scanners, 8 years apart). The findings are consistent with the role of the aforementioned regions in depressive illness. Recent brain imaging studies have shown that the hippocampus undergoes selective shape shrinking in stress-related neuropsychiatric disorders such as depression. Pallidum structure is believed to control the hedonic tone and reward sensitivity. Reduction of complexity in pallidum may therefore relate to anhedonia. The caudate, part of the striatum, plays a key role in cognitive control. The abnormalities of striatum structure may cause cognitive dyscontrol in persons, which may predict a risk for the development of depression. Taken together, our findings suggest that the fractal dimension of subcortical brain structures could be a biomarker to predict the risk for depression.

References

1. Peterson BS, Warner V, Bansal R, Zhu H, Hao X, Liu J, Durkin K, Adams PB, Wickramaratne P, Weissman MM. Cortical thinning in persons at increased familial risk for major depression. Proc Natl Acad Sci. 2009; 106(15):6273-6278.

2. Hao X, Talati A, Shankman SA, Liu J, Kaiser J, Tenke CE, Warner V, Semanek, D, Wickramaratne PJ, Weissman MM, Posner J. Stability of cortical thinning in persons at increased familial risk for major depression across eight years. Biological Psychiatry: Cognitive Neuroscience and Neuroimaging. 2017, in press.

3. Weissman MM, Berry OO, Warner V, et al. A 30-Year Study of 3 Generations at High Risk and Low Risk for Depression. JAMA Psychiatry. 2016;73(9):970-977.

4. Patenaude B, Smith SM, Kennedy D, Jenkinson MA, Bayesian Model of Shape and Appearance for Subcortical Brain. NeuroImage. 2011; 56(3):907-922.

5. Zhao G, Denisova K, Sehatpour P, Long J, Gui W, Qiao J, Javitt DC, Wang Z. Fractal Dimension Analysis of Subcortical Gray Matter Structures in Schizophrenia. PLoS One. 2016 May 13;11(5):e0155415.

Keywords: Depression, MRI, Subcortical Shape Analysis

Disclosure: Nothing to Disclose.

T137. Association of Fasting Insulin Levels and Depression Severity With Resting-State Functional Connectivity in Depressed Adolescents

Sarah Leslie, Elizabeth Weisman, Alexis Staver, Danielle Wall, Alma Andrade, Yevgeniya Zaiko, Alexander Onopa, Melissa Packer, Owen Phillips, Natalie Rasgon, Manpreet Singh*

Stanford University School of Medicine, Stanford, California, United States

Background: Depression paired with insulin insensitivity is a growing problem in youth worldwide. While insulin resistance and mood disorders have typically been compartmentalized as distinct conditions, recent evidence suggests that shared mechanisms may link insulin resistance, obesity, and worsening depression. Among the most compelling, are mechanisms that involve aberrant connectivity in brain networks involved in approach motivation, which are relevant in this population due to the potential presence of food reward-seeking, anhedonia, or a combination of both, due to anhedonic compensation. To investigate whether these shared mechanisms interact, we analyzed resting state functional connectivity data in relation to fasting insulin and clinical measures of depression in a sample of overweight, depressed youth.
Methods: Resting-state functional MRI data and fasting insulin levels were collected from 36 youth, aged 9 to 17, who are overweight or obese and experiencing moderate to severe symptoms of depression. Using an anterior cingulate (ACC) seed-based region of interest analysis, we investigated inter-subject associations between fasting insulin levels and resting-state functional connectivity (RSFC). Individual RSFC measures were then extracted from significant ACC/insulin connectivity regions and correlated with scores on the Children's Depression Rating Scale-Revised (CDRS-R).
Results: Higher levels of fasting insulin were associated with alterations in resting-state connectivity between the bilateral ACC and the putamen, nucleus accumbens (NAcc), and precuneus. Higher levels of fasting insulin and greater depression severity both correlated with stronger ACC-putamen/NAcc connectivity and with stronger negative ACC-precuneus connectivity.
Conclusions: Alterations in resting-state connectivity between the ACC and the precuneus, and between the ACC and the putamen/NAcc, are associated with both fasting insulin and with severity of depression symptoms, suggesting a neural interaction linking impaired insulin sensitivity and depression. Future longitudinal investigations that add a temporal component to understanding the relations among these systems involved in approach motivation and reward, may help elucidate the shared mechanisms underlying metabolic disorders and mood disorders. By understanding when and how disruptions in these systems emerge, we may be able to identify ways to successfully prevent or treat them.
Keywords: Frontostriatal, Resting State Functional Connectivity, Adolescent Depression, Insulin Resistance, Obesity
Disclosure: Part 1: National Institute of Mental Health, Grant, National Institute of Aging, Grant, Brain and Behavior Foundation, Grant, Stanford Child Health Research Institute, Grant, Johnson and Johnson, Grant, Neuronetics, Grant, Sunovion, Advisory Board.

T138. Effects of Early Life Stress on Depressive and Metabolic States in Adult Individuals at Genetic Risk

Benedetta Bigio*, Francis Lee, Danielle Zelli, Timothy Lau, Paolo de Angelis, Anjali Ferris, Susan Harvey, Jonathan Lai, Anisha Kalidindi, Natalie Rasgon, Bruce McEwen, Carla Nasca

Rockefeller University, New York, New York, United States

Background: Early life experiences play a powerful role in adulthood mental and physical health. For example, naturally occurring variations in maternal care are known to increase the individual likelihood to depressive disorders in adulthood. Arising from the postulate of insulin resistance (IR) as a pathophysiological link in diabetes, depression and dementia ("the 3 D's"), converging literature supports a role for the epigenetic modulator of pathogenic mechanisms common to the 3D's, acetyl-L-carnitine (LAC), as a biomarker to identify a severe phenotype of depression characterized by greater severity and earlier onset. Moreover, low LAC levels are associated with childhood abuse, specifically emotional neglect in subjects with treatment resistant depression. Reverse translational studies rodents with depressive-like phenotypes have reduced LAC levels in the plasma and mood regulatory brain regions (e.g.: hippocampus and prefrontal cortex) and that low LAC levels are associated with IR.
Methods: We implemented a novel mating model using wild-type or heterozygous BDNF val66met (hets) dams and controlling for paternal care to assess the effects of maternal care on molecular, metabolic and behavioral outcomes in male hets offspring. in individuals at genetic risk for the development of the depression, diabetes, progressing to dementia (3D's). A computerized setup was devised for recording of the behavioral observation sessions (i.e., three sessions/day between postnatal days 1 and 7, between 7:00pm and 8:00pm, 12:00am and 1:00am and 4:00am and 5:00am with red lights on and animals in their active phase). Behavioral analysis was performed on the basis of previously employed ethological parameters. Behavioral and molecular analyses in adulthood mice were performed as previously described by four observers who were blind to the sample conditions. For the RNAseq, we used a design with balanced-blocks by multiplexing samples, in order to eliminate potential confounding caused by lane effects. Thus, each sample was provided with a unique adapter and all samples were put in the same pool and sequenced with Illumina Hiseq2500 to yield the desired sequencing depth. The Fastq files were evaluated for quality control using FastQC and trimmed with Trimmomatic. Alignment was performed using the Tophat2 and statistical analyses were performed with EdgeR. Genes of interest were confirmed by RT-PCR and ran in triplicates. Two-tailed t-tests and multiple regression were used as appropriate to specific analyses.
Results: We find that wild-type and hets dams differ in the amount of maternal care provided to offspring assessed by in and out of the nest caring and self-maintenance behaviors.

Our data show that depressive-like states manifest only in individuals at genetic risk (hets) that receive less maternal care. Indeed, hets receiving less maternal care, as compared to hets receiving more maternal care, show depressive-like traits, including abnormal social interactions. These behavioral differences are concomitant with a peripheral metabolic dysregulation (i.e., IR) and reduced LAC levels. Molecularly, we find striking different transcriptomic profiles in the hippocampal ventral dentate gyrus (vDG), a critical brain region for antidepressant responses. Among the most significant differentially expressed genes, RNAseq, qPCR and protein analyses revealed a reduction in the known marker of stress responses mGlu2, a regulator of glutamate tone, in the vDG of less nurtured hets as compared to high nurtured hets. Finally, at the behavioral characterization, more aged hets that received low nurturing showed cognitive impairments in spatial and contextual memory at the Y-maze, suggestive of dementia-like symptoms, which were not apparent before. Such cognitive impairments were not observed in high nurtured hets at either age.

Conclusions: These findings suggest that early life experience have huge influences on adulthood depressive and metabolic states in individuals at genetic risk. Our findings also point to positive maternal care as a mean to "block" manifestation of a genetic predisposition.

Keywords: Early Life Stress, Insulin Resistance, Depression, Neuroendocrinology, Maternal Behavior

Disclosure: Nothing to Disclose.

T139. Midbrain Microcircuit Dysfunction in Repeated Social Stress

Stacy Ku*, Carole Morel, Allyson Friedman, Barbara Juarez, Ming-Hu Han

Icahn School of Medicine at Mount Sinai, New York, New York, United States

Background: Major depressive disorder (MDD) is a highly debilitating mental disorder that afflicts over 15% of the US population. While chronic stress is a precipitating factor for the development of depression, MDD is highlighted by a complex etiology and heterogeneous symptomology, and current therapeutics are beneficial in only a subset of patients. A number of clinical studies have found dysfunctional gamma-Aminobutyric acid (GABA) neurotransmission in MDD patients. Preclinical models of stress-induced depression have shown neuroadaptations within the mesocorticolimbic circuit that may underlie the anhedonic aspect of depression. In particular, the ventral tegmental area (VTA) is a key substrate for mediating susceptibility or resilience to chronic stress. We recently revealed that the VTA dopaminergic (DA) projection to the nucleus accumbens (VTA-NAc) is critical for the expression of social avoidance and anhedonia observed in susceptible mice (Chaudhury D et al., Nature 2013; Friedman AK et al., Science 2014). While there are rich local GABA synaptic inputs onto VTA-NAc DA neurons (Changeux JP, Nat Rev Neurosci 2010), their role in regulating social behaviors is unknown. Thus, we investigated the local GABA inhibitory control of VTA DA activity in encoding behavioral maladaptations.

Methods: Repeated social defeat stress (RSDS) is a well-established animal model of depression that utilizes chronic stress to induce long-lasting neurophysiological and behavioral changes that are reversible by chronic antidepressant treatments. While only "susceptible" subjects develop depression-like behaviors including social avoidance and anhedonia, a subset of animals exhibit control-like "resilient" behaviors. To identify the contribution of VTA GABAergic microcircuit regulation of VTA-NAc DA neuroadaptations involved in susceptible mice following RSDS, we utilized viral-mediated techniques with transgenic mice for cell- and circuit-specific investigations in combination with electrophysiological and optogenetic techniques.

Results: We first investigated the neurophysiological changes in VTA GABA neurons following RSDS in GAD-Cre mice. Using in vitro electrophysiology and in vivo phototagging recordings of VTA GABA neurons, we found that susceptible mice displayed significantly decreased VTA GABA firing activity and intrinsic excitability, as compared to control mice. Using an optostimulation rescue paradigm, we demonstrated the necessity of VTA GABA microcircuit function and reversed the social avoidance behavior of susceptible mice. To define the microcircuit role in social avoidance behaviors, we determined the inhibitory control of the VTA-NAc DA pathway encoding social avoidance. We recorded the spontaneous inhibitory postsynaptic currents on VTA-NAc DA neurons, and determined decreased inhibitory tone on VTA-NAc DA neurons in susceptible mice compared to control. We will determine the sufficiency of VTA GABA hypoactivity in inducing susceptible behaviors, before we publish our findings.

Conclusions: While VTA DA neuron dysfunction has been implicated in social avoidance behaviors, the role of VTA GABA synaptic transmission in VTA DA-mediated social behavior have not yet been investigated. We show here that VTA GABA neurons exhibit a potent inhibitory control on DA neuronal activity, and a pathological decrease in VTA GABA neuron activity and excitability underlie social avoidance behaviors. To further dissect the VTA microcircuit, we revealed a decreased inhibitory tone on VTA-NAc DA neurons in susceptible mice. These findings emphasize the importance of parsing the mechanisms of GABA neurotransmission in depression to identify novel drug candidates for treatment of MDD, and provide insight into the neuronal cellular and circuit composition of stress-induced depression.

Keywords: Social Defeat Stress, Electrophysiology, Mesolimbic Circuitry

Disclosure: Nothing to Disclose.

T140. Vascular Endothelial Growth Factor in Bipolar Depression: Potential Biomarker for Diagnosis and Treatment Outcome Prediction

Arielle Cohen*, David Edberg, Brendan Martin, Angelos Halaris

Loyola University Chicago Stritch School of Medicine, Maywood, Illinois, United States

Background: Vascular Endothelial Growth Factor (VEGF) is involved in brain plasticity and neurogenesis. It has been

implicated in the neurotrophic model of depression, which is based on the finding that stress can cause a decline in levels of neurotrophins. In contrast, VEGF levels appear to increase in mood disorders. Notably, VEGF doesn't cross the blood brain barrier, and so it may work through effects on brain vasculature. Some studies show VEGF levels are increased in patients with bipolar disorder and decreased after successful treatment with lithium. We sought to explore this relationship and hypothesized that modulation of inflammation by co-administration of a cyclooxygenase (COX-2) inhibitor, celecoxib, would enhance this neurotrophic factor in the depressive phase of bipolar disorder.

Methods: Participants were bipolar depressed patients whose depression failed to remit following one or more adequate trials with an antidepressant, or who were experiencing a breakthrough depressive episode despite being maintained on a mood stabilizer and/or an atypical antipsychotic agent. In a double-blind study, patients were placed on escitalopram and randomized to receive add-on celecoxib (27 participants) or placebo (21 participants). There were 42 healthy controls. Plasma levels of VEGF were drawn at three time points over eight weeks.

A univariable exact logistic regression analysis was used to assess associations between remission and drug therapy. A binomial distribution was specified for the response variable, while a logit link was used to estimate the odds ratio for the explanatory variable against a referent. A Wilcoxon Rank sum test and univariable exact logistic regression analysis were used to compare VEGF levels. A linear mixed effects model was used to estimate VEGF values over time by drug therapy after adjusting for sex, age, and BMI.

Results: Patients receiving Celebrex were 14.34 (95 CI: 2.59-153.17) times more likely to experience remission compared to those on the placebo ($p < 0.001$).

The Wilcoxon Rank sum test found that bipolar patients had significantly higher levels of VEGF at baseline compared to healthy controls ($p = 0.01$). The logistic regression found that the AUC is 0.67 (95 CI: 0.54 – 0.80) and the VEGF cut point is 8.21 (Sensitivity: 0.86; Specificity: 0.50). Patients with VEGF levels above the cut point tended to be classified accurately as having bipolar disorder, while participants below the cut point only had a 50% chance of being well classified.

Patients' baseline VEGF scores were compared and found to be equivalent ($p = .99$). At all time points, patients receiving Celebrex (Mean = 16.10, Standard Error [SE] = 1.43) had comparable VEGF values to patients receiving placebo (Mean = 14.51, SE = 1.75, $p = 0.49$). The interaction between drug therapy and time was not significant ($p = 0.27$), indicating VEGF values did not change significantly over time based on drug therapy.

Baseline VEGF was a poor predictor of treatment response (OR = 0.97, 0.90-1.05, $p = 0.44$) with an AUC of 0.53 (95 CI: 0.33-0.73).

Conclusions: The success of the add-on celecoxib suggests that the disease was modulated through inflammatory mechanisms. However, the relationship between VEGF and the inflammatory modulation is unclear. Increased VEGF levels during the depressive phase of bipolar disorder agree with similar findings in major depression. A high VEGF level tended to accurately predict bipolar disorder, although not all patients had high VEGF levels, suggesting differential VEGF expression. One possible explanation is that VEGF levels could be increased through a neuroprotective mechanism in a disease state, allowing for neurogenesis to occur. Another possibility is that high VEGF levels could indicate a predisposition to mood disorders. Plasma measures of VEGF may have diagnostic utility and help guide personalized treatment. Treatment response was not predicted by baseline VEGF, which is different than what was found in a study on major depression in the same lab. VEGF levels did not change with the study treatment for either study group or the total cohort. Extending the length of the study may allow detection of changes in neurogenesis. Findings may also differ in the manic phase of bipolar disorder.

Keywords: Neurotrophins, Vascular Endothelial Growth Factor, Inflammation, Bipolar Disorder, Celecoxib

Disclosure: Nothing to Disclose.

T141. Activity-Dependent Response Reduction of GABA-A Receptors in Major Depressive Disorder

Agenor Limon*, Firoza Mamdani, Marquis Vawter, Francis Lee, Alan Schatzberg, Richard Myers, Jack Barchas, Huda Akil, Stanley Watson, William Bunney, Adolfo Sequeira

University of California, Irvine, Irvine, California, United States

Background: Major depressive disorder (MDD) is a leading cause of disability worldwide characterized by altered neuronal activity and abnormal excitability in brain regions involved in the control of stress and emotion. Transcriptional alterations of inhibitory GABA receptors have been found in postmortem brain samples of the dorsolateral prefrontal cortex (DLPFC) from individuals with MDD. The functional consequences of these alterations are unknown because of the lack of GABA specific ion flux measurements in human membranes. To address this problem, we optimized a novel approach to directly measure ion currents from synaptic GABA receptors isolated from postmortem brains of subjects with MDD to contrast with controls. We show in our preliminary work that GABA-A receptors become less efficient upon repetitive stimulation in the DLPFC of subjects with MDD compared to controls.

Methods: We microtransplanted human synaptic membranes containing native GABA receptors, from the postmortem DLPFC of 16 controls and 22 MDD subjects, into Xenopus oocytes. The oocyte membrane provides a functional platform for the direct electrophysiological characterization of native human synaptic receptors. Ion currents through GABA receptors were used to determine the maximum amplitude of the responses, their kinetic properties and the decay of the current amplitude upon repeated stimulation. Excitatory AMPA receptors present in the same preparations were also recorded with 100 μM kainate to calculate the electrophysiological excitation to inhibition ratio (E/I). All electrophysiological parameters were determined for each subject.

Results: Functional responses of native GABA receptors in controls and MDDs were similar in maximum amplitude,

activation time and desensitization ($P > 0.05$, two tailed t-tests). No effects of postmortem interval, RIN, gender or pH were observed for these parameters on functional responses of GABA receptors. There was a trend for an association between age and maximum GABA current ($r(38) = 0.32$; $P = 0.056$). Responses to kainate were similar between MDD cases and controls ($P > 0.05$), consequently, the E/I ratio was not different between groups. Interestingly, repetitive application of GABA elicited a differential run-down of GABA receptor currents between control and MDD cases. To facilitate the comparison with other in vitro studies, GABA current rundown was defined as the decrease of the peak current amplitude after six consecutive applications of GABA,10 s duration and 40 s interval between applications (Ragozzino et al., 2005; PNAS, 102: 15219). The response to the 6th application was reduced to 42 ± 7 % in control vs 34 ± 8 % in MDD (Mean ± SD, $P = 0.006$, two tailed t-tests) compared to the first application. Comparison between MDD who did not commit suicide ($n = 14$) and MDD cases who committed suicide ($n = 8$) shows similar rundown (35 ± 7 % in MDD vs 34 ± 10 in MDD-suicide).

Conclusions: Our results show that synaptic GABA receptors from the DLPFC of MDD cases have significantly smaller responses after repeated stimulation compared to controls, suggesting that in conditions of high synaptic demand (e.g. stress) GABAergic signaling responsivity might be compromised and the inhibitory/excitatory balance lost in MDD. Potential pro-excitatory alterations caused by enhanced rundown of GABA receptors may underline a state-dependent abnormal excitability in the DLPFC of individuals with MDD. These results if confirmed should point to novel pharmacological targets aiming to modulate GABA responsivity in MDD.

Keywords: Major Depressive Disorder, GABA-A receptors, inhibitory synaptic transmission

Disclosure: Nothing to Disclose.

T142. Multimodal Control of Dendritic Excitatory Synaptic Integration by Nucleus Accumbens Medium-Sized Spiny Neuron Collaterals

Hugo Tejeda*, Shiliang Zhang, Jocelyn Wu, Marisela Morales, Antonello Bonci

National Institute on Drug Abuse/National Institutes of Health, Baltimore, Maryland, United States

Background: Current views state that neurons integrate fast excitatory and inhibitory synaptic transmission to produce a well-defined output signal (patterned action potentials in the post-synaptic neuron), though these processes are, in fact, not well understood. The nucleus accumbens (NAcc) receives dense innervation from excitatory limbic afferents, with strongest inputs arising from the ventral hippocampal formation. Anatomical studies have suggested that medium-sized spiny neuron (MSN) collaterals (local synapses between pairs of MSNs) comprise an overwhelming fraction of inhibitory synapses in MSNs. However, electrophysiological studies have suggested that fast, inhibitory MSN collateral synapses between MSNs are sparse and weak, suggesting that MSNs may play a limited role in NAcc excitation-inhibition balance. In addition to synthesizing GABA, MSNs are highly enriched with opioid peptides. However, there is a critical knowledge gap in our understanding of how MSNs in the NAcc transform integrated excitatory, inhibitory, and opioidergic signals into coordinated activity within the circuit.

Methods: Here, we utilized a combination of whole-cell slice electrophysiology, anatomy, optogenetics, and genetic tools to determine how MSN collaterals in the NAcc influence synaptic integration.

Results: We find that dopamine D1 receptor-containing (D1 MSNs) and dopamine D2 receptor-containing (D2 MSNs) MSNs inhibit activity of neighboring MSNs utilizing GABAA receptor signaling. MSN collaterals target distal dendrites of other MSNs and utilize GABAA receptors to shunt incoming excitatory inputs. Moreover, MSN collateral inhibition is slow relative to fast-spiking interneuron inhibition, highlighting the diversity of inhibition within NAcc circuits. As a population, MSN collateral inhibition outweighs fast-spiking interneuron inhibition. Moreover, D1 MSN collaterals utilize dynorphin, an endogenous opioid receptor peptide, to decrease excitatory, but not inhibitory, inputs to MSNs via presynaptic kappa-opioid receptors. Finally, both D1 and D2 MSN collaterals are recruited by ventral hippocampal inputs to shape the flow of excitatory inputs and limit activation of NAcc MSNs.

Conclusions: This study highlights a previously unknown role of MSN collaterals in governing excitation-inhibition balance in NAcc circuits. We also provide a comprehensive model wherein recruitment of NAcc MSNs shapes synaptic integration to sculpt NAcc neuronal dynamics. These results are of importance as pertubations in NAcc MSN activity and dynorphin/kappa-opioid receptor dysfunction are major players in psychiatric disorders.

Keywords: Nucleus Accumbens, GABA, Dendrites, Excitation-Inhibition Balance, Dynorphin

Disclosure: Nothing to Disclose.

T143. Intrinsic Functional Organization and Network Roles in MDD and PTSD Change With Cognitive Behavioral Therapy (CBT)

Zhen Yang, Shi Gu, Nicolas Honnorat, Desmond Oathes, Steven Bruce, Russell Shinohara, Irem Aselcioglu, Phillip Cook, Kristin A. Linn, Theodore D. Satterthwaite, Danielle S. Bassett, Yvette Sheline*

University of Pennsylvania, Philadelphia, Pennsylvania, United States

Background: With increased conceptualization of neuropsychiatric disorders as involving large-scale functional network disorganization, researchers have become increasingly interested in applying graph theory-based tools to better understand the neural mechanisms of CBT. It has been proposed that correcting the imbalanced communication among functional networks plays an important role in the efficacy of CBT treatment responses. However, prior research has examined the neurobiological signature of CBT either at a coarse level (i.e., focusing on global topological features of the network) or has focused on a specific network. Thus, whether and how the functional interaction between specific networks within dynamic brain

systems contributes to CBT treatment responses remains unknown.

Methods: Our longitudinal sample included 64 participants (22 MDD, 42 PTSD) who entered CBT treatment and 32 healthy controls. Patients did not have additional comorbid psychiatric or medical diagnoses and were not on psychotropic or central nervous system-active drugs. For each subject, a high resolution T1 and two runs of resting-state functional MRI images (~7min/run) were collected on a Siemens 3T Trio scanner. Depressive and anxiety symptoms were assessed using the Montgomery-Asberg Depression Rating Scale (MADRS) and Anxious Arousal subscale of the self-report Mood and Anxiety Symptoms Questionnaire (MASQ-AA).

After standard preprocessing, intrinsic functional connections (network edges) between 220 functionally defined regions (network nodes) were estimated using a wavelet coherence in the range of 0.01-0.08 Hz. Each subject's node community membership was determined using a community detection algorithm based on modularity maximization. To examine the functional role of each network, we categorized each network into one of four categories based on its within- and between-network connectivity profile: connector (hub) versus provincial (non-hub) and cohesive versus incohesive. Univariate analyses examined how changes in depressive symptoms (measured using MADRS) were associated with changes in within- or between- network connectivity for each of the 10 networks. In multivariate analyses, the dependent variable was the change score of the MADRS. Predictors included change scores of within- and between-network connectivity of all 10 networks, and the change scores of the in-scanner head motion. We used the Elastic Net, a state of the art machine learning method, which selects a sparse set of predictors for a regression model. For inference on the Elastic Net results, we used the covariance test to determine which predictors were significant for predicting the normalized MADRS change scores at a significance level of $p < 0.001$ with Bonferroni correction for 21 predictors.

Results: Univariate analyses showed that greater improvement in depression was associated with a greater decrease in within-network connectivity (partial r = 0.49; confidence interval: 0.09 ~ 0.77) and a greater decrease in between-network connectivity (partial r = 0.71; confidence interval: 0.53 ~ 0.86) in the Ventral Attention (VA) network. With CBT treatment, there was an increase in the VA role as an incohesive provincial (decrease in within-network and decrease in between-network connectivity) and this change was associated with greater symptom amelioration. Multivariate predictors selected by the Elastic Net showed that when the tuning parameter was set to favor sparse models, the between-network connectivity of the VA network was the only predictor selected. As the Elastic Net was allowed to introduce more predictors (by adjusting the sparseness turning parameter), up to four networks were selected to predict MADRS change scores: within- and between-network connectivity of the VA network, within-network connectivity of the Salience network, and between-network connectivity of the Fronto-Parietal cognitive control network.

Conclusions: This study examined the effect of CBT upon network organization to determine how the functional interaction between specific networks contributes to treatment response. We determined a primary relation between decreases in between-network connectivity of the VA network that were associated with reduction of MADRS scores during CBT. In addition, using the Elastic Net, a regression approach that naturally performs model selection, we found that additional network variables, including within-network connectivity of the VA network, within-network connectivity of the Salience network and between-network connectivity of the Fronto-Parietal cognitive control network, improved prediction of decreases in MADRS score. These results underscore the importance of graph theory-based tools for understanding neural mechanisms of CBT and highlight the network basis of depressive symptom improvement following CBT.

Keywords: Cognitive Behavior Therapy, Graph Theory, MDD, PTSD, Network-Analysis, fMRI

Disclosure: Nothing to Disclose.

T144. Does Ketamine Anesthesia During Electroconvulsive Therapy Offer Efficacy Advantages Over Standard Methohexital Anesthesia? A Randomized Pilot Study

Styliani Kaliora, Raphael Braga, Ketan Hirapara, Samuel Bailine, Georgios Petrides*

Northwell Health, Glen Oaks, New York, United States

Background: Ketamine is an anesthetic medication which, when administered intravenously in sub-anesthetic doses (0.3-0.5mg/kg), was shown to have rapid antidepressant effects in patients with major depression. Because of its minimal antiepileptic properties and short half-life, ketamine has been used as an anesthetic agent during Electroconvulsive Therapy (ECT). It is used mostly when it is difficult to elicit an adequate seizure with the most commonly used anesthetic agent, methohexital. The antidepressant effects of the anesthetic doses of ketamine (1.0- 1.5mg/kg) have not been investigated in humans. We sought to examine whether the use of ketamine as an anesthetic during ECT for depression offers efficacy advantages over the commonly used methohexital anesthesia.

Methods: Patients undergoing ECT for an episode of unipolar or bipolar depression without psychotic features were randomly assigned to receive general anesthesia either with i.v. ketamine (1.0-1.5 mg/kg) or i.v. methohexital (0.5-1.0/kg). Inclusion criteria were diagnosis of major depressive episode, age 18-65, and a Hamilton rating scale for depression HRSD-24 > 21. Patients with current or past history of any psychotic disorder, substance abuse, neurological and cognitive disorders were excluded. DSM-IV diagnosis was established by a SCID interview. Raters were masked to anesthetic medication assignment. ECT was administered with bilateral placement with seizure threshold (ST) determination at the first session and at 1.5x ST at the following treatments. Primary outcome measure was number of treatments to respond, with response defined as HRSD < 10. Patients who received fewer than 10 ECT without meeting response criteria were considered non-completers. Ratings were performed by raters blinded to anesthetic medication.

Results: A total of 31 patients signed informed consent and were randomized to ketamine ($n = 16$) or methohexital ($n = 15$). The 2 treatment groups did not differ statistically in any demographic characteristics, with mean (SD) for age for ketamine: 48.01 (15.04) and methohexital: 48.34 (9.27). Twenty-one patients completed the study, 10 in the ketamine group and 11 in the methohexital group, while 6 and 4 dropped from each group respectively, without responding or completing 10 treatments. Both groups showed clinically and statistically significant decrease in mean HRSD, from 28.62 (1.85) to 9.49 (2.79) for the ketamine group and from 27.20 (1.90) to 8.93 (3.68) for the methohexital group. For completers, the mean number of treatments (SD) to reach response was not statistically different between the 2 groups: ketamine = 7.82 (3.43), methohexital = 8.60 (3.72), $p = 0.6235$. The trajectory of the response was not different ($p = 0.838$)

Conclusions: Ketamine anesthesia does not seem to increase the efficacy of ECT for depression as measured by the number of treatments needed to reach response compared to standard methohexital anesthesia.

Keywords: Ketamine, Electroconvulsive Therapy, Methohexital, Anesthesia

Disclosure: Nothing to Disclose.

T145. Electric Field Characteristics of Low-Field Synchronized Transcranial Magnetic Stimulation (sTMS)

Zhi-De Deng*, Sarah Lisanby

National Institute of Mental Health, Washington, District of Columbia, United States

Background: Conventional magnetic neurostimulation systems use a current-carrying coil to generate a time-varying magnetic field pulse, which in turn produces a spatially varying electric field—via electromagnetic induction—in the central or peripheral nervous system. An alternative approach to generating the time-varying magnetic field is by means of moving permanent magnets. Several systems have been proposed, involving rotation of high-strength neodymium magnets. One of these systems, termed synchronized transcranial magnetic stimulation (sTMS), was explored as a treatment of major depressive disorder (MDD). The sTMS device is comprised of a configuration of three cylindrical neodymium magnets mounted over the midline frontal polar region, the superior frontal gyrus, and the parietal cortex. The speed of rotation for the magnets was set to the patient's individualized peak alpha frequency of neural oscillations, as obtained by pretreatment electroencephalography recorded from prefrontal and occipital regions while the patient remained in eyes-closed, resting state. The hypothesized mechanism of action is that entrainment of alpha oscillations, via exogenous subthreshold sinusoidal stimulation produced by sTMS, could reset neural oscillators, enhance cortical plasticity, normalize cerebral blood flow, and altogether ameliorate depressive symptoms. In a multicenter, double-blind, sham-controlled trial of sTMS treatment of MDD, there was no difference in efficacy between active and sham in the intent-to-treat sample. In a subset of per-protocol patients, there was significant treatment effect at six-weeks. No direct electrophysiological evidence of the hypothesized mechanism of sTMS was reported, nor was the stimulation intensity and distribution well characterized. In this work, we evaluate the electric field characteristics of sTMS using the finite element method.

Methods: The finite element model was implemented in COMSOL Multiphysics (COMSOL, Burlington, MA) using its version of the IEEE Specific Anthropomorphic Mannequin (SAM) phantom. The head model (stator) has uniform, isotropic electrical conductivity of 0:33 S/m and relative permeability of 1. Three cylindrical magnets (rotators) are positioned along the midline: Magnet #1 is located over the frontal pole just above the eyebrows. Magnet #2 is 7.1 cm away from Magnet #1, approximately overlying the superior frontal gyrus. Magnet #3 is 9.2 cm away from Magnet #2, approximately overlying the parietal cortex. Each magnet is 2.54 cm in diameter and height, diametrically magnetized, with a residual flux density of 0.64 T. The axes of rotations are perpendicular to the sagital plane; and the rotation velocity is set to 10 Hz, corresponding to approximately peak alpha frequency. The resulting adaptive mesh consists of 56,825 tetrahedral elements. A stationary solution was first obtained using a direct solver (MUMPS), and then the time-dependent problem (in 10 degrees rotation steps) was solved. This assumes that the transient effects of initiating the rotating magnets have decayed, and the final solution reflects steady-state behavior.

Results: The electric field distribution of a single rotating magnet was first simulated in a sphere head model with radius of 8.5 cm. As the magnet rotates, the electric field switches from a figure-8 pattern (when the magnetic dipole is perpendicular to the head sphere at multiples of half-period) to a circular pattern (when the magnetic dipole is parallel to the head at multiples of quarter-period). The peak induced electric field strength at the surface of the head is approximately 0.05 V/m, in the direction parallel to the rotation axis of the magnet.

The electric field distribution of the full sTMS configuration in the SAM head model shows broadly distributed over midline frontal polar, medial frontal, and parietal regions. The peak induced electric field strength at the surface of the head is approximately 0.06 V/m. At a depth of 1.5 cm from the head surface, corresponding to the depth of the cortex, the electric field strength attenuates to approximately 0.02 V/m.

Conclusions: We evaluated the electric field characteristics of the sTMS system of rotating magnets using the finite element method. We found that the maximum induced electric field strength at the level of the cortex is approximately 0.02 V/m, which is an order of magnitude lower compared to those delivered by transcranial current stimulation and low field magnetic stimulation. Future work would include simulation of sTMS in anatomically-accurate head models derived from individual brain scans and treatment parameters. Direct electrophysiological data should also be collected to validate the proposed mechanism of action.

Keywords: Synchronized Transcranial Magnetic Stimulation (sTMS), Major Depressive Disorder (MDD), Electric Field Modeling

Disclosure: Nothing to Disclose.

T146. cAMP-Dependent and NMDA Receptor-Independent Action of Ketamine in a Cellular Model System: Ketamine Reveals a Biosignature Consistent With Other Antidepressants, but With a Rapid Time Course

Nathan Wray, Jeffrey Schappi, Harinder Singh, Mark Rasenick*

University of Illinois College of Medicine, Chicago, Illinois, United States

Background: Previous studies demonstrated that all extant classes of antidepressants increase coupling between the G protein, Gαs and adenylyl cyclase, resulting in persistent cAMP elevation. This is apparently due to Gαs being released from constraints of a lipid-raft environment to cholesterol-poor regions of the plasma membrane. Peripheral tissue and postmortem brain from depressed human subjects show a greater proportion of Gαs in lipid rafts. This is determined either with biochemical methods (raft isolation) or imaging using a fluorescent Gαs, the latter being suitable for high-content screening. Furthermore, most of the antidepressant compounds examined accumulate, slowly (3 days) in lipid rafts, a treatment period consistent with that required for Gαs translocation. Ketamine shows rapid and transient antidepressant action in humans and we wished to explore whether this was mimicked in a cellular model.
Methods: C6 glioma cells or primary astrocytes were treated with ketamine, harvested and lipid-raft Gαs was determined. C6 cells expressing GFP-Gαs were treated similarly with ketamine and the mobility of GFP-Gαs was determined by Fluorescence Recovery after Photobleaching (FRAP). FRAP was carried out either in C6 cells with stable expression of GFP-Gαs or in primary astrocytes or neural stem cells (derived from IPSC from human fibroblasts) by infecting cells with a mammalianized baculoviral vector coding for GFP- Gαs. BDNF, CREB and p-CREB were quantified after immunoblotting. cAMP was determined in cells infected with a fluorescent cAMP sensor delivered in a mammalianized baculoviral vector (Montana Molecular).
Results: Brief (15-minute) ketamine treatment (as low as 1μM) evoked the biochemical hallmark in C6 cells (translocation of Gαs from lipid rafts) seen after prolonged treatment with several species of antidepressant drugs. This effect was robust, but, similar to clinical parameters, was not long lasting, waning 24 hours after treatment (and washout). This is not mimicked by other NMDA antagonists, suggesting an additional site for ketamine action. Furthermore, knockdown of NR-1 eliminated NMDA receptor expression, but did not alter cellular response to ketamine as determined by FRAP or cAMP increase. Ketamine increased functional coupling of Gαs and adenylyl cyclase to increase intracellular cyclic adenosine monophosphate (cAMP). cAMP-dependent protein kinase phosphorylated cAMP response-element-binding-protein (CREB), which promoted BDNF synthesis. These somewhat delayed events were dependent on cAMP as they were blocked by the cAMP antagonist, Rp-cAMPS Furthermore, BDNF expression was increased in primary astrocytes after ketamine treatment. The ketamine metabolite, hydroxynorketamine, showed responses indistinguishable from ketamine in time or dose and did so also in cells where NMDA receptor was knocked down.
Conclusions: These results reveal a novel antidepressant mechanism in glia following a brief ketamine treatment that may contribute to ketamine's antidepressant effect. The cAMP dependence and NMDA-receptor independence suggests a possible alternative pathway for ketamine action. Furthermore, the translocation of GFP-Gαs produced by ketamine and all tested compounds with antidepressant activity might serve as a useful platform for identifying compounds with potential antidepressant activity and for predicting clinical response
Keywords: GPCR, Antidepressants, cAMP Signalling, BDNF, Lipid Rafts
Disclosure: Part 1: Pax Neuroscience, Stock / Equity, **Part 4:** Otsuka, Consultant.

T147. Individualized Modeling of Large-Scale Functional Network Dynamics With fMRI and rTMS

Shan Siddiqi*, Ludwig Trillo-Alvarez, Nicholas Trapp, Carl Hacker, Timothy Laumann, Alexandre Carter, David Brody

Harvard Medical School, McLean Hospital, Belmont, Massachusetts, United States

Background: While rTMS of the dorsolateral prefrontal cortex (DLPFC) has demonstrated efficacy for a wide variety of neuropsychiatric illnesses, its efficacy remains variable. This may be partly explained by inter-individual variability in functional topography of the DLPFC. The recent advent of individualized resting-state network (RSN) mapping enables identification of subject-specific network profiles for any DLPFC site. These methods have not yet been used to predict the expected brain network changes induced by rTMS.
Methods: Five healthy volunteers each underwent three baseline resting-state fMRI sessions. Individualized RSN mapping was used to construct detailed network maps for each subject. Single sessions of excitatory rTMS were applied to various DLPFC target sites in each individual. A total of 10 sessions were conducted in the first three subjects, including four at ventral attention network (VAN), four at default mode network (DMN), and two at frontoparietal control network (FPCN). Resting-state fMRI was collected immediately before and after each stimulation session and functional connectivity (FC) was calculated between the target site, VAN, DMN, FPCN, dorsal attention network, and language network. rTMS-induced change in FC was calculated between the target site and the RSN most correlated with that site. Each session was considered to be a "hit" if the target site's FC with the targeted network decreased more than its FC with any of the other five prefrontal networks. The primary outcome of the study was the overall "hit rate," or the overall percentage of sessions in which yielded "hits" across all subjects and targets. A planned interim analysis was conducted after the first 10 sessions, which included three of the five subjects included in the study.
Results: In six of the 10 (60%) rTMS sessions, FC between the stimulation site and the targeted network decreased more than any other network. Compared with an expected hit rate of 20% by random chance, single-proportion z-test showed $p < 0.001$ (one-tailed). Each RSN showed similar hit rates between individuals and within individuals.

Conclusions: Individualized RSN mapping can be used to predict the resting-state functional connectivity changes induced by single sessions of rTMS of the DLPFC. While this demonstrates that it is possible and feasible to target specific RSNs in specific individuals, we are collecting further data in order to refine the predictive model to achieve clinical significance. This enables the possibility of planning precision medicine interventions to selectively modulate brain networks in a disease-specific and patient-specific manner.
Keywords: Repetitive Transcranial Magnetic Stimulation (rTMS), Functional MRI (fMRI), Large Scale Networks, Precision Medicine for Mood Disorders
Disclosure: Part 1: SigNeuro, LLC, Stock / Equity.

T148. Stress-Induced Neuronal CSF1 Provokes Microglia-Mediated Neuronal Remodeling and Depressive-Like Behavior

Eric Wohleb*, Rosemarie Terwilliger, Catharine Duman, Ronald Duman

University of Cincinnati College of Medicine, Cincinnati, Ohio, United States

Background: Major depressive disorder (MDD) is a recurring neuropsychiatric illness that affects up to 17% of the population and causes significant social and economic burden. Clinical and basic research show that depressive symptoms stem from neuroplasticity deficits in the medial prefrontal cortex (PFC). Moreover, recent studies indicate that microglia, resident immune cells in the brain, play an important role in neuroplasticity. Neuron-derived molecular signals, such as transforming growth factor (TGF)-β, fractalkine (CX3CL1), and colony-stimulating factor (CSF)-1, direct microglia function and disruptions in these pathways may contribute to microglia activation and subsequent synaptic loss observed in models of depression. The primary objective of the current study is to determine the role of microglia in neuroplasticity deficits and development of depressive-like behavior in male and female mice.
Methods: In these studies male and female mice were exposed to 14 days of chronic unpredictable stress (CUS) and brains were collected for mRNA analyses. To further examine neuron-microglia interactions transgenic Thy-1 (GFP) mice, in which layer V pyramidal neurons express yellow fluorescent protein (GFP) were used and brains were collected for immunohistology to assess microglia activation and dendritic spine density on medial PFC pyramidal neurons. Engulfment of pyramidal neuron elements by microglia were assessed with Iba-1 immunohistology. Further studies examined anxiety- and depressive-like behavior in the open field, forced swim, sucrose consumption, and novelty-suppressed feeding tests. In the final experiments, viral-mediated knockdown of CSF1 was performed in the medial PFC of male mice.
Results: Initial mRNA analyses showed that 14 days of CUS increased levels of CSF1 in the medial PFC of male and female mice. In line with these findings postmortem dorsolateral PFC samples from depressed individuals also showed elevated CSF1 mRNA levels. Moreover, CUS increased CSF1 receptor expression in enriched microglia from the frontal cortex of male and female mice. Immunohistology showed that Iba-1+ microglia in male mice displayed an activated morphological phenotype in the medial PFC. In contrast, female mice showed no significant morphological changes in the medial PFC. Confocal imaging of Thy-1(GFP) mice revealed that CUS increased microglia phagocytosis of GFP+ pyramidal neuron dendritic elements in the medial PFC of male mice. Consistent with these findings, dendritic spine density was significantly reduced on pyramidal neurons in the medial PFC of male mice. In follow-up studies, viral-mediated CSF1 knockdown in the medial PFC of male mice attenuated CUS-induced microglia activation and prevented anxiety- and depressive-like behaviors.
Conclusions: These findings show that stress-induced alterations in neuron-derived signals provokes microglia-mediated neuronal remodeling in the medial PFC, which contributes to the development of anxiety- and depressive-like behaviors.
Keywords: Major Depression Disorder, Chronic Stress, Microglia, Colony Stimulating Factor-1
Disclosure: Nothing to Disclose.

T149. Relationships Between Inflammatory Markers and Suicide Risk Status in Major Depression

Licinia Gananca, Hanga Galfalvy, Sebastian Cisneros, Zahra Basseda, Thomas Cooper, Maria Oquendo, Michael Grunebaum, J. John Mann, M. Elizabeth Sublette*

Columbia University/New York State Psychiatric Institute, New York, New York, United States

Background: Pro-inflammatory status has been implicated in the depressed phase of mood disorders and in suicidal behaviors. Both polyunsaturated fatty acid status (PUFAs) and cytokine levels have been associated with suicide, independent of depression severity. How these inflammatory markers may relate with each other to influence suicide risk is less clear.
Methods: We measured arachidonic acid (AA%), docosahexaenoic acid (DHA%), and eicosapentaenoic acid (EPA%) as a percentage of total plasma phospholipids; and serum interleukin-6 (IL-6), interleukin-1β (IL-1β) and tumor necrosis factor α (TNF-α) in 80 patients with major depressive disorder in a current major depressive episode with a Hamilton depression score of at least 16, and 24 healthy volunteers. We performed separate ANOVAs comparing each individual PUFA and cytokine species in the following risk-stratified groups: suicide attempt within 5 years ($n = 20$); severe current suicidal ideators (having intent or plan) with no recent history of attempts ($n = 22$); low-risk current non-ideators who are also lifetime non-attempters ($n = 38$); and healthy controls ($n = 24$), and performed post hoc analyses to identify species associated with suicide risk. Sex and age were tested as covariates in the models. Results were not corrected for multiple comparisons.
Results: Of PUFAs and cytokines studied, only low DHA% ($p = 0.014$) and low IL-1β ($p = 0.011$) differed between groups. DHA% was lower in recent suicide attempters than in severe ideators ($p = 0.005$) and low-risk depressed patients ($p = 0.007$). IL-1β was lowest in attempters, differentiating

them from severe ideators ($p = 0.008$), low-risk depressed patients (at a trend level, $p = 0.059$) and controls ($p = 0.002$). No main effects of sex and age were seen so these covariates were removed from the models.
Conclusions: Our results suggest that suicide attempt status is a more useful marker than suicidal ideation for suicide risk related to neuroinflammation. We found that two indices of neuroinflammation differed among levels of suicide risk. Based on prior studies, the association of lower DHA with prior suicide attempts was expected, but the association of suicide attempt with lower IL-1β was surprising. However, interactions between DHA and IL-1β are complex: IL-1β is known to influence cyclooxygenase, involved in metabolism of DHA to docosanoids; and oxidized fatty acids can stimulate synthesis of IL-1β precursors. Future mechanistic studies would be useful to better understand the patterns of immune response associated with suicidal behaviors.
Keywords: Omega-3 Fatty Acids, Cytokines, Suicide
Disclosure: Nothing to Disclose.

T150. Examining Baseline and Longitudinal Changes in Brain Functional Activation as a Function of Recovery After a First Manic Episode - An Exploratory Study

Fabiano Nery*, Hongbo Wen, Maxwell Tallman, Caleb Adler, Melissa DelBello

University of Cincinnati, Cincinnati, Ohio, United States

Background: Neurophysiological mechanisms underlying the early course of bipolar disorder, particularly the recovery from a first manic episode are unknown. Mounting evidence suggest that the ventrolateral prefrontal cortex (VLPFC) and amygdala play an essential role in emotion and attention regulation in bipolar disorder. However, the longitudinal and dynamic changes in functional activation in these two brain regions during and following a manic episode are poorly understood. Here, we explored whether baseline brain activation during a parametric continuous performance task in a cohort of adolescents experiencing their first manic episode predicts recovery from the manic episode.
Methods: 3D T1-weighted magnetic resonance images were obtained from 26 adolescents (mean age ± SD: 15.2 ± 1.4 years old, range: 12 to 17 years old, 13 girls) hospitalized during their first manic or mixed episode of bipolar I disorder. Subjects were assessed longitudinally for up to 12 months after their first hospitalization, and received fMRI scanning at baseline, after reaching euthymia from their index episode, and at 12 months of follow-up. Longitudinal Interval Follow-up Examination (LIFE) was used to assess manic and depressive symptoms during follow-up. Symptomatic recovery was defined as 8 weeks with minor residual symptoms or no symptoms, assessed by the LIFE. We used fMRI to study brain regional activation while subjects performed a distorted continuous performance task with 3 levels of attentional demand. Region-of-interest analyses were performed to test the hypotheses. We used Cox regression survival analysis, controlling for age and sex, to explore the association between baseline activation in bilateral VLPFC and amygdala and symptomatic recovery during follow-up.
Results: During follow-up, 10 patients (38.5%) reached symptomatic recovery. Baseline activation in the right VLPFC predicted symptomatic recovery for two levels of attentional demand (Hazard Ratio (HR): 4.0, $p = 0.03$, for lowest level of attentional demand; HR: 2.1; $p = 0.07$, for mid-level of attentional demand; and HR: 5.6, $p = 0.009$, for the highest level of attentional demand). This association remained significant when controlling for baseline manic and depressive symptom scores, with no change in HR. There was no association between baseline activation in the left VLPFC and chance of symptomatic recovery during follow-up for any of the 3 levels of attentional demand. Baseline right amygdala activation was only associated with symptomatic recovery for the highest level of attentional demand (HR: 4.6, $p = 0.048$). When controlling for baseline manic and depressive rating scores, the risk association became a trend to significance, without change in effect size (HR: 4.1, $p = 0.06$). Baseline activation in left amygdala did not predict symptomatic recovery after a first manic episode. In the 12-month follow-up examination, right VLPFC activation decreased and right amygdala activation increased at the higher level of task level.
Conclusions: Our results suggest that during a first manic episode, greater baseline activation of the right VLPFC is associated with a greater probability of symptomatic recovery, despite the attentional demand level, potentially as a correlate of better prefrontal function. Right amygdala activation is only associated with a hazard of recovery at higher levels of task complexity, perhaps indicating a failed prefrontal modulation of amygdala as the attentional demands increase. These results suggest that those individuals with better prefrontal cortex modulation are more likely to experience recovery from their first manic episode. Longitudinal studies with larger samples are warranted to further investigate the role of prefrontal cortex-amygdala network in determining outcomes in bipolar disorder.
Keywords: Bipolar Disorder, Functional MRI (fMRI), Recovery, Ventrolateral Prefrontal Cortex, Amygdala
Disclosure: Part 1: Jansen, Honoraria, Eli Lilly, Employee, Spouse, **Part 2:** Eli Lilly, Employee, Spouse, **Part 3:** Eli Lilly, Employee, Spouse. **Part 4:** Jansen, Honoraria, **Part 5:** Eli Lilly, Employee, Spouse.

T151. Electrical Field Interaction With the Cortical Column Determines Responsiveness to Electroconvulsive Treatment

Miklos Argyelan*, Todd Lencz, Styliani Kaliora, Carlo Cottone, Marom Bikson, Georgios Petrides, Anil Malhotra

Hofstra Northwell School of Medicine, Glen Oaks, New York, United States

Background: Electroconvulsive therapy (ECT) is known to be the most effective treatment for treatment resistant depression (UK ECT Review Group 2003; Fink and Taylor 2007), yet its mechanism of action remains unknown (Grover et al. 2005; Kellner et al. 2012), and treatment response is variable. To date, we are unable to predict ECT treatment response or determine the most effective treatment option for an individual patient, which frequently leads to

significant delay in delivering effective treatment and can result in unnecessary exposure to unwanted side-effects.

One source why the effect of treatment may be variable across individuals is due to the patient's brain traits or neural status. In a recent study (Argyelan et al. 2016), we showed that baseline rs-fMRI signal amplitude in the subgenual cingulate cortex (SCC) correlated with ECT response in a cohort of 16 depressed subjects, explaining approximately 25% of the variance in the clinical data. Notably, the SCC has been implicated as a central node of the frontolimbic network of emotion regulation (Mayberg 1997; Lener and Iosifescu 2015) and has been targeted in clinical trials of deep brain stimulation (Mayberg et al. 2005; Choi et al. 2015).

Another equally important source of the intersubject variance of ECT efficacy can be due to the inevitable variations of the applied electrical field and its interactions with the patient's brain. This effect can be seen as analogous to "pharmacokinetics" for ECT: just as the liver may dictate how a pharmacologic intervention reaches its target, individual differences in skull and cortical anatomy may impact how electrical current reaches various parts of the brain. Thus, different individuals with identical baseline functional biomarkers (rs-fMRI and task-based fMRI) may have different responses to ECT because of structural anatomic differences.

Methods: We utilize structural scan data and electrical field modeling to calculate electrode-specific, individualized electrical field brain maps to predict ECT response. Twenty depressed patients (years; females) underwent MRI scanning less than 72 hours before starting electroconvulsive treatment (ECT). Inclusion criteria were: (1) between 18 and 70 years of age; (2) DSM-IV diagnosis of Major Depression, unipolar without psychotic features, or Bipolar I or Bipolar II Depression without psychotic features confirmed by SCID-IV interview; (3) pretreatment 24-item Hamilton Rating Scale for Depression (HAM-D) score $\geqq 21$.

During the study, the patient received bilateral ECT (Thymatron System) three times a week, and patients' symptoms were assessed with the HAM-D before every ECT. Remission was defined as two consecutive HAM-D ratings 10. The only psychotropic medication allowed during the study was lorazepam up to 3 mg per day for anxiety or insomnia.

Electrical Field (EF) maps were calculated in SimNIBS (Thielscher et al., 2015) environment. After segmentation of the structural MRI T1-weighted images, SimNIBS builds the three-dimensional tetrahedral mesh model of the head. The segmentation identifies five tissue types: white and gray matter of the brain, cerebrospinal fluid, skull, and scalp, and assigned them different conductivity values: 0.126 S/m, 0.275 S/m, 1.654 S/m, 0.01 S/m, and 0.465 S/m respectively. SimNIBS uses finite element methods (FEM) to solve the Laplace's equation, using Dirichlet boundary conditions at the electrodes (900 mA). The electrodes were placed based on the individual head model in the graphical user interface according to the standards used in our ECT center.

It is well established that EF along the axons induces more neural excitation than EF which is transverse to this direction (Peter T. Fox et al., 2004). Therefore, it is a reasonable assumption that the projection of the EF to the longitudinal direction of the cortical columns (normal to the pial surface identified in Freesurfer) is a better measure to predict electrical influence on the brain tissue. The EF projection (EFP) was calculated by performing the dot product between the EF and cortical column vectors in all voxels of the gray matter.

The magnitudes of the EF and EFP vector maps were calculated, and these scalar images were transformed to the MNI native space to perform voxel based correlations with clinical response and clinical responsiveness.

Results: The baseline HAM-D was 28.9 $\pm$ 5.6 and it decreased to 12.1 $\pm$ 8.4, indicating an average 57.5 $\pm$ 27.0 % change in the symptoms of depression. Patients received on average 6.8 ECT before getting into remission (range: 2–12). The clinical responsiveness (HAM-D score decrease per ECT treatment) was 10.7 $\pm$ 9.2.

Electrical field modeling of bifrontal electrode placements resulted in field maps with strong frontal influence bilaterally, the estimated fields in these areas were over 100 V/m. T Voxel-wise correlation of the EF maps with clinical response (ð«HAM-D) and clinical responsiveness (ð«HAM-D/time) failed to provide significant results in any brain areas.

As expected, electrical field projection (EFP) modeling resulted in more variable field maps due to the individual heterogeneity of cortical gyrification. Voxel wise correlation of EFP maps with clinical responsiveness (ð«HAM-D/time) yielded strongly significant results (FWE corrected, $p < 0.05$) in SCC, dorsal ACC, and right hippocampus. The results indicated that the higher the electrical field projections (the component of the electrical field which is parallel to the cortical column) were estimated in these regions the faster the response was possible.

Conclusions: The electrical field itself was not associated with clinical response in any brain regions, but its interaction with the underlying neural tissue was highly predictive for faster clinical response. In the future, individually optimized electrode placements (based on electrical modeling) can help to achieve faster and more reliable response in ECT. The generalization of our results on a 300+ cohort is now under way.

Keywords: Depression, Electroconvulsive Therapy, Subgenual Cingulate Cortex, Electrical Field Modeling, Individual Variability

Disclosure: Nothing to Disclose.

T152. Alteration of Monoaminergic Neuronal Firing by Acute Administration of Cariprazine: An in Vivo Electrophysiological Study

Pierre Blier*, Anna Herman, Nika Adham, Bela Kiss, Bence Farkas, Mostafa El Mansari

University of Ottawa Institute of Mental Health Research, Ottawa, Canada

Background: Dysregulated dopamine (D), serotonin (5-HT), and norepinephrine (NE) signaling has been linked to schizophrenia and depression. In vitro, cariprazine acts as a dopamine D3-preferring D3/D2 and 5-HT1A receptor partial agonist. It also acts as a 5-HT2A receptor antagonist. This in vitro receptor affinity profile endows cariprazine with

the potential to have clinical efficacy as a treatment for depressive episodes in addition to its established clinical efficacy in diminishing schizophrenia symptoms. Therapeutic implications for depression are supported by the antidepressant-like effects of cariprazine in rodent models of anhedonia and clinical studies that demonstrated its efficacy as monotherapy in bipolar depression and as an adjunctive therapy in major depressive disorder. This study used in vivo extracellular single cell recording to determine whether cariprazine treatment leads to functional alterations of monoamine systems in the intact rat brain.

Methods: In vivo electrophysiological recordings were carried out in male Sprague-Dawley rats under chloral hydrate anesthesia via electrodes lowered stereotaxically in the dorsal raphe nucleus (DRN; 5-HT neurons), locus coeruleus (LC; NE neurons), and hippocampus CA3 (excitatory pyramidal neurons). To measure the effect of cariprazine on neuronal activity in the DRN and LC, cumulative doses of cariprazine (50 μg/kg) and/or the selective 5-HT1A receptor antagonist WAY100635 (50 μg/kg) or the preferential 5-HT2A receptor agonist 2,5-dimethoxy-4-iodoamphetamine (DOI; 100 μg/kg) were administered systemically by intravenous injection. In the hippocampus, a small current (-1 to +1 nA) was applied to the quisqualate (1.5 mM) barrel of a multibarrel glass micropipette to activate CA3 pyramidal neurons to their physiological firing range (10-15 Hz). Cariprazine (10 mM) and 5-HT (15 mM) were then co-administered locally through iontophoresis. Drug-induced changes in neuronal firing were recorded and analyzed.

Results: In the DRN, cumulative doses of cariprazine induced a complete inhibition of the firing of 5-HT neurons, which was fully reversed by a single dose of WAY100635 ($n = 15$). In the LC, the inhibitory effect of DOI was reversed by cariprazine to 80% of baseline levels (ED50 = 65 μg/kg; $n = 7$) in a dose-dependent manner. Cariprazine alone had a negligible effect on the firing of LC NE neurons ($n = 6$). In the hippocampus, both cariprazine and 5-HT inhibited pyramidal neuronal activity following microiontophoretic application; this suppression was significantly attenuated by WAY100635 (cariprazine, $P = .007$; 5-HT, $P = .001$). When cariprazine and 5-HT were concomitantly administered by iontophoresis, cariprazine did not block the inhibitory effect of 5-HT on pyramidal neuronal firing ($n = 9$, $P > .05$).

Conclusions: These results suggest that after acute administration, cariprazine behaves in vivo as a 5-HT1A receptor agonist on 5-HT neurons in the DRN, a 5-HT2A receptor antagonist on neurons controlling the firing of LC neurons, and a full agonist at 5-HT1A receptors located on the pyramidal neurons of the hippocampus. The modulatory actions of cariprazine on 5-HT and NE transmission may contribute to the therapeutic efficacy of cariprazine, especially as it relates to depression or mood symptoms.

Keywords: Cariprazine, Monoamines, Electrophysiology

Disclosure: Part 1: Bristol Myers Squibb, Consultant, Janssen, Consultant, Sunovion, Consultant, Lundbeck, Consultant, Allergan, Consultant, Pfizer, Inc., Consultant, Otsuka, Consultant. **Part 2:** Forest, Consultant, Pfizer, Consultant, **Part 4:** Allergan, Grant, Janssen, Grant, Otsuka, Grant, Lundbeck, Grant.

T153. Influence of Solar Insolation in Springtime on the Age of Onset of Bipolar Disorder: A Multisite Study

Michael Bauer*, Tasha Glenn, Peter Whybrow

University Hospital Carl Gustav Carus, Technische Universität Dresden, Dresden, Germany

Background: There is considerable evidence of circadian rhythm dysfunction in patients with bipolar disorder including disturbances in the sleep/wake cycle, activity patterns, melatonin secretion, as well as suggestive associations with clock gene polymorphisms and epigenetic alterations. Clinical symptoms that are frequently reported include sleep timing disturbances, irregular daily schedules, and an evening preference. Even small changes to circadian rhythms such as the shift to daylight savings time may have adverse mental health consequences. As the onset of bipolar disorder is highly variable, it is important to understand the factors that may influence it, including environmental. We previously found a large, significant, inverse relation between the maximum monthly increase in solar insolation (incoming solar radiation striking the Earth's surface) and the age of onset of bipolar disorder. The aim of this multisite study was to confirm that the relations found previously were not sample specific, by repeating the analyses using significantly more data from geographically dispersed countries.

Methods: Solar insolation is defined as the amount of electromagnetic energy from the sun received on earth for a given surface area at a given time, expressed in kilowatt hours/square meter/day (kWh/m2/day) (42). All solar insolation data were obtained from the National Aeronautics and Space Administration (NASA) Surface Meteorological and Solar Energy (SSE) database version 6.0, which is based on global data collected by satellite for 22 years between 1983 and 2005. Clinical data were collected from 5536 patients with bipolar disorder at 50 sites in 32 countries on 6 continents. Onset occurred at 456 locations in 57 countries. Variables included solar insolation, birth cohort, family history, polarity of first episode and country physician density.

Results: 58.2% of the patients were female, the unadjusted mean age of onset was 25.4 ± 10.6 years. The largest maximum monthly increase in solar insolation occurred in the northern latitudes such as the Nordic countries, Russia, Estonia and Canada, and in warm dry areas in Chile, US, Mexico, Greece, and South Africa. The smallest changes occurred near the equator in Uganda, Colombia, Malaysia and Brazil. There was a significant, inverse association between the maximum monthly increase in solar insolation at the onset location, and the age of onset. This effect was reduced in those without a family history of mood disorders and with a first episode of mania rather than depression. The maximum monthly increase occurred in springtime. There was a large birth cohort effect, with the youngest group having the youngest onset. All prior relationships were confirmed using both the entire sample, and only the youngest birth cohort (all estimated coefficients $p < 0.001$).

Conclusions: Large increase in springtime solar insolation may impact the onset of bipolar disorder, especially with a family history of mood disorders. Major societal changes that may affect vulnerability to a circadian challenge need

investigation: exposure to LED lighting, mobile devices backlit with LEDs, and the 24-hour society. Limitations of this multicenter study were: the data collection process was not standardized; there was no individual data on behaviors or exposures that affect circadian rhythms; the sample was not demographically representative of the country populations. In the future, circadian symptoms and clock gene polymorphisms may help define endophenotypes of bipolar disorder, including early onset.

Keywords: Bipolar Disorder, Epidemiology, Circadian Rhythms

Disclosure: Part 1: Allergan, Advisory Board, Janssen, Advisory Board, Lundbeck, Advisory Board, Otsuka, Advisory Board, Neuraxpharm, Advisory Board, Servier, Advisory Board, Lilly, Honoraria, Pfizer, Honoraria, Servier, Honoraria.

T154. Diffusion MRI Tractography Fiber Cluster Topography Analysis of Frontostriatal Connectivity in Healthy Subjects

James Levitt*, Fan Zhang, Marek Kubicki, Martha Shenton, Lauren O'Donnell

VA Boston Healthcare System/Harvard Medical School, Brockton, Massachusetts, United States

Background: Key processes during brain development including neurogenesis, neuronal maturation and migration establish appropriate connections in the developing brain (Sekar, 2016; Stephan, 2012). Demonstrating brain miswiring thus implicates the presence of a developmental disorder. Prior research into white matter (WM) abnormalities in putative neurodevelopmental disorders, such as schizophrenia, have largely focused on using standard measures of WM integrity, such as fractional anisotropy (FA) and mean diffusivity (MD) via diffusion MRI (dMRI), (Fitzsimmons, 2013; Kubicki, 2007). Such an approach supports a deficit in structural connectivity but not in brain miswiring. Thus, a measure of brain wiring pattern, which we here propose, is an advance over standard dMRI measures. Specifically, we assess the pattern of long-range axonal anatomical connectivity between the prefrontal cortex (PFC) and striatum in 100 healthy subjects (n = 100 subjects from the Human Connectome Project (HCP); age: 22 to 35; sex: 46 females and 54 males). These tracts have been divided into 2 types of input: functionally segregated, anatomically parallel pathways, and functionally integrative, anatomically converging pathways, which is strongly supported by both animal tract tracing studies and human imaging studies (Draganski, 2008; Haber, 2003). Alternatively, this division has been characterized as reflecting a topographical (parallel pathways) vs. a non-topographical (converging) organization (e.g., Averbeck, 2014). It has been suggested that if striatal target regions receive mixed input from different cortical areas, such target regions might serve as integrative hubs for the processing of information (Averbeck, 2014, Haber, 2011). This has been quantified in animal tracer injection studies, where Averbeck (2014) demonstrated that overlap in the striatum decayed exponentially as distance increased between cortical injection sites. We propose a novel method using fiber clustering of dMRI tractography to assess the pattern of frontostriatal brain wiring. This technique allows us to assess 1) the degree of convergence of cortical projections to the striatum (which we here use as an estimate of overlap); and, 2) the extent to which this convergence is a function of the distance between prefrontal cortical tract origination locations. If this relationship is nonlinear, it supports the idea that the pattern of frontostriatal projections is not exclusively topographically organized but also allows for overlap and integration.

Methods: To enable the identification of fiber tract parcels from the prefrontal cortex (C) and the striatum (S), we first conduct a population-based (across the 100 subjects) tractography parcellation using a well-established data-driven fiber clustering pipeline (O'Donnell, 2007, 2012). The fiber clustering allows a fine whole brain tractography parcellation (2000 fiber clusters per subject) according to the WM anatomy (i.e. fiber geometric trajectory). Then, fiber clusters of interest (i.e. from C to S) from the whole brain WM are identified according to their connected anatomical brain regions. For the prefrontal cortex, we study multiple Freesurfer cortical regions [5] including rostral-middle-frontal, pars-opercularis, pars-triangularis, rostral-anterior-cingulate, caudal-anterior-cingulate, lateral-orbito-frontal, and medial-orbito-frontal gyri, while for the striatum, we study the caudate Freesurfer region. In this study, we identify 20 fiber clusters that connect C and S. For high sensitivity in identifying all potential fiber clusters of interest, a cluster is considered to connect C and S if it connects these regions in over 40% of subjects. To quantify the topographical relationship of these fiber clusters, we measure the mean distances between the endpoints of the fiber clusters within the prefrontal cortex (i.e. cortical distance) and the mean distances between the endpoints of the corresponding fiber clusters terminating in the striatum. The striatal degree of convergence, or overlap, is defined as one over the mean distance between the clusters' fiber endpoints connecting to the S region, such that low distances correspond to high overlap.

Results: We examined the relationship between the cortical distances and the corresponding striatal overlaps of the obtained fiber clusters that connect the prefrontal cortex (C) and the striatum (S) regions. We generated a plot (not shown) based on the 20 fiber clusters and thus a total of (20 * 19) / 2 = 190 pairs of fiber clusters, yielding 190 data points. A 2-term exponential model was fit to the data points. The amount of overlap decayed exponentially as a function of cortical distance, such that for the fiber cluster pairs with close cortical distances, their corresponding overlap was high and decreased rapidly as the cortical distances increased; however, for the cluster pairs with distant cortical distances, their corresponding overlap tended to be stable. This supports a frontostriatal projection pattern that is not strictly linearly topographically organized.

Conclusions: The distinction between the two types of tract input (topographic vs non-topographic) may have clinical importance as it has been suggested that information processing via topographically organized, segregated tracts allows for the refining of skills already learned, whereas non-topographically organized, integrative tracts may allow for integration of information and reward-based learning of new skills. Using fiber cluster topography analysis in healthy subjects using in vivo dMRI, initial results appear to support

the idea that the prefrontal cortical projection wiring pattern to the striatum is not strictly topographically organized. In future studies, this approach will allow us to assess other conditions such as schizophrenia to test for the degree of variation from a normal frontostriatal wiring pattern.
Keywords: Corticostriatal Connectivity, Diffusion MRI, Tractography, Brain Wiring
Disclosure: Nothing to Disclose.

T155. Maximizing Signal Detection of Functional Imaging With Machine Learning for Early Phase Development of Treatments for Neurodegenerative and Psychiatric Disorders

Mark Schmidt*, Dawn Matthews, Ana Lukic, Miles Wernick, Stephen Strother

Janssen Research & Development, LLC, Beerse, Belgium

Background: Functional imaging of neuronal activity can provide insight aiding in the staging, diagnosis, and phenotyping of neurodegenerative and psychiatric disorders, prediction of clinical change, and detection of treatment response. For example, neurodegenerative disorders such as Alzheimer's disease (AD), Frontotemporal Dementia (FTD), and progressive aphasias have distinctive, progressive spatial patterns of neuronal decline, and psychiatric disorders and treatments have also been shown to affect distinctive neuronal circuits. Measurement of glucose metabolism measurement with 2-fluoro-deoxyglucose (FDG) PET reflects these changes in synaptic activity and neuronal function, while functional MRI can provide closely coupled metrics of blood flow. However, image data is complex, and using standard univariate methods to evaluate regions of interest or images can be limited in producing disconnected regional changes that are difficult to relate to the network nature of brain activity and treatment response, reflect background noise as well as neuronal effects, and often lack statistical power. A key advance in image data utilization can be achieved through multivariate machine learning methods that make use of relationships between regional brain activity, segregate signal from background noise, and quantify the contribution of network patterns varying in dose response and temporal progression. This enables increased statistical power, more interpretable results, and a deeper understanding of the relative contributions of the neuronal networks to clinical status. Moreover, these methods can help to address hurdles in therapeutic development that include difficulty of diagnosis for patient selection, heterogeneity in clinical rates of progression, and subtlety of treatment response. We have applied multivariate machine learning to functional imaging data to demonstrate (a) the discrimination of different dementia phenotypes and underlying pathology, (b) the prediction of rate of clinical progression for patient stratification, and (c) the detection of treatment effect upon brain networks in relation to dose and duration, relevant to the development of effective treatments.
Methods: We applied the NPAIRS multivariate machine learning software framework (Strother et al, 2010) to the FDG PET and perfusion images of patient groups defined based upon clinical diagnosis, known pathology (amyloid status) (data from ADNI, UCSF Memory and Aging Center, NINDS, Technische Universität München), and for drug evaluation, and treatment arm (data on file with ADMdx). Images were quality controlled, processed to a common resolution and spatial template, and intensity normalized. Principal Component Analysis (PCA) was performed, followed by the mathematical combination of PCs into Canonical Variates (patterns, CVs), optimized based upon an intensive, iterative split half resampling process during which training and test data sets were compared to generate metrics of reproducibility and prediction. Each classifier produced one or more CVs where each patient's expression of a pattern was quantified as a numeric value that could then be evaluated using descriptive statistics and correlation to clinical endpoints. Classifiers were validated using Leave-One-Out testing and testing of independent subject populations where adequate subjects were available. To evaluate dementia discrimination, groups of patients with known clinical diagnosis and amyloid (A) status were compared including: normal controls (A-), AD (A+), Frontotemporal Dementia (behavioral, A-), Nonfluent Progressive Aphasia (A-), and Corticobasal Syndrome (CBS, A+ and A-). The relationship between baseline classification and subsequent rate of clinical progression was evaluated using Mild Cognitive Impairment (MCI, A+) patients. Subject FDG PET scans were scored using our previously developed AD Progression classifier and correlated with change in MMSE, CDR-sum of boxes and other clinical endpoints. To evaluate use in treatment effect detection, FDG PET and machine learning were applied in a set of 12 healthy volunteers who received the anxiolytic buspirone with imaging at baseline (placebo), after a first acute dose, second acute dose, first steady state condition, second steady state condition, and after partial washout, and in a set of 11 healthy volunteers who received placebo, an acute 100 mg dose, and an acute 300 mg dose of an NK1 antagonist compound in a double blinded randomized crossover design.
Results: Classifiers developed using different dementias showed that each dementia phenotype and underlying pathology was characterized by a pattern of functional effects and perfusion changes that could be discriminated from normal controls and other dementias. In A+ MCI patients, baseline AD Progression score correlated with subsequent rate of change in MMSE and CDR-sum of boxes. The analysis of buspirone identified a pattern of metabolic changes that emerged upon the first acute administration, increased with the second acute administration, plateaued at an elevated level during steady state, and decreased upon partial washout. The NK1 antagonist analysis revealed a dose-dependent pattern of metabolic effects that corresponded to clinical effects observed in a separate chronic study.
Conclusions: The combination of functional imaging with machine learning can enable a deeper understanding and identification of disease, individual response, and treatment effect, aiding in the development of treatments for neurodegenerative and psychiatric disorders. Analysis at voxel level, and utilizing information from disparate but functionally connected regions of the brain, may allow a more sensitive and informative method for detection of the effects of disease or pharmacological treatment.
Keywords: Positron Emission Tomography Imaging, Machine Learning, Neurodegenerative Disease

Disclosure: Part 1: Janssen Pharmaceutical, Employee, Novartis, Stock / Equity, Johnson & Johnson, Stock / Equity, **Part 2:** Janssen Pharmaceutical, Employee, Novartis, Stock / Equity, Johnson & Johnson, Stock / Equity, **Part 5:** Janssen Pharmaceutical, Employee.

T156. Expert Consensus Survey Results: Defining a Long-Acting Injectable Antipsychotic Therapeutic Trial and How to Determine Adherence and Response for Patients With Schizophrenia/Schizoaffective Disorder or Bipolar Disorder

Martha Sajatovic*, Matthew Byerly, Christoph Correll, John Kane, Faith DiBiasi, Heather Fitzgerald, Ruth Ross

Case Western Reserve University, Cleveland, Ohio, United States

Background: Although the potential of long-acting injectable antipsychotics (LAIs) in preventing relapse and improving medication adherence in patients with serious mental illness has been well-recognized, LAIs are under-utilized in clinical practice. Clear consensus on what constitutes an adequate LAI therapeutic trial and how LAIs should be used to determine treatment adherence and response has not been established. Expert consensus survey methodology could help inform clinical decision-making. This analysis evaluated expert opinion on defining adequacy of an LAI therapeutic trial and how LAIs should be used to determine adherence to and efficacy of pharmacologic therapy for patients with schizophrenia/schizoaffective disorder or bipolar disorder.
Methods: A 50-question survey was completed by 34 experts including researchers in mood and psychotic disorders with expertise in LAI trials, and prescribers with extensive clinical experience with multiple LAI antipsychotics. Respondents rated importance or appropriateness of predefined options on a 9-point Likert scale (1 = not important at all or not appropriate and 9 = extremely important or extremely appropriate); consensus was determined using Chi-square test ($p < 0.05$) of the distribution of scores across the 3 ranges (1–3, 4–6, 7–9). We calculated the mean and 95% confidence interval (CI) of the ratings for each item. A categorical rating of first, second, or third line was designated based on the lowest category in which the CI fell. Thus, items were considered first line if the bottom boundary of their CI was > 6.5. Items for which the bottom of the CI fell between 3.5 and 6.5 were considered second line, and those for which the bottom of the CI was < 3.5 were considered third line. It is important to note that the terms first line, second line, and third line are used differently in the context of the expert consensus method than they are sometimes used in clinical practice i.e. third-line options are NOT recommended (rated as inappropriate), not important, or refer to statements with which the panel disagrees.
Results: To define an adequate LAI trial, the experts considered both number of cycles (once steady state plasma levels are reached) and duration of LAI treatment important for schizophrenia and bipolar disorder. Although no first line consensus was achieved regarding the number of LAI cycles needed to establish a therapeutic trial, defining an adequate LAI trial as either the time it takes to achieve steady state or time to reach steady state plus 1-2 additional injection cycles both received high second line ratings (rated a 9 by 16% and 18%, respectively, and first line (>6.5) by 45% and 50%, respectively, for a patient with schizophrenia/schizoaffective disorder). Regarding adequate duration of treatment, high second line consensus (3.5 – 6.5) was achieved on treating patients with schizophrenia/schizoaffective disorder for >12 weeks after initiation of LAI treatment. For patients with bipolar disorder, no consensus was reached concerning duration of treatment in weeks but 66% ($N = 21/32$) of experts indicated that a patient's history of mood cycling intervals would influence their clinical decision concerning duration of an adequate trial. With respect to what constitutes treatment adherence and therapeutic efficacy, the majority of experts (21 %, $N = 7/33$ strongly agreed (rating of 9); 67%, $N = 22/33$ agreed (rating of 7-9)) believe that an adequate LAI trial is needed to determine this in patients with schizophrenia/schizoaffective disorder. Opinions were divided on whether 1 or 2 trials of different LAIs would be required to determine treatment resistance to oral antipsychotics; 50% of experts considered 1 trial and 44% considered 2 trials to be required before determining treatment resistance to oral antipsychotics for patients with schizophrenia. The level of agreement was lower for bipolar disorder in which most experts (9 %, $N = 3/33$ strongly agreed; 52%, $N = 17/33$ agreed) indicated that an LAI trial is needed to determine treatment resistance to oral antipsychotics in patients with bipolar disorder.
Conclusions: Experts felt that multiple cycles of LAIs are needed to ensure an adequate therapeutic trial, with the least conservative definition being the number of cycles needed to achieve steady state and a more conservative definition involving 1-2 cycles beyond steady state or at least 12 weeks of treatment. The majority of experts (21%, strongly agreed, 67%, agreed) believe that an adequate LAI trial is needed to determine what constitutes treatment adherence and therapeutic efficacy in patients with schizophrenia/schizoaffective disorder. Overall, consensus among experts was lower for bipolar disorder than for schizophrenia, underscoring the need for further studies on the use of LAIs in bipolar disorder. The lack of a first line consensus on the number of LAI cycles needed to establish a therapeutic trial highlights the need for further research and additional medical education regarding the use of LAIs for the treatment of schizophrenia/schizoaffective disorder and bipolar disorder.
Keywords: Long-Acting Injectable Antipsychotics, Treatment Adherence, Therapeutic Trial
Disclosure: Part 1: Neurocrine Biosciences, Inc., Consultant, Otsuka Pharmaceutical Development & Commercialization, Inc., Consultant, H. Lundbeck A/S, Consultant. **Part 2:** Otsuka Pharmaceutical Development & Commercialization, Inc., Consultant.

T157. Interoceptive Dysfunction Across Psychiatric Disorders: Preliminary Results From a Large-Scale Naturalistic Study

Sahib Khalsa*, Rayus Kuplicki, Hung-Wen Yeh, Katie Clary, Justin Feinstein, W. Kyle Simmons, Martin Paulus

Laureate Institute for Brain Research, Tulsa, Oklahoma, United States

Background: Interoception refers to how the brain receives, processes, and integrates internal signals from the body to influence motivated behavior. Interoceptive dysfunction has been proposed to impact the expression of disorders of mood and anxiety (Paulus and Stein, 2010) and substance use (Goldstein et al., 2009), but there is limited data on this topic. This study aimed to determine whether a modified heartbeat perception task can be used to quantify interoceptive dysfunctions in such individuals.

Methods: Participants were selected from the first 500 subjects of the Tulsa 1000, a naturalistic study that is assessing and longitudinally following 1000 individuals, including healthy comparisons and treatment-seeking individuals with mood and/or anxiety, substance use, and eating disorders. The Tulsa 1000 aims to determine how disorders of affect, substance use, and eating behavior organize across different units of analysis with a focus on predictors of long-term prognosis, symptom severity, and treatment outcome. Participants were screened into the study on the basis of a treatment-seeking history and a dimensional psychopathology score: Patient Health Questionnaire (PHQ-9) $\geq$ 10 and/or Overall Anxiety Severity and Impairment Scale (OASIS) $\geq$ 8, Drug Abuse Screening Test (DAST-10) score $>$ 2, or Eating Disorder Screen (SCOFF) score $\geq$ 2. Each participant underwent approximately 24 hours of testing over the course of one year including a standardized diagnostic assessment, self-report questionnaires, behavioral and physiological measurements indexing RDoC domains, magnetic resonance imaging focusing on brain structure and reward-related processing, fear processing, cognitive control/inhibition and interoceptive processing, and blood/microbiome collection.

This report focuses on a multi-unit analysis of cardiac interoception obtained during baseline assessment in the healthy comparison (HC; $n = 55$, 29 female), mood/anxiety (MA; $n = 247$, 176 female), and substance use (SU; $n = 138$, 71 female) subgroups. The remaining 60 out of 500 subjects either were in the eating disorders group (n = 27) or were excluded for poor performance on a control task (n = 33). Physiology, behavior, and self-report were recorded during several conditions of a novel heartbeat tapping task: pressing a button to indicate a felt heartbeat (guess), pressing only when they were sure (no-guess), pressing when sure during a prolonged inspiratory breath hold (breath hold), and pressing after hearing a tone (tone). The breath hold condition was included because it represents a non-invasive homeostatic perturbation with survival significance that was expected to modulate interoceptive signaling. The tone condition served as an exteroceptive control.

Five linear mixed effects models were run in R with y ~ Trial*Group + Psychotropic medication as a covariate, where y was the number of heartbeats, number of taps, subjective ratings of heartbeat intensity, and subjective ratings of task difficulty and performance confidence.

Results: There were significant main effects of trial for all tests (all F's $>$ 9, all p's $<$ 0.0001), but no effect of psychotropic medication status. The only main effect of group was on intensity (F = 11.6, p $<$ 0.0001), and there were trial by group interactions on all tests except confidence (all F's $>$ 2.73, p's $<$ 0.013).

Heartbeats: HCs showed elevated heart rates during breath holding compared to guessing (t = 3.3, $p = 0.0009$, increase of 2.6 beats). MA showed lower changes in heart rate between breath-hold vs. guessing than HCs (t = -2.8, $p = 0.005$, -2.4 beats).

Tapping behavior: The main effect of trial showed that varying the instructions did in fact alter performance on the task. Compared to HCs, SU tapped more times (indicating more felt heartbeats) only during the condition when they were instructed not to guess vs. when they were instructed to guess (t = 2.8, $p = 0.005$, 13.3 taps).

Subjective ratings: SU reported more intense sensations compared to HC across all interoceptive trials. However, trial by group interactions indicated SU perceived less intensely during tone vs. guessing conditions as compared to HC (t = -4.1, $p < 0.0001$, -19 intensity units), and had more difficulty detecting heartbeats during tone vs. guessing conditions as compared to HC (t = 3.3, $p = 0.001$, 15.9 difficulty units). While HC subjects were more confident in their performance on the tone (t = 10.8, $p < 0.0001$, 43 confidence units) and breath hold (t = 3.5, $p = 0.0004$, 14 units) tasks, there were no group differences.

Conclusions: Mood and anxiety disorders showed evidence of blunted physiological responses to endogenous interoceptive perturbation. In comparison, substance use disorders showed evidence of an amplified interoceptive and attenuated exteroceptive subjective experience. These initial findings, which await replication in the next 500 subjects, are consistent with the presence of dysfunctional interoceptive processing across psychiatric disorders. Future work will examine how such interoceptive dysregulation relates to neurobiological units of analysis, clinical outcomes, and illness progression.

Keywords: Anxiety Disorders, Major Depressive Disorder, Substance Use Disorder, Interoception, Research Domain Criteria (RDoC)

Disclosure: Nothing to Disclose.

T158. Pharmacological Inhibition of Hypocretin/orexin-1 Receptors Prevents Hyperarousal and Rewarding Behaviors Induced by Optogenetic Stimulation of Norepinephrine or Dopaminergic Neurons

Pascal Bonaventure*, Andrew Whittle, William Giardino, Brock Shireman, Timothy Lovenberg, Christine Dugovic, Luis de Lecea

Janssen Research & Development, LLC, San Diego, California, United States

Background: Orexin (aka hypocretin) neuropeptides are produced by a cluster of neurons within the lateral hypothalamus projecting widely through the brain. Orexins mediate their effects by stimulating two closely related G protein-coupled receptors, orexin-1 and orexin-2 receptors that are co-located or selectively located in specific brain areas suggesting differentiated roles. The orexin-2 receptor is almost the exclusive orexin receptor on the histaminergic neurons in the tuberomammilary nucleus which play a critical role in wake promotion, and on paraventricular neurons which are involved in the hypothalamic-pituitary-adrenal axis regulation. Orexin-1 receptors are selectively expressed on norepinephrine neurons in the locus coeruleus (LC) and on neuronal population shown to be important for

regulating motivating and rewarding behaviors. Here, we studied the ability of a selective orexin-1 receptor antagonist (compound-56, (N-({3-[(3-Ethoxy-6-methylpyridin-2-yl)carbonyl]-3-azabicyclo[4.1.0]hept-4-yl}methyl)-5-(trifluoromethyl)pyrimidin-2-amine) to attenuate behavioral responses that are dependent on functional orexin-1 receptor signaling. We tested whether systemic administration of compound-56 in mice was sufficient to modulate arousal resulting from optogenetic stimulation of the norepinephrine neurons (NE) of the LC neurons or behavioral reward elicited by real-time optogenetic stimulation of dopaminergic (DA) neurons of the ventral tegmental area (VTA).

Methods: LC-NE stimulation: Male Tyrosine Hydroxylase - Cre (TH:Cre) mice were injected bilaterally with AAV5-DIO-ChR2-eYFP in the LC and implanted with bilateral optical cannulae directly above the injection sites. Four weeks later, allowing for recovery and adequate viral expression, experiments commenced after mice were acclimated to being connected to fiber-optic cables. Mice were then injected subcutaneously with either vehicle or 10 mg/kg compound-56 or its inactive enantiomer three hours into the light phase and 30 minutes later monitored for 5 minutes in their home cage to provide baseline data. Mice were then stimulated with blue light. Changes to arousal were assessed visually and via locomotion. One week later the treatment groups were reversed and the study repeated.

VTA DA neuron stimulation: Male DAT:Cre mice were injected unilaterally with AAV5-DIO-ChR2-eYFP in the VTA and implanted with a unilateral optical cannula directly above the injection site. Four weeks later, allowing for recovery and adequate viral expression during habituation to a reverse light-cycle, experiments commenced after mice were acclimated to being connected to fiber-optic cables. In the middle of the dark/active phase, mice were moved to the experimental room. Mice were injected subcutaneously with either vehicle, Compound-56 (10 mg/kg), or its inactive enantiomer. Twenty minutes following injection, mice were connected to a blue light laser and placed into one of two identical sides of a rectangular black polystyrene box. Mice were stimulated continuously with blue light when they crossed into the side of the box opposite from which they were originally placed. Stimulation ceased immediately when the mouse transitioned back to the original side of the apparatus. Pairings of optogenetic stimulations with sides of the box and injection treatments were fully counterbalanced. Sessions were video recorded on each trial day and scored to determine the percentage of time spent on stimulation side for each trial. Frequency control trials used parameters of 1Hz (i.e. low-frequency stimulation) and 0Hz (i.e. no light [but still connected to laser via fiber optic patch cable]).

Results: LC-NE stimulation resulted in a significant increase from baseline in all measurements of locomotion, terminating in many animals in a behavioral arrest as NE levels in the LC were depleted. Treatment with compound-56 but not its inactive enantiomer inhibited this increase. All control animals eventually encountered the behavioral arrest, and treatment with compound-56 was able to prevent arrest from occurring during the timeframe of the experiment. VTA-DA stimulation produced real time place preference. Following injection of compound-56 but not its inactive enantiomer, real time place preference was completely blocked, as mice spent about half of their time on the stimulated-paired side.

Conclusions: Compound-56 was effective at inhibiting hyperarousal resulting from LC-NE stimulation using the fixed optogenetic parameters tested in this study. This is likely due to reduced orexin signaling in LC-NE neurons limiting the extent to which they can increase their firing rate in response to optogenetic stimulation. Compound-56 completely negated the place preference observed following stimulation of VTA-dopaminergic neurons. These neurons and neurons of the nucleus accumbens to which they project both express orexin-1 receptors, suggesting that orexin signaling is critical for eliciting the behavioral responses associated with normal function of the mesolimbic circuitry. The clear inhibitory effect of compound-56 on reward-seeking behavior suggests that that compound-56 is a potent regulator of VTA DA-dependent hedonic states relevant to psychiatric conditions that are driven in part by dysregulated reward processing.

Keywords: Orexin Receptor Antagonist, Locus Coeruleus, Ventral Tegmental Area (VTA), Optogenetics, Reward Circuitry

Disclosure: Part 5: Janssen, Employee.

T159. Kinect 4: A Phase 3, One-Year, Open-Label Trial of Valbenazine in Participants With Tardive Dyskinesia

Stephen Marder*, John Kane, Stewart Factor, Roland Jimenez, Dao Thai-Cuarto, Grace Liang

West Los Angeles VA Medical Center & Semel Institute at UCLA, Los Angeles, California, United States

Background: Tardive dyskinesia (TD) is a persistent movement disorder associated with prolonged exposure to antipsychotics and other dopamine receptor blocking agents. Historically, clinicians were limited to off-label treatments, but the evidence for these various therapies was limited at best. Valbenazine (INGREZZA) is a highly selective vesicular monoamine transporter 2 (VMAT2) inhibitor that is now approved for the treatment of tardive dyskinesia (TD) in adults. The efficacy and safety of valbenazine have been evaluated in several trials with 6-week, randomized, double-blind, placebo-controlled periods (KINECT, KINECT 2, KINECT 3) and two with longer-term extensions in which participants received valbenazine for up to 12 weeks (KINECT) or 48 weeks (KINECT 3). Findings of another long-term study are reported here.

Methods: KINECT 4 (NCT02405091) included a 48-week open-label treatment period and a 4-week washout/follow-up period. The study included adults (18-85 years) with moderate or severe TD (as qualitatively assessed by a blinded, external reviewer using a video of the participant's Abnormal Involuntary Movement Scale [AIMS] assessment at screening) and a diagnosis of schizophrenia/schizoaffective disorder, or mood disorder (e.g., bipolar disorder, major depressive disorder). Participants were required to be psychiatrically stable prior to study entry; stable regimens of concomitant medications to treat psychiatric disorders were allowed during the study. All participants received a starting dose of once-daily valbenazine 40 mg, which could be escalated to 80 mg at the end of Week 4 if both of the following criteria were met: Clinical Global Impression of Change-Tardive Dyskinesia (CGI-TD) score of ≥ 3

(minimally improved to very much worse) and acceptable safety/tolerability with the 40 mg dose, based on investigator judgment. From Weeks 4 to 48, a decrease to 40 mg was allowed if the participant was unable to tolerate the dose increase (80/40 mg group). Participants who were unable to tolerate the 40 mg dose were discontinued from the study. Efficacy and safety analyses were conducted in the safety population (all participants who received valbenazine and had any available postbaseline data). Efficacy assessments included the AIMS total score (sum of items 1-7), based on consensus scoring by 2 blinded central AIMS video raters (at baseline and Week 8 [first visit after dose escalation]) and by the investigator or site rater (at baseline, Week 8, and Week 48 [last on-treatment visit]). Additional efficacy assessments included: CGI-TD and Patient Global Impression of Change (PGIC) scores at Weeks 8 and 48; and CGI-TD and PGIC response, defined as a score of 1 or 2 (very much improved or much improved) at Week 48. Safety assessments included treatment-emergent adverse events (TEAEs). All outcomes were analyzed using descriptive statistics.

Results: Of the 167 participants who entered the study, 149 (89.2%) reached the Week 8 visit (40 mg, $n=33$; 80 mg, $n=105$; 80/40 mg, $n=11$) and 103 (61.7%) reached the Week 48 visit (i.e., treatment completers; 40 mg, $n=20$; 80 mg, $n=74$; 80/40 mg, $n=9$). Demographic and baseline characteristics in the overall safety population ($N=163$) were: mean age, 57.4 years; male, 52.8%; white, 67.5%; schizophrenia/schizoaffective disorder, 73.0%. Mean AIMS scores, as rated by central AIMS video raters, were generally similar across dose groups at baseline (40 mg, 10.2; 80 mg, 10.0; 80/40 mg, 9.3) and Week 8 (40 mg, 5.8; 80 mg, 6.5; 80/40 mg, 4.5), with AIMS mean score change from baseline (CFB) to Week 8 indicating TD improvement in all dose groups (40 mg, 4.5; 80 mg, -3.5; 80/40 mg, -4.9). Per site raters, AIMS mean scores were as follows: baseline (40 mg, 14.2; 80 mg, 15.0; 80/40 mg, 12.8), Week 8 (40 mg, 7.0; 80 mg, 9.6; 80/40 mg, 5.5), CFB to Week 8 (40 mg, -7.1; 80 mg, -5.4; 80/40 mg, -7.4). Per site raters, treatment completers experienced ongoing TD improvement with longer duration treatment, based on AIMS mean score CFB to Week 48 (40 mg, -10.2; 80 mg, -11.0; 80/40 mg, -7.2). Mean scores for CGI-TD (Week 8: 40 mg, 2.5; 80 mg, 2.6; 80/40 mg, 2.0; Week 48: 40 mg, 1.7; 80 mg, 1.6; 80/40 mg, 2.3) and PGIC (Week 8: 40 mg, 2.2; 80 mg, 2.5; 80/40 mg, 2.4; Week 48: 40 mg, 1.6; 80 mg, 1.7; 80/40 mg, 2.0) indicated clinically meaningful long-term improvements in all valbenazine dose groups. CGI-TD response was achieved by the majority of treatment completers (40 mg, 90.0%; 80 mg, 95.9%; 80/40 mg, 66.7%); the same was true for PGIC response at Week 48 (40 mg, 90.0%; 80 mg, 89.2%; 80/40 mg, 77.8%). Two-thirds (67.5%) of participants reported ≥ 1 TEAE at any time during valbenazine treatment, 14.7% discontinued due to ≥ 1 TEAE, and 12.9% reported ≥ 1 serious TEAE. One death occurred due to breast cancer (80 mg), judged by the investigator as not related to study drug. From Week 4 to Week 48, the only TEAEs that occurred in $\geq 5\%$ of all participants (combined dose groups) were urinary tract infection (8.5%) and headache (5.2%). CFB in vital signs, ECG parameters, and laboratory test values were generally small and not clinically significant.

Conclusions: Consistent with previous Phase 2/3 clinical trials, once-daily administration of valbenazine was associated with improvement of TD based on various efficacy measures including AIMS total score reduction and CGI-TD and PGIC mean scores. Based on the CGI-TD and PGIC, approximately 70-90% of treatment completers had a clinician- or self-rating of much improved or very much improved. Valbenazine was generally well tolerated and no safety signals were detected.

Keywords: Tardive Dyskinesia, Valbenazine, Clinical Trial

Disclosure: Part 1: Allergan, Advisory Board, Lundbeck, Consultant, Teva, Advisory Board, Takeda, Advisory Board, Otsuka, Advisory Board, **Part 4:** Forum, Grant, Neurocrine, Grant.

T160. Lithium Preserves Neurocognitive Functions by Proving a Decrease in Prevalence of Neurological Diseases & Myocardial Infarction: The Lithium Archive Project

Ronald Fieve*, Parthy Shah, James Prosser

Columbia University Medical Center, New York, New York, United States

Background: Lithium has been used as a mood stabilizer medication for bipolar disorder for more than 50 years. Laboratory and animal studies have demonstrated that lithium salts have cytoprotective actions, and may therefore ameliorate or prevent a variety of neurological and possibly other disease states. In a rat model of cerebral vascular accident, post-infarct lithium treatment has been demonstrated to reduce both stroke volume and functional deficits compared to control treatments. We are interested in examining if a disease-sparing effect of lithium may be observable in human patients taking regular oral lithium therapy. This study compares the prevalence of cardiovascular and neurological disease in psychiatric outpatients receiving and not receiving lithium.

Methods: This study consists of a retrospective chart review of adult psychiatric outpatients at the New York State Psychiatric Institute Lithium Clinic and two affiliate lithium clinics of the Columbia University Medical Center and the Foundation for Mood Disorders. Who were receiving and not receiving regular oral lithium treatment to compare the prevalence of severe neurological and cardiovascular disorders in them. These clinics specialize in the treatment of mood disorders and the majority of patients were diagnosed with either major depression or bipolar disorders. All patient diagnoses were made by board-certified psychiatrists, and patients were assigned to lithium treatment as appropriate for their individual diagnoses. All patients in the practice underwent yearly physical exams and blood chemistries performed by an independent medical practice. Data extracted from the medical records included patient demographic information, diagnosis, treatment information, and any reported medical or neurological disorders. One thousand one hundred and sixteen patients were studied. Of these 1,116, 626 (56%) received lithium treatment. Odds ratios were calculated to assess the risk of having a disorder for patients receiving lithium compared to patients not receiving lithium. A logistic regression model was used to examine the predictive role of lithium in the prevalence of neurological and cardiovascular disease among the study subjects.

Results: To date, 1,116 patients have been entered in the database (53.8% female, and 45.9% male), ranging in age from 18 to 88 years old (mean = 42.2 yrs; sd = 14.8 yrs). Of these, 626 patients (56%) received lithium treatment, with the average duration of lithium therapy being 2.75 years (range 0.1 – 30.0 yrs; sd = 4.82 yrs). The frequency of any neurological disease in this group was low: there were 178 unique disorders occurring in 117 patients (10.8% of all patients). The frequency of specific conditions ranged from 39 patients with a history of Migraine Headache, to zero patients with Huntington's Chorea, Down's Syndrome, Lewy Body Disease, and Multi-Infarct Dementia. For seizures, the OR was 0.096 (95% CI: 0.021 – 0.428). For dementia – not otherwise specified (D-NOS), the OR was 0.112 (95% CI: 0.017 – 0.921). For amyotrophic lateral sclerosis (ALS), the OR was 0.112 (95% CI: 0.015 – 0.920). For myocardial infarction (MI), the OR was 0.291 (95% CI: 0.112 – 0.753). Odds ratios for all other disorders (Alzheimer's Disease, Cerebrovascular Disease, CNS Neoplasm, Down's Syndrome, Huntington's Disease, Lewy-Body Disease, Migraine Headache, Multi-Infarct Dementia, Multiple Sclerosis, Optic Atrophy/Neuritis, Parkinson's Disease, Stroke, Syncope) were found to be equivalent to unity. For ALS, D-NOS, Seizures, and MI, lithium treatment was found to be negative predictor of disease occurrence when patient age, duration of clinic attendance, and use of any anti-psychotic medication were controlled.

Conclusions: Patients receiving long-term lithium treatment for psychiatric illness have a significantly lower prevalence of seizure disorders, ALS, Dementia (NOS), and MI, compared to psychiatric outpatients not receiving lithium therapy. These results suggest that long-term lithium treatment may protect against some neurological disorders and myocardial infarction in human patients taking lithium.

Keywords: Lithium, Mood Disorders, Dementia, ALS, Seizure

Disclosure: Nothing to Disclose.

T161. Risk of Toxoplasmosis in the Old Order Amish: Gender Differences, Heritability and Household Effects

Teodor Postolache*

University of Maryland School of Medicine, Baltimore, Maryland, United States

Background: Toxoplasma gondii (T. gondii) is a highly prevalent pathogen and a major foodborne contributor to mortality, morbidity and economic loss. Infection during pregnancy has major implications for the development of the new born in 20 to 60% of cases, with consequences varying from minor to severe (mental retardation, seizure disorder, visual impairment. T. Gondii infection has also been associated with schizophrenia, bipolar disorder, and cognitive deficits in the elderly and suicidal behavior. For prevention, it is important to identify modifiable risk factors ideally in a high prevalence environment allowing adjustment for multiple confounders, to individualize risk by gender and other subgroups, and to separate household effects from non-household effects (heritability, occupational). We now investigate gender differences in risk factors for Toxoplasma gondii infection, heritability and household effects in the Old Order Amish, a population with high seroprevalence rates.

Methods: 843 participants in the Amish Wellness Study (336 men and 507 women, mean age = 46.6 ± 16.8) had T. gondii IgG titers measured by ELISA and a risk factor exposure questionnaires obtained. Logistic regression was used to test the association between seropositivity and risk factors, adjusting for age and sex. Heritability of T. gondii, and of its risk factors, was estimated using log transformed T. gondii titers and mixed models that included fixed effects for age and sex and random kinship effect. Potential environmental contributions were estimated using a likelihood ratio test with household included as an additional random effect. Heritability and household effects were reciprocally adjusted.

Results: 478 participants (56.7%) were seropositive. Heritability for T. gondii IgG serointensity = 0.22 ($p = 0.05E-08$) and for T. gondii seropositivity was 0.28 ($p = 1.9E-05$). Seropositive status was associated with cleaning the litter-box (OR 2.30, 1.08-4.87), working with animals (OR 1.66, 1.18-2.33), eating rear/ raw meat (OR 1.61, 1.02-2.56 and OR 2.19, 1.17-4.10 respectively), or consuming unpasteurized milk/ yogurt (OR 1.74, 1.14-2.66). Major gender differences were present, with the food related factors (meat and milk) being significant in women, and the animal related factors (litterbox and contact with animals) – only in men. An exploration of heritability of the risk factors identified robust household effects for all factors (when adjusting for heritability), with working with animals, rare meat, and unpasteurized milk resisting Bonferroni adjustment for multiple comparisons (household effect 0.43-0.62, $p < 0.000002$ for each), but not changing the letterbox ($p = 0.02$). We also identified a significant heritability (when adjusted for household, only for eating rare meat. ($H2 = 0.37$, $p = 0.002$).

Conclusions: Prevention of congenital toxoplasmosis (infection in women during pregnancy) should focus on food, and not on cat litter. Preventative education methods should be gender-specific, and may have a major impact on brain and behavioral health. The heritability effect for rare meat consumption suggests the need for examining genetic associations and mediation. Household is where educational efforts geared towards reducing the rate of T. gondii infection and potentially rates of mental illness and suicidal behavior should have the highest chance to be most effective.

Keywords: Gender Differences, Neuroinfection, Neuroinflammation, Sex Differences

Disclosure: Nothing to Disclose.

T162. Psychotomimetic Effects of Single-Dose Ketamine in Children: Exploring Effects of Age and Alpha-2 Agonist Pretreatment

John Newcomer*, Robert Kennedy, Michael Yingling, Julia Schweiger, Nuri Farber, Ginger Nicol

Charles E. Schmidt College of Medicine, Florida Atlantic University, MIAMI, Florida, United States

Background: Sub-anesthetic doses of NMDA receptor antagonist drugs have been used for conscious sedation and treatment of pain and depression in humans, including

clinical use in children for burn management and fracture reductions where ketamine offers less respiratory depression than a commonly used combination of midazolam-fentanyl. However, NMDA antagonists like ketamine produce dose-dependent, transient psychotomimetic effects in adult humans with limited safety data in children. Animal models used to measure NMDA antagonist-induced neurotoxicity observed with sustained NMDA receptor blockade –along with limited clinical observations- suggest 1) that NMDA antagonist-induced neurotoxicity and psychotomimetic effects may both be age dependent (i.e., expressed post-puberty) and 2) that certain treatments –like alpha-2 adrenergic agonists- may attenuate NMDA antagonist-induced neurotoxicity, with initial experimental results in adult humans suggesting alpha-2 adrenergic agonist attenuation of ketamine-induced psychotomimetic effects. We report on a double-blind, placebo-controlled study of the effect of the alpha-2 agonist, dexmedetomidine, on the psychotomimetic effects of ketamine in children receiving single-dose, sub-anesthetic IV ketamine treatment for conscious sedation during limb fracture reductions in an emergency department (ED) setting. The study hypothesized 1) ketamine-induced psychotomimetic effects, and 2) attenuation of psychotomimetic effects with a) younger age and b) pretreatment using dexmedetomidine.

Methods: Children ages 7-17 years undergoing clinically indicated ketamine sedation for orthopedic fracture reduction in the ED were randomized (stratified to achieve approximately equal numbers of <12 vs. >12 years-of-age children in each group) to co-administration of dexmedetomidine versus placebo during sedation for ED management of fractures. Following initial pain control with oral oxycodone (0.2 mg/kg), medical assessments, patient stabilization, and informed consent, participants were administered dexmedetomidine (0.7 mcg/kg) or placebo, followed by 1.0 mg/kg of IV ketamine. Adverse events were recorded using a clinician-administered adverse events and side effects scale. The Brief Psychiatric Rating Scale for Children (BPRS-C) was administered prior to ketamine dose, during ketamine treatment, and again prior to discharge from the emergency room, with planned primary analysis focused on BPRS positive symptom change from the pre-ketamine to on-ketamine condition. Cognitive testing was obtained following on-ketamine BPRS ratings. Analyses on BPRS subscale and other outcomes were performed using repeated measures ANOVA (ANCOVA when baseline values were non-zero), using time as the repeated measure (pre- vs. during ketamine), and treatment condition (dexmedetomidine vs. saline) and age (<12 vs. >12) as independent variables.

Results: Forty children (mean age: 11.0 years (SD = 2.3 years), 70.0% male, 22.5% black) were randomized to ketamine+saline ($n=18$) vs. ketamine+dexmedetomidine ($n=22$). Ketamine treatment at these doses produced mild but statistically significant psychotomimetic effects, consistent with prior reports of dose-dependent psychotomimetic effects in adult humans. BPRS positive symptoms (hallucinations, delusions, peculiar fantasies), absent in all subjects prior to ketamine treatment, were manifested during ketamine treatment, as indicated by a significant main effect of time ($F[1,36]=19.98$, $p<0.0001$). A trend-level time by treatment by age interaction for BPRS positive symptoms was also observed ($F[1,36]=3.93$, $p=0.055$), explained by greater psychotomimetic effects in older children receiving ketamine without dexmedetomidine pre-treatment. Frequently reported (>5%, baseline-corrected) side effects were sedation (56.3%), dizziness (50.0%), "pleasant" dreams (31.4%) and visual hallucinations (29.7%).

Conclusions: The results of this study indicate that single-dose, sub-anesthetic IV ketamine may be feasibly used in a well-controlled ED setting for conscious sedation during medical procedures, with generally well-tolerated adverse events, consistent with prior reports. However, ketamine treatment in children, as in adults, is associated with psychotomimetic effects. Trend level observations in this sample are also consistent with prior work in animals and humans suggesting 1) that younger children may be less susceptible to psychotomimetic effects and 2) that pretreatment with alpha-2 agonists may attenuate NMDA antagonist induced effects on the brain. Future studies are needed to better characterize these effects.

Keywords: Ketamine, Pain, NMDA Antagonists, Pediatric

Disclosure: Part 1: Otsuka, Grant, Amgen, Consultant, Indivior, Consultant, Legal, Consultant, **Part 2:** Legal, Consultant, **Part 4:** Otsuka, Grant.

T163. Comparing Anatomic and Functional MRI Predictors of Cognition via Machine Learning

George James*, Bradford Martins, Ming-Hua Chung, Clinton Kilts

Psychiatric Research Institute/University of Arkansas for Medical Sciences, Little Rock, Arkansas, United States

Background: Functional MRI's translation into patient care is limited by our incomplete understanding of how the brain encodes normative variance in cognition. Both brain structure and function have been proposed as encoding cognition; however, the neuroimaging field lacks the methodology for unbiased direct comparison of these modalities. To address this limitation, we adapted the machine learning algorithm LASSO (Least Absolute Shrinkage and Selection Operator) to identify the combination of structural and functional MRI features that optimally predict diverse cognitions in a well-characterized normative sample. Consistent with past findings (James 2016, Siegel 2016), we hypothesized that fMRI would better predict higher-order cognitions whereas structural MRI (sMRI) would better predict domain-specific (or lower-order) cognitions.

Methods: Fifty-three healthy adults [mean(sd) age = 32(9.7) years, 31 (58%) females] were enrolled in the Cognitive Connectome project (Gess, 2014), an initiative pairing clinical neuropsychological testing with MRI neuroimaging evaluation. Participants underwent sMRI (MPRAGE sequence, 1x1x1 mm^3 resolution) and resting-state fMRI (rsfMRI, EPI sequence, (TR/TE/FA = 2000 ms/ 30 ms /90, 3x3x3 mm^3 resolution). Anatomic volumes (cortical and subcortical) were extracted from sMRI data via FreeSurfer v5.3.0 to serve as anatomic features. rsFMRI data underwent preprocessing in AFNI with extraction of 30 ICA networks via GIFTv4.0; pairwise correlation among these independent networks served as functional features (resting-state functional connectivity, or rsFC). Age and education were

regressed from all features to reduce multicollinearity, a confounding variable for using LASSO. Each feature underwent z-score normalization to allow direct comparison in LASSO. LASSO was implemented with 10-fold cross-validation in Matlab vR2017a to predict clinical neuropsychology performance from the combined sMRI features (160 volumes) and rsFC features (435 correlation pairs). LASSO identified the subset of features which optimally predicted performance for each neuropsychology test. These features were then ranked by unique percent variance explained (R-squared, R2).

Results: Consistent with hypotheses, higher order cognitions were predominantly explained by rsFC features. Two attentional measures were solely explained by rsFC: selective attention (D-KEFS Color-Word Stroop task) was explained by frontocingulate connectivity (R2 = 0.18), whereas attentional switching (Test of Everyday Attention test 4: visual elevator task) was explained by dorsomedial prefrontal and posterior cingulate connectivity (4 features, cumulative R2 = 0.51). A third attentional measure (D-KEFS Trail-Making IV, also attentional switching) was explained by sMRI (strongest predictor, precentral sulcus volume, R2 = 0.23) but also dorsomedial prefrontal and posterior connectivity rsFC (7 features, cumulative R2 = 0.51).The higher-order cognition of working memory was also largely explained by rsFC. Spatial working memory (Spatial Span Reverse) was solely explained by rsFC between frontocingulate and superior temporal networks (R2 = 0.23). Verbal working memory (Digit Span Backward) was explained by both sMRI (top 2 features, cumulative R2 = 0.45) and rsFC (4 features, cumulative R2 = 0.20). Digit Span Sequencing was equally explained by rsFC (R2 = 0.55) and sMRI (R2 = 0.39) features, but the large cumulative variance (R2 = 0.94) suggests overfitting, limiting interpretability.

Domain-specific cognitions had comparable contributions from structural and functional predictors – including language, whose cumulative R2 contributions from sMRI and rsFC features were respectively 0.22 and 0.44 for Boston Naming Test, 0.17 and 0.61 for COWAT Letter, and 0.55 and 0.38 variance for COWAT Animals. Visuospatial awareness (Judgment of Line Orientation Task) was predominantly explained by sMRI features (R2 = 0.48) with smaller rsFC contribution (R2 = 0.18). Motor behavior was an exception, which – contrary to hypotheses – was almost exclusively explained by rsFC features (right finger tapping speed: cumulative R2 = 0.80 explained by 8 rsFC features; left finger tapping speed: cumulative R2 = 0.68 explained by 6 rsFC and 1 sMRI features).

Conclusions: Results largely supported our a priori hypotheses, with the exception of rsFC features predicting motor performance. The large R2 values for some cognitions suggest overfitting, supporting the use of a larger dataset. Future work will extend this methodology to larger datasets (e.g. the Human Connectome Project) and incorporate additional neuroimaging modalities (diffusion weighted imaging and task-based fMRI).

Keywords: Cognition, Machine Learning, Resting-state fMRI, Cortical Thickness, Multimodal Neuoimaging

Disclosure: Nothing to Disclose.

T164. Morphological Brain Alterations and Changes in Hedonic Ingestive Behaviors Associated With Bariatric Surgery

Arpana Gupta*, Emeran Mayer, Elizabeth Gallagher, Ravi Bhatt, Tiffany Ju, Kristen Coveleskie, Jean Stains, Yijun Chen, Anna Balioukova, Jennifer Labus, Erik Dutson, Claudia Sanmiguel

University of California, Los Angeles, Los Angeles, California, United States

Background: In the United States, 65% of adults are considered either overweight or obese. Neuroimaging studies have identified obesity-related differences in morphological and functional scans, suggesting a possible role of the brain in the pathophysiology of obesity. Obese humans have significantly greater responses in gustatory, somatosensory, and reward-processing regions in response to palatable food and to visual food cues. It has been shown that bariatric surgery decreases the brain's reward network hyperactivity associated with obesity. The aim of the study is to investigate morphological changes in the brain's extended reward system one month after undergoing laparoscopic sleeve gastrectomy (LSG) and the associated changes in body mass, weight loss, appetite and hedonic ingestive behaviors.

Methods: Structural MRI was acquired in 16 female subjects at baseline (mean age = 39.5 ± 8.7yrs) and 1-month post-surgery. Voxel-based morphometry analyses using a general linear model controlling for age were conducted in FSL-VBM to determine differences in grey-matter pre-vs. post-surgery and paired t-tests were run to determine the changes in clinical and behavioral variables. All significance testing was conducted at $p < .05$, corrected for multiple comparisons using the Family-Wise Error method. Correlations between significant changes in brain morphometry and changes in obesity and behavioral variables were conducted ($p < .05$).

Results: Bariatric surgery resulted in significant reductions in BMI (45.0 ± 5.3 before vs. 40.3 ± 5.5 kg/m2 after surgery (t = 16.23, $p < .0001$) and in adiposity (total body fat mass: 119 kg vs. 106 kg, pre-vs. post-surgery (t = 16.82, $p < .0001$). LSG also resulted in decreased hedonic eating (Yale food addiction symptoms count of 3.62 pre-vs. 1.19 post-surgery (t = 4.80, $p < .0001$). Brain Morphological Changes: After surgery, significantly decreased grey matter (GM) densities were observed in the left anterior (t = 4.16,$p = .03$), middle, (t = 5.25, $p = .02$), posterior (t = 4.66, $p = .03$), and inferior insula (t = 3.88, $p = .04$); along with the left hippocampus (t = 5.14, $p = .02$). Correlations with Clinical variables: Post-operative reductions in total body fat mass were correlated to changes in middle insula GM (r = .53, $p = .04$). Changes in lean body mass were correlated with changes to the GM at the posterior insula (r = -.61, $p = .01$) and the inferior insula (r = -.55, $p = .03$). Changes in the Yale Food Addiction scores were correlated with changes in the middle insula GM (r = .59, $p = .02$).

Conclusions: Bariatric surgery results in a significant decrease in measures of obesity and hedonic eating as well as structural changes at the brain's reward network core regions. Postoperative changes in the brain's reward network were associated with reductions in hedonic eating and

adiposity, suggesting an effect of bariatric surgery on brain control of ingestive behaviors.

Keywords: Obesity, Hedonic Ingestion, Reward Network, Bariatric Surgery

Disclosure: Nothing to Disclose.

T165. Subtle Neurocognitive Deficits Associated With Obsessive-Compulsive Symptoms During Childhood: A Latent Class Analysis Applied in a Brazilian High Risk Cohort

Marcelo Batistuzzo*, Marcelo Hoexter, Eduardo Vattimo, Pedro Alvarenga, Roseli Shavitt, Lorenna Teixeira, Maria Conceição do Rosário, Euripedes Miguel, Giovanni Salum

Psychiatric Institute HC-FMUSP, Sao Paulo, Brazil

Background: More than a risk factor for obsessive-compulsive disorder (OCD), obsessive-compulsive symptoms (OCS) can be associated with certain brain circuits which, in turn, could appear as deficits in neurocognitive tasks. Although previous studies described neurocognitive deficits in children with OCD, little is known if those deficits are also presented in children with OCS in the general population. In this study, we use latent class analysis in a large school sample from Brazil to represent OCS and neurocognitive performance in three different domains: motor coordination, visuospatial/visuoconstructive ability, and executive functions. We hypothesized that OCS would be associated with all three cognitive domains.

Methods: The sample was constituted by 2,512 children (no siblings) residents of two Brazilian cities: Sao Paulo and Porto Alegre. All children were evaluated with the clinical instruments, whereas 2,401 subjects were assessed with any neuropsychological measure. This cohort has the peculiarity of having accessed a high risk for development of psychiatric disorders during childhood, according to an index of individual and familial screened psychopathology. More details about the constitution of the sample can be found in Salum et al. (2015).

Two instruments were used to extract the latent variable 'OCS' from subjects. The first one was the Brazilian Portuguese version of the Development and Well-Being Assessment (DAWBA) (Goodman, Ford et al. 2000), administered by lay interviewers. To compose the OCS latent variable we only used the OCD-related questions from DAWBA implicated in behavior (not impairment or time-consuming): 9 items (F2a-g, F3 and F4) that can assume three values each "no" = 0, "a little" = 1 and "a lot" = 2. The second instrument was the self-report measure Child Behavior Check List (CBCL) that, in our case, was fulfilled by the parent's child. OSC were quantified according to the 8 item scale proposed by Nelson and colleagues (nº 9, 31, 32, 52,66, 84, 85, 112) (Nelson et al., 2001). As in DAWBA, the values also can assume 0, 1 and 2, and the total scores vary from 0-16.

Different instruments were used in order to evaluate the three major neurocognitive domains: a) motor coordination (MO) were assessed by the Luria motor tasks (Luria, 1973); b) visuospatial/visuoconstructive abilities (VA) evaluated through Block Design (Wechsler Intelligence Scale for Children, 2002), Bender Gestalt task (Bender, 1938) and Rey Osterrieth Complex Figure test, copy part (Osterrieth, 1944; Rey, 1959); c) executive functions (EF), divided among three subdomains: a) working memory, measured by the Digit Span backward (Wechsler, 1991) and Corsi Block Tapping Task backward (Wechsler, 1997); b) inhibitory control, evaluated by means of the Go/NoGo task (Bitsakou et al., 2008) and the Conflict Control task (Hogan et al., 2005); c) temporal processing, assessed with the time anticipation task with short and long intervals (Toplak and Tannock, 2005).

We used confirmatory factor analysis (CFA) and to extract scores from the DAWBA and CBCL. In total, 17 items from both scales (only the OCS related items) were entered in a unidimensional model that was designated to evaluate the latent OCS trait. Then, the OCS latent value was extracted for each subject. We used the 'lavaan' library for R! (version 3.2.3 – www.r-project.org) and r-studio (https://www.rstudio.com) to extract CFA scores. We also used CFA to group the neurocognitive tasks in three high-ordering models. Finally, we regressed out age effects (in months) and use linear mixed-effects models (LMEM - using the 'nlme' library for R!) to test the influence of the OCS scores in neuropsychological measures, controlling for age, site (i.e. city: Sao Paulo or Porto Alegre), evaluator (each psychologist that performed the assessment) and general psychopathology factor ("p-factor").

Results: All CFA models showed good fit indexes. Results are presented next for: a) the unidimensional model that composed the OCS factor (CFI = 0.950, TCI = 0.940, RMSEA = 0.043) and three hi-order models for b) MC (CFI = 0.997, TCI = 0.994, RMSEA = 0.036), c) VA (CFI = 0.985, TCI = 0.984, RMSEA = 0.065) and d) EF (CFI = 1.000, TCI = 1.001, RMSEA < 0.001)

LMEM analyzes revealed that neurocognitive functions were associated with OCS: the higher the amount of symptoms, the lower MC ($\beta = -0.061$, $p = 0.023$) and VA ($\beta = -0.046$, $p = 0.004$) performance, but not with EF ($p = 0.325$). These results were controlled for age, site, IQ and evaluator. However, after adding the 'p-factor' in the LMEM results turn to be negative.

Conclusions: Our findings suggest that neurocognitive functions were affected by the presence of OCS in our sample of Brazilian scholars, especially regarding motor coordination problems and lower visuospatial/visuoconstructive abilities. On the other hand, contrary to our hypothesis EF, a hierarchically superior ability that is systematically reported in OCD patients. It is possible that this function is not impacted yet in a high-risk population with subclinical symptoms. Finally, although our results didn't survive when controlling for general psychopathology, the subtle interference of neurocognitive performance reported over OCS is important precisely because this was a dimensional evaluation in children that mostly present subclinical symptoms.

Keywords: Obsessive-Compulsive Symptoms, Neurocognitive Functioning, Childhood, Latent Class Analysis

Disclosure: Nothing to Disclose.

T166. Abuse Potential of NKTR-181 in Recreational Opioid Users: Results From a Randomized, Double-Blind Crossover Oral Study

Xue Snow Ge*, Lin Lu, Janet Jobes, Margaret Ziola, Sunny Xie, Ivan Gergel, Jack Henningfield, Michael Eldon

Nektar Therapeutics, San Francisco, California, United States

Background: NKTR-181 is a new molecular entity, full mu-opioid agonist currently in Phase 3 clinical trials as a potential treatment of moderate to severe chronic pain. The primary hypothesis underlying its development is that clinically meaningful opioid analgesia can be achieved in combination with decreases in acute brain-mediated side effects, such as euphoria, sedation and respiratory depression, by slowing the rate of drug entry into the brain. With NKTR-181, the slow rate of CNS entry is inherent to the drug itself and there currently exists no known chemical or physical method to alter the drug to increase its speed of entry from systemic blood to the brain, and hence its abuse potential and other brain mediated effects. In a Phase 3 clinical trial, NKTR-181 at 100 to 400 mg twice daily produced significant analgesia throughout 12 weeks of double-blind treatment of chronic low-back pain. For the same dosage range, an exploratory human abuse potential (HAP) study in recreational opioid users reported Drug Liking and Drug High scores for NKTR-181 dramatically lower than those for immediate-release oxycodone, and closely resembling those for placebo. Here we present results of a double-blind, double-dummy, randomized, crossover oral HAP study comparing substantially higher dose levels of NKTR-181 to oxycodone.

Methods: All subjects were healthy adults, 18 to 55 years of age, who had used opioids for non-therapeutic purposes ≥ 10 times in the past year, including ≥ 1 use in the past 8 weeks. Subjects were also required to not be physically opioid-dependent, as confirmed by a negative naloxone challenge during screening. They were also qualified for participation consistent with FDA's 2017 abuse potential guidance by pretesting with 40 mg oral oxycodone and placebo to assure that oxycodone was discriminated and tolerated with elevated drug-liking scores.

Subjects remaining eligible entered an in-patient, double-blind treatment phase where they randomly received single oral doses of each of six treatments, according to a crossover design. Treatments were NKTR-181 at 400 mg, 600 mg, and a supratherapeutic 1200 mg, oxycodone at 40 mg and 60 mg and placebo, each separated by washouts of ≥ 5-day. NKTR-181 doses and its matching placebo were administered as oral tablets. Oxycodone HCl doses were administered as over-encapsulated oral tablets with matching placebo capsules. Subjects provided ratings of liking and other effects at scheduled times for 24 hours post dose on subjective measures consistent with the FDA abuse potential guidance. Other measures included pupil diameter and opioid- and abuse-related AEs, as well as blood samples for PK evaluation.

Results: Among 54 recreational opioid users who completed all 6 treatments, onset of Drug Liking was significantly more rapid for both oxycodone dose levels than for any of the NKTR-181 dose levels, as measured by the LS mean rate of increase of the Drug Liking curve. For the first hour, the rates were 1.6, 4.5, and 6.1 mm/hr for NKTR-181 at 400, 600, and 1200 mg, respectively, compared with 14.2 and 20.5 mm/hr for oxycodone at 40 and 60 mg ($P<0.0001$ for all comparisons). During the first hour, the area under the effect (AUE) values were 0.8, 1.7 and 2.4 for NKTR-181 at 400, 600 and 1200 mg, respectively, compared with 5.8 and 9.5 for oxycodone at 40 and 60 mg ($P<0.0001$ for all comparisons, except $P=0.0002$ for 1200 mg NKTR-181 vs. oxycodone 60 mg).

The least-squares (LS) mean Emax for Drug Liking "at this moment" (primary outcome measure) was 62.0 and 67.9 for NKTR-181 at 400 and 600 mg, respectively, compared with 76.6 and 81.5 for oxycodone 40 and 60 mg, respectively, and with 53.2 for placebo ($P<0.0001$ for all comparisons between NKTR-181 and oxycodone). For the supratherapeutic NKTR-181 dose, Drug Liking Emax was 76.7 ($P=0.0071$ vs oxycodone 60 mg).

Peak ratings for Drug High and Take Drug Again were also significantly lower for all NKTR-181 dose levels compared with 60 mg oxycodone. Emax values for Drug Liking and Drug High after NKTR-181 400 mg in this study matched the values for NKTR-181 400 mg in the prior HAP study, at 62.0 vs 62.3 and 21.3 vs 22.6, respectively. Similar consistencies were observed among studies for all other subjective abuse potential measurements.

Conclusions: In recreational opioid users, NKTR-181 exhibited significantly less abuse potential compared with oxycodone, a conventional Schedule II opioid. For NKTR-181 at therapeutic dose levels of 400 and 600 mg, Drug Liking was significantly lower than oxycodone at dose levels of 40 and 60 mg. At all dose levels of NKTR-181, likeability, pupil diameter and several other measures during the critical early post-dose time period were significantly lower in magnitude and slower in onset than for oxycodone, and had a time profile consistent with a slowed rate of brain entry. For NKTR-181 at the supratherapeutic dose level of 1200 mg, Drug Liking, Drug High, and Take Drug Again scores were significantly less than those from a therapeutic 60 mg dose of oxycodone.

Overall, these findings are consistent with data from other clinical trials, including Phase 3 data, which characterize NKTR-181 with a significantly lower overall abuse potential as compared with prototypic Schedule II opioids.

Keywords: Abuse Potential, Mu-Opioid Receptor Agonist, Drug Liking, Chronic Pain Treatment, Controlled Substance Scheduling

Disclosure: Part 1: Nektar Therapeutics, Employee, **Part 2:** Nektar Therapeutics, Employee, Nektar Therapeutics, Stock / Equity, **Part 5:** Nektar Therapeutics, Employee.

T167. Elevated Soluble Epoxide Hydrolase Activity is Associated With Worsening Anxiety and Cognition: a Pilot Longitudinal Study in Breast Cancer Patients Receiving Adjuvant Therapy

Pei-an Betty Shih, Jun Yang, Marbella Cervantes, Frank Penedo, Christophe Morisseau, Lei Wang*

Northwestern University Feinberg School of Medicine, Chicago, Illinois, United States

Background: Cancer patients receiving chemotherapy often report increased difficulties in their ability to remember and concentrate, and worsening of depressive and anxiety symptoms. Inflammation is associated with neurodegeneration and impaired neurogenesis, hence dysregulation in the inflammation responses may lead to cognitive decline and related traits. This longitudinal study examined relationships of the ex vivo activity of soluble epoxide hydrolase (sEH), a key enzyme with inflammation resolution properties, with measures of cognition, depression, and anxiety in female breast cancer patients receiving chemotherapy and healthy control women. We first assessed whether there are differences in the longitudinal changes in cognition, depression, and anxiety between groups, followed by elucidating associations of sEH activity with longitudinal changes of cognitive function, anxiety, and depression to identify inflammation-mediated effects on these traits.
Methods: Seven female breast cancer patients (mean age [SD]: 54.21 ± 10.17) and seven age-, sex-education-matched healthy controls (mean age [SD]: 53.8 ± 6.4) were included in the study. Psychopathology traits and ex vivo sEH activity were measured at both baseline and the follow-up visits (post-chemo treatment visit for breast cancer patients and follow-up visit for controls, mean follow-up days: 202 vs. 219, NS) in all subjects. Cognitive function was measured by Verbal Fluency, Ray's Auditory Verbal Learning Test (RAVLT), WMS-III Letter Number Sequencing (LNS), WMS-III Spatial Span (SS), Trail Making A & B, and WAIS-IV Digit Symbol Coding (DSC). Depression was measured by Inventory for Depressive Symptomatology (IDS) and anxiety was measured by Hamilton Anxiety Scale (HAMA). Within-individual longitudinal change of clinical traits and sEH activity was assessed by the difference of the two timepoints. Nonparametric test statistics (Wilcox test and Spearman's rank correlation coefficient) were used to assess strength of association. All analyses were done in R.
Results: At the baseline visit, sEH activity trended lower in cancer patients compared to controls (11.14 vs 15.53, $p = 0.09$). Combining all available data of the two visits, the strength of association increased (11.46 vs. 14.61, $p = 0.05$). Longitudinal change of sEH activity appeared to be higher in cancer patients compared to controls (+0.98 vs -3.3, NS). Longitudinal changes of cognition measures were not significantly different between the two groups. Change of depression score (IDS) was significantly higher in patients (+3.43 vs -0.33, $p = 0.04$) as well as the anxiety score (+2.29 vs. -0.57, $p = 0.03$). Higher longitudinal sEH activity was significantly associated with increased longitudinal anxiety in controls (Rho = 0.8944, $p = 0.04$). Change in longitudinal sEH activity was inversely associated with SS Forward score (Rho = -0.703, $p = 0.015$) and positively associated with Trailmaking B score (Rho = 0.6027, $p = 0.04$) in all subjects.
Conclusions: Breast cancer patients demonstrated a number of adverse longitudinal traits in depression and anxiety compared to control women. While this is the first study showing a decreased sEH activity in breast cancer patients compared to healthy controls, the significant association between increased longitudinal change of sEH activity and poorer outcome in anxiety and cognition suggests that dysregulation of inflammation, marked by the aberrant sEH activity, may contribute to outcomes in cognitive function, depression, and anxiety. Replication with a larger sample is necessary.

Keywords: Inflammation, Breast Cancer Chemotherapy, Cognition, Depression, Anxiety
Disclosure: Nothing to Disclose.

T168. One Size Does Not Fit All: Challenges in Advance Care Planning and End-Of-Life Treatment Preferences by Latino and Non-Latino White Cancer Patients

Veronica Cardenas*, Sarah Friedman, Jesse Nodora, Sandahl Nelson, Paulette Gabbai-Saldate, Katrina Ng, Yael Cohen-Arazi, Joseph Ma, Elena Martinez, Eric Roeland

University of California, San Diego, La Jolla, California, United States

Background: Advance care planning (ACP) is a process whereby patients communicate their end-of-life treatment preferences and select a surrogate to make decisions on their behalf if they become incapacitated. Despite a growing US Latino population, much remains unknown regarding ACP for Latinos. The purpose of this study was to examine the association between ethnicity and ACP, including individual surrogate markers such as code status documentation, advance directive (AD)/Physician Order for Life-Sustaining Treatment (POLST) completion, and/or palliative care consultation in deceased Latino vs. Non-Latino White cancer patients. Additionally, this study examined the selection of end-of-life treatment preferences and relationship of the selected surrogate decision maker documented by each racial/ethnic group.
Methods: In order to examine the relationship between race/ethnicity and ACP, data from an NCI-designated cancer center were obtained to conduct a retrospective analysis of randomly selected matched pairs of deceased Latino and Non-Latino White cancer patients (2011-2016). The pairs were matched based on sex, age (at diagnosis/death), and cancer type. Conditional logistic regression was used to assess ethnicity (Latino vs. Non-Latino White) and the presence of ACP (yes/no). Secondary aims examined the association between race/ethnicity and the presence of individual ACP surrogate markers using separate logistic regression models. An exploratory aim used descriptive statistics to examine patient selections to accept or decline potentially life-prolonging treatment and to evaluate the relationships with their selected surrogates. All analyses were completed using SAS 9.4.
Results: Of the 152 matched pairs, code status was documented for 125 (82%) of Latinos and 117 (77%) of Non-Latino White patients. A palliative care consultation was recorded for 60 (39%) Latinos and 50 (33%) Non-Latino White patients. Completion of an advance directive or POLST was documented for 25 (16%) Latino and 47 (31%) Non-Latino White patients. Spanish language was preferred by 17 (68%) of the 25 Latino patients who completed AD/POLST documents. There were no significant differences in the presence of any ACP, code status documentation, or palliative care consultation among the 152 eligible matched pairs. However, cancer patients with AD/POLST completion were 58% less likely to be Latino than Non-Latino White. End-of-life treatment preference on AD/POLST documents revealed that 3 (12%) Latino patients would prefer life-

prolonging treatment compared to 7 (14.9%) of Non-Latino-White patients. Moreover, 11 (44%) of Latino patients would prefer no life-prolonging treatment compared to 28 (59.6%) of Non-Latino White patients. Last, 11 (44%) of Latino patients did not specify an end-of-life treatment preference compared to 12 (25.6%) of Non-Latino White patients. Among Latino patients, the selected surrogate decision maker's relationship to patient included 13 (52%) adult child, 3 (12%) spouse/partner, and 9 (36%) not specified or unknown. For Non-Latino White patients, the selected surrogate decision maker was 19 (40.5%) spouse/partner, 8 (17%) adult child, 5 (10.6%) parent, 2 (4.3%) sibling, 1 (2.1 %) nephew/niece, 3 (6.3%) friend, and 9 (19.1%) not specified or unknown.

Conclusions: Advance care planning in cancer care has historically proven difficult to implement with only 20-30% completion across all racial/ethnic groups. We found lower rates of AD/POLST completion in Latino patients, which is consistent with prior studies. These findings strengthen the evidence for continued improvement of AD/POLST completion among all racial/ethnic groups, but perhaps underscore a particular challenge amongst Latinos. Additionally, this study begins to explore patterns in specific details that may enhance the experience of ACP among Latinos such as the importance of family involvement in decision-making. A limiting factor of this study is the small sample size limited to a single academic medical center, which impedes the generalizability of the results and requires further evaluation.

Keywords: Advance Care Planning, Disparities, Hispanic/Latinos, Disparities, Cancer Quality of Care

Disclosure: Nothing to Disclose.

T169. Biomarkers and New Therapeutics in TBI and PTSD: Neurosteroid Signatures to Phase II Randomized Controlled Trials

Chris Marx*, Jennifer Naylor, Jason Kilts, Steven Szabo, Ryan Wagner, Gillian Parke, Rajendra Morey, Gerry Grant, Michael Hauser, Murray Stein, Lawrence Shampine

Duke School of Medicine and Durham VA, Durham, North Carolina, United States

Background: Neurosteroids are enriched in brain and exhibit pleiotropic actions in the central nervous system that are highly relevant to the pathophysiology of traumatic brain injury (TBI) and post-traumatic stress disorder (PTSD). Allopregnanolone (ALLO) is a positive GABAA receptor modulator and downstream metabolite of pregnenolone (PREG); it has neuroprotective, neurotrophic, anti-inflammatory, analgesic, and neurogenesis-enhancing properties in rodent models. ALLO also increases myelination. We thus quantified neurosteroid and inflammatory biomarker candidates in participants with TBI and/or PTSD enrolled in the INjury and TRaumatic STress (INTRuST) Biorepository Cohort. We also conducted a randomized controlled trial (RCT) examining the potential utility of neurosteroids as therapeutic interventions utilizing PREG as a precursor loading strategy to enhance downstream ALLO levels in veterans with mild TBI. Diffusion tensor imaging (DTI) at baseline and post-treatment was conducted in a subset of participants to interrogate potential target engagement for this neurosteroid intervention.

Methods: Serum samples were collected from INTRuST Biorepository sites. Neurosteroids were quantified by mass spectrometry-based methods and inflammatory markers [cytokines, c-reactive protein (CRP)] were quantified using commercially available kits. In a separate investigation, we conducted an RCT with a neurosteroid intervention in veterans with mild TBI. Veterans with TBI were randomized to PREG or placebo for 8 weeks. A subset of patients received diffusion tensor imaging (DTI) at baseline and post-treatment.

Results: Neurosteroid and inflammatory marker quantifications of INTRuST Biorepository samples demonstrate that ALLO ($p<0.001$), PREG ($p=0.041$), and androsterone ($p=0.008$) levels were significantly decreased in male participants with PTSD, with or without a history of TBI ($n=107$), compared to control subjects ($n=103$). Decreased ALLO ($p=0.001$) and androsterone ($p=0.056$) levels persisted after controlling for age and smoking. ALLO ($p<0.001$) levels were similarly reduced in male participants with a history of TBI (with or without PTSD; $n=129$) compared to controls. ALLO was inversely correlated with PTSD and depression symptoms. IL-6, IL-8, TNF-α, and CRP were significantly elevated in both PTSD and TBI groups compared to control participants.

Results of an RCT using PREG in TBI included 53 randomized participants; 44 participants had at least one post-randomization assessment, and were thus included in the modified intent-to-treat analysis. PREG significantly outperformed placebo by approximately 4 points for the primary behavioral endpoint (Criterion D of the CAPS), which includes sleep difficulties, trouble concentrating, irritability or anger, increased startle response, and hypervigilance - symptoms that overlap considerably with post-concussive syndrome symptoms. In addition, treatment with PREG
appeared to enhance fractional anisotropy assessed by DTI compared to baseline, potentially consistent with its actions on myelination in rodent models.

Conclusions: ALLO, PREG, and androsterone levels were significantly decreased in male participants with PTSD, with or without a history of TBI. ALLO levels were similarly reduced in male participants with a history of TBI (with or without PTSD). ALLO was inversely correlated with PTSD and depression symptoms. Inflammatory markers were also altered in TBI and PTSD. Results from an RCT using a novel neurosteroid intervention in veterans with TBI showed that PREG outperformed placebo by 4 points for the primary behavioral endpoint, and may enhance myelination as assessed by DTI post-treatment. Neurosteroids, pro-inflammatory cytokines, and CRP are promising biomarker candidates for TBI and PTSD. In addition, PREG may have therapeutic utility for the treatment of TBI and associated sequelae.

Keywords: Biomarker, Novel Therapeutics, Neurosteroid, TBI, PTSD

Disclosure: Part 1: N/A (pending patents; no patents issued; no licensing in place; VA 208 waiver in place), Patent.

T170. Poster Withdrawn

T171. Discovery and Characterization of NPT520-34, a Novel Therapeutic Candidate for the Treatment of Parkinson's Disease and Related Neurodegenerative Disorders

Diana Price*, Asma Khan, Wolfgang Wrasidlo, Douglas Bonhaus

Neuropore Therapies, Inc., San Diego, California, United States

Background: The goal of the study is to develop small molecule therapeutics which reduce the accumulation of neurotoxic misfolded and aggregated proteins [e.g., α-synuclein (ASYN) in Parkinson's disease (PD), Beta-amyloid in Alzheimer's disease or Huntingtin in Huntington's disease] and the associated neuroinflammation which have been implicated in neurodegeneration and progression of the disease. NPT520-34 is a structurally novel, orally bioavailable and brain penetrating compound with favorable ADME and safety profiles that is being evaluated for the treatment of PD and related neurodegenerative disorders. Here we present the results of in vivo pharmacology experiments demonstrating that 1-3 months of NPT520-34 administration produced reductions in ASYN, a neuronal protein whose dysregulation has been clearly implicated in the pathogenesis of PD. Moreover, these reductions in ASYN neuropathology are accompanied by improvements in motor function and normalizations in potentially translatable CNS imaging markers for inflammation and the dopamine system.
Methods: The mThy1-alpha-synuclein transgenic mouse model of PD was utilized for studies of NPT520-34 administration. This transgenic mouse model overexpresses wild-type human ASYN under the Thy-1 promoter (commonly referred to as Line 61 transgenic mice; Rockenstein et al., 2002), and develops extensive accumulation of ASYN in areas relevant to PD (Rockenstein et al., 2002; Chesselet et al., 2012; Games et al., 2013), neurodegeneration (including loss of tyrosine hydroxylase immunoreactivity in the striatum (Masliah et al., 2000; Lam et al., 2011)), inflammation (Rabl et al., 2017), and motor and non-motor functional deficits (Fleming et al., 2004). Male transgenic and non-transgenic littermates (3-3.5 months) were assigned to treatment groups in a pseudo-randomized manner based on a baseline assessment of grip strength, and then received vehicle or NPT520-34 (5 or 10 mg/kg, ip) once daily for 3 months. Behavioral assessments were included in the 3-month study and commenced at approximately 10 weeks of treatment and included measures of spontaneous locomotor activity and grip strength. Mice were euthanized following 3 months of NPT520-34 administration and then biological samples were collected to enable bioanalysis of NPT520-34 in plasma and brain and immunohistochemical evaluations of ASYN pathology, inflammation (glial fibrillary acidic protein - GFAP), and striatal dopamine transporters (DAT) in brain.
Results: NPT520-34 administration at 5 and 10 mg/kg (1 or 3 months) produced statistically significant ($p < 0.05$) reductions in the accumulation of aggregated alpha-synuclein (20-71 % in hippocampus, cortex and striatum) and associated inflammation in a transgenic mouse model of Parkinson's disease and dementia with Lewy bodies that overexpresses human wild type ASYN. The beneficial effects of NPT520-34 were persistent, being detected in studies of both one month and three months duration. NPT520-34 treatment ameliorated ASYN pathology to a degree previously determined to be necessary for normalized levels of striatal DAT and reduction of motor symptoms. Accordingly, reductions in ASYN in the 3-month study were accompanied by statistically significant functional benefits of improved grip strength and normalized dopaminergic sensitive locomotor activity patterns in mice treated with 5 or 10 mg/kg NPT520-34.
Conclusions: NPT520-34 has good drug-like properties (orally bioavailable and brain penetrating), an excellent safety profile (devoid of overt cardiovascular and genetic safety liabilities and no findings of concern in initial 7-day duration toxicity studies in dogs and rats) and produces statistically significant reductions in ASYN neuropathology that have been demonstrated to ameliorate additional PD-related pathology and motoric deficits in animal models. Demonstration of beneficial effects on both ASYN protein accumulation and neuroinflammation with consequent improvements in motor function along with the development of imaging markers for early clinical development warrants advancement of NPT520-34 into preclinical development and hold promise as a therapeutic candidate for the treatment of Parkinson's disease and potentially other neurodegenerative disorders.
Keywords: Parkinson's Disease, Animal Models, Experimental Therapeutics
Disclosure: Part 1: Neuropore Therapies, Inc., Employee, Neuropore Therapies, Inc., Stock / Equity, **Part 2:** Neuropore Therapies, Inc., Employee, **Part 5:** Neuropore Therapies, Inc., Employee.

T172. ITI-214, a Potent and Selective PDE 1 Inhibitor: A New Approach to the Treatment of Neurodegenerative Diseases, Including Parkinson's Disease

Jennifer J. O'Brien, Sophie Dutheil, Stephanie Cruz, Lawrence P. Wennogle, Cedric O'Gorman*, Peng Li, Robert E. Davis, Gretchen L. Snyder, Joseph P. Hendrick

Intra-Cellular Therapies, Inc., New York, New York, United States

Background: Neurodegenerative diseases, including Parkinson's disease, Alzheimer's disease, multiple sclerosis and amyotrophic lateral sclerosis, are accompanied by chronic immune activation. Microglia, the resident brain macrophages, under pathological conditions release pro-inflammatory or anti-inflammatory factors depending on state characteristics. It has been suggested that release of IL-1a, TNF and C1q from microglia initiates a cascade of events that lead to activation of a subset of astrocytes and ultimately cause the death of neurons and oligodendrocytes in neurodegenerative disease (Liddelow, 2017). Preventing activation of microglia under pathological conditions may represent a viable therapeutic strategy for treating

neurodegenerative disorders in which neuroinflammation and microglia activation are known components of the disease.

Recently, it has been recognized that elevated intracellular cyclic nucleotide levels control microglia mediated neuroinflammation. The second messenger cyclic adenosine monophosphate (cAMP) acts primarily as a negative modulator of inflammatory cell responses, including cytokine secretion and leukocytes recruitment. Hence, increasing the levels of intracellular cyclic AMP using cyclic AMP analogs, adenylyl cyclase (AC) activators, or phosphodiesterase (PDE) inhibitors, has been shown to antagonize the changes in microglial cell morphology and their production of proinflammatory cytokines in the context of inflammation. Similarly, increasing intracellular cAMP attenuates lipopolysaccharide (LPS)-induced responses in microglia, suggesting that both cAMP is a key mediator of neuroinflammation and is a critical regulator of microglia homeostasis.

Methods: We have developed ITI-214, a potent and specific inhibitor of PDE1 and tested its ability to reverse neuroinflammation in a microglia cell line and in animal models. PDE1 is a dual cyclic nucleotide phosphodiesterase and likely provides a substantial percent of cAMP and cGMP hydrolytic activity in microglia.

Results: ITI-214 reduced LPS-induced increases in TNFα, IL-1β, and Ccl2 mRNA expression by $>50\%$, both in BV2 (murine microglia cell line) cells in vitro and in mouse brain in vivo (striatum, cortex and hippocampus). ITI-214 administration to mice also increased expression of the anti-inflammatory cytokine IL-10 in the brain, supporting an anti-inflammatory property of the compound. These results were replicated when mRNA levels were measured in microglia isolated from the brains of mice treated with LPS and ITI-214. Thus, ITI-214 reduces brain inflammation both by reducing the expression of pro-inflammatory and enhancing the release of anti-inflammatory molecules.

To further understand the actions of ITI-214 on resting and LPS-activated microglia, we examined transcriptional regulation using RNA-Seq on BV2 cells mRNA transcripts. We identified a subset of genes whose transcript expression was significantly changed with PDE1 inhibition. Using gene ontology software (AmiGO 2), these genes were significantly enriched in cell migration and extravasation pathways as well as inflammatory pathways. Of the genes induced by LPS, we found a subset that were attenuated by ITI-214, all of which were significantly associated with inflammatory pathways, demonstrating the unique properties of ITI-214 based on its ability to inhibit PDE1 activity in microglia. Curiously, transcripts that were significantly changed with a PED4 inhibitor, rolipram, were largely distinct from those altered by our PDE1 inhibitor.

Conclusions: These data demonstrate the therapeutic potential for controlling neuro-inflammation via exposure to ITI-214 mediated PDE1 inhibition. Taken together with our previous findings demonstrating that ITI-214 improves motor control in animal models of Parkinson's disease, enhances cognition and increase wakefulness without stimulating locomotor activity (Snyder, et al. 2015), ITI-214 may represent a viable therapeutic strategy for preventing or slowing neurodegeneration in patients with Parkinson's disease and other neurodegenerative or neuropsychiatric diseases in which neuroinflammation and microglia activation is a known component of the disease. We are presently studying the effects of ITI-214 in patients with Parkinson's disease to further understand the role of PDE1 inhibition in improving common symptoms and reducing neuroinflammation in this disease.

Keywords: Parkinson's Disease, Phosphodiesterase 1 Inhibitor, Neuroinflammation

Disclosure: Part 5: Intra-Cellular Therapies, Inc., Employee.

T173. Defects of Myelination are Common Pathophysiology in Syndromic and Idiopathic Autism Spectrum Disorder

BaDoi Phan, Stephanie Page, Morganne Campbell, Joseph Bohlen, Courtney Thaxton, Jeremy Simon, Emily Burke, Joo Heon Shin, Andrew Kennedy, David Sweatt, Benjamin Philpot, Andrew Jaffe, Brady Maher*

Johns Hopkins School of Medicine, Baltimore, Maryland, United States

Background: Autism Spectrum Disorder (ASD) is genetically heterogeneous in nature with convergent symptomatology that suggests the potential for dysregulation of common molecular pathways across the spectrum of the disorder. Autosomal dominant mutation in Transcription Factor 4 (TCF4) causes the rare ASD known as Pitt-Hopkins Syndrome (PTHS) that is characterized by intellectual disability, failure to acquire language, deficits in motor learning, and gastrointestinal abnormalities. To understand TCF4's role in neuronal development and PTHS, we analyzed transcriptional changes in the brains of five independent mouse models of PTHS. We then compared this transcriptional profile to two additional syndromic forms of ASD caused by mutations in MECP2 and PTEN. Lastly, we integrated the transcriptional profiles of these syndromic ASD mouse models to RNA sequencing datasets from postmortem human idiopathic ASD to identify common molecular pathways disrupted across the spectrum of ASD.

Methods: Five independent mouse models of PTHS syndrome with unique mutations in the Tcf4 gene were analyzed in this study (Tcf4+/tr, Tcf4R579W, Tcf4del5740579, Actin-Cre::Tcf4+/floxed, Nestin-Cre::Tcf4+/floxed). Tissue was micro-dissected from mouse cortex and flash frozen. For the developmental timecourse data, TCF4 mRNA and protein levels were quantified in TCF4+/tr mice and wildtype littermates from embryonic day 12 (E12) through postnatal day 42 (P42), $N=3$ per condition. For RNA sequencing data in Tcf4+/tr mice, 6 samples from each genotype at P1, P21, and P42 were sequenced with 100bp paired-end reads at 50 million reads per sample. Reads were aligned to the mm10 mouse reference genome and differential expression (DE) was determined at the gene, exon, and junction levels co-varying for age and un-modeled batch effects. DE features were used in subsequent gene ontology enrichment, Ingenuity Pathway analysis, and cell type-specific expression analysis (CSEA). In addition, we analyzed differentially expressed genes across the TCF4 truncation model of PTHS and two other ASD mouse models, MeCP2 knockout and homozygous Pten mutation. Lastly, we compared our PTHS mouse models with sporadic

human ASD by comparing DE genes to Simons Foundation Autism Research Initiative (SFARI) ASD risk gene database and we also searched for overlap using weighted gene co-expression network analyses (WGCNAs) from postmortem human ASD RNA sequencing data.
Results: We observed Tcf4 RNA and protein levels were significantly blunted during cortical development and throughout postnatal life. Differential expression analysis identified 36 differentially expressed genes (DEGs) at P1 and 1832 DEGs in adult mice (FDR < 0.05). DEGs between PTHS mouse models were most similar in adult mice. DEGs enriched for gene ontology terms associated with synaptic transmission, forebrain development, axon development, and myelination. CIBERSORT analysis predicated a decrease proportion of oligodendrocytes in PTHS mice. Comparisons between PTHS models and other syndromic ASD models found a significant negative correlation between transcriptional changes underlying PTHS compared to Rett syndrome (MeCP2, $P < 0.0023$) and PTEN-related disorders (PTEN, $P < 2.2e\text{-}16$) with common DEGs showing enrichment for gene-sets associated with myelination. Lastly, we identified significant overlap and positive correlation of gene fold-change between adult PTHS mouse DEGs with DEGs from ASD and 15q duplication postmortem brain ($P < 0.001$).
Conclusions: By comparing DE in mouse models of PTHS with human ASD and 15q duplication, we observed similar trends in differential expression between datasets, particularly among convergent ASD genes. The observed positive correlation between altered gene regulation in mouse models of syndromic ASD with those in idiopathic human ASD strongly supports the hypothesis that defects in myelination are a common pathophysiology in ASD. Our cross-species framework demonstrates the usefulness of mouse models to study human ASD and highlights novel targets and pathways for potential pharmaceutical intervention.
Keywords: Autism Spectrum Disorder, Oligodendrocytes, Rett Syndrome, TCF4, PTEN
Disclosure: Nothing to Disclose.

T174. Optogenetic Identification of Granule Cells and Population-Specific Neural Correlates of Space in the Dentate Gyrus

Sang Hoon Kim, Douglas Goodsmith, Stephanie Temme, Guo-li Ming, James Knierim, Hongjun Song, Kimberly Christian*

University of Pennsylvania, Philadelphia, Pennsylvania, United States

Background: The hippocampus is a critical locus for several forms of learning and memory. In rodents, principal hippocampal neurons exhibit spatially-modulated firing that may provide a framework for the integration of environmental features necessary for contextually-embedded memories. It is not fully understood how different subregions, and different neuronal populations within each subregion, contribute to the encoding of spatial information. Among hippocampal subregions, the dentate gyrus is a site of robust plasticity in the form of adult neurogenesis, which results in the continuous integration of maturing granule cells into the local circuitry. Although several computational models of hippocampal-dependent learning and the dentate gyrus ascribe unique properties to adult-born granule cells as they mature, the in vivo properties of this population are difficult to assess using traditional electrophysiological techniques. To be able to discriminate dentate granule neurons of varying ages, we developed an optogenetic approach to permit recording of identified granule cells at the single cell level. Characterizing the properties of distinct populations of dentate gyrus neurons, and how these properties may evolve for adult-born neurons, could generate insight into the mechanisms underlying information processing in the hippocampus that contribute to memory formation.
Methods: We used an optogenetic approach to birthdate and identify dentate granule cells of various ages. Taking advantage of the transient expression of Tbr2 in intermediate neural progenitor cells, we developed a line of mice that allows for inducible expression of channelrhodopsin (ChR2) in specific cohorts of dentate granule neurons. After tamoxifen-induced recombination to express ChR2 in granule cell populations born within a restricted temporal window, we surgically implanted a custom chronic recording drive with 8 independently-adjustable tetrodes and an optical fiber. Animals were allowed to recover from surgery and then we slowly lowered tetrodes to the dentate gyrus over a 1-2 week period. Light pulses were delivered to identify cells that responded with short latencies (< 5ms) and high fidelity that were potentially directly activated via ChR2. Upon identification of clearly isolated units, including both light and non-light responsive cells, animals were allowed to freely forage in multiple distinct environments while the neural activity was recorded. Data were then analyzed offline to quantify parameters of spatial firing including place field size and quality.
Results: Our preliminary data suggest that putative adult-born neurons exhibit spatially-modulated firing that differs across distinct environments. Consistent with recent reports describing the activity of diverse populations of dentate gyrus neurons, we observed several patterns of spatial firing among all cells recorded from the dentate gyrus. These spatial firing patterns correlated with other features of neural activity including spontaneous firing rate during rest and sleep sessions.
Conclusions: Previous reports have suggested that the dentate gyrus is involved in behavioral discrimination among similar stimuli. Adult neurogenesis has been thought to contribute to established dentate gyrus functions, and its dysregulation has been implicated in pathology related to epilepsy and affective disorders. Our data suggest that functional properties of the dentate gyrus emerge from distinct populations, including adult-born neurons, that respond to environmental cues in a selective and non-uniform manner. Understanding the electrophysiological properties of these different dentate gyrus populations at a single cell level can provide a basis for future therapeutic strategies to counteract cognitive deficits arising from hippocampal dysfunction.
Keywords: Adult Hippocampal Neurogenesis, Dentate Gyrus, Optogenetics
Disclosure: Nothing to Disclose.

T175. Behavioral and Neurochemical Effects of Selective Inhibition of Phosphodiesterase 4D in Mice: Evidence for a Key Role of This Enzyme Subtype in Memory

Chong Zhang, Ying Xu, Benjamin Auerbach, Richard Salvi, Mark Gurney, James O'Donnell*

University at Buffalo, SUNY, Buffalo, New York, United States

Background: Inhibitors of phosphodiesterase-4 have been shown to have a range of neuropsychopharmacological effects, including improving measures of memory. Development of these compounds for clinical use has been limited due to their side-effects, notably nausea and emesis. While it has been speculated that development of subtype-selective PDE4 inhibitors (i.e., with selectivity for one of the subtypes expressed in brain, PDE4A, PDE4B, or PDE4D), might exhibit a better therapeutic index, development of selective inhibitors has proven difficult due to the highly conserved nature of the catalytic region/inhibitor binding site. Recently, it has proven possible to develop selective, allosteric inhibitors of PDE4B and PDE4D; assessment of their effects suggests different behavioral profiles. The present study examined the behavioral and neurochemical effects of BPN14770, a novel allosteric inhibitor of PDE4D, in a humanized mouse model; these mice express the primate-specific allosteric binding site for the inhibitor in the N-terminal region of the enzyme.
Methods: Behavioral and neurochemical measures were conducted using a transgenic mouse line in which a key amino acid in the UCR2 region of the PDE4D enzyme was mutated to the primate form (tyrosine to phenylalanine). Assessment of the neurochemical effects of BPN14770 included inhibition of 3H-rolipram binding to mouse brain membranes, increase in brain cyclic AMP/phospho-CREB, and changes in synaptic marker proteins; effects on LTP in hippocampal slices also were determined. Behavioral effects focused on memory measures using the novel object recognition test; brain and plasma levels also were determined. Potential for emetic side-effects were determined using a surrogate mouse model assessing anesthesia time in mice administered ketamine and xylazine.
Results: The PDE4D-selective inhibitor BPN14770 exhibited two-site binding for inhibition of 3H-rolipram binding to brain membranes from humanized mice; by contrast, rolipram, a nonselective PDE4 inhibitor, exhibited one-site binding. BPN14770 increased brain cyclic AMP and phospho-CREB, as well as phospho-synapsin and PSD95; it also enhanced hippocampal LTP. It increased indices of memory in the novel object recognition test; this effect was blocked by the PKA inhibitor H89. In all tests, particularly the novel object recognition test, BPN14770 was more potent in humanized mice than in wild-type mice. BPN14770 showed activity in the ketamine/xylazine test, but at doses 100-fold above those effective in the behavioral tests.
Conclusions: The selective, allosteric inhibitor of PDE4D, BPN14770, exhibited linear kinetics, penetrated well into the brain, increased cyclic AMP signaling and its downstream targets, and enhanced cellular and behavioral indices of memory. Its increased potency in humanized mice compared to wild-type mice, particularly in the tests of memory, suggest a significant role for the PDE4D subtype in the mediation of its effects. Its considerable dose differential for its effects in the behavioral tests compared to the ketamine/xylazine test, suggests a therapeutic index superior to that of rolipram.
Keywords: Memory, Cyclic AMP, Phosphodiesterase-4 (PDE4), Drug Discovery - New Approaches
Disclosure: Nothing to Disclose.

T176. Cortical Inhibition and Excitation in Late-Life Depression: A TMS Study of Baseline Abnormalities and Changes With Pharmacotherapy

Jennifer Lissemore*, Apoorva Bhandari, Benoit Mulsant, Eric Lenze, Charles Reynolds, Jordan Karp, Tarek Rajji, Yoshihiro Noda, Reza Zomorrodi, Etienne Sibille, Zafiris J Daskalakis, Daniel Blumberger

University of Toronto, Toronto, Canada

Background: Most studies on the neurobiology of depression have investigated patients under the age of 60. However, in older adults, depression is prevalent, and treatment resistance and relapse are common. Improvement of current treatments for depression in older populations requires a clearer understanding of the neurobiological mechanisms underlying late-life depression (LLD), including the influence of molecular aging and the neurophysiological correlates of successful LLD treatment. There is evidence in younger adults that GABA and glutamate neurotransmitter systems are impaired in depression and may normalize with treatment, yet there remains a prominent gap in our understanding of GABAergic and glutamatergic functioning at baseline and with treatment in older adults with depression. We present here the first study of GABAergic and glutamatergic neurotransmission in LLD patients both at (i) baseline and (ii) with treatment.
Methods: Using transcranial magnetic stimulation (TMS), a unique tool for the in vivo investigation of human cortical neurophysiology, the present study examined cortical inhibition and excitation in 92 LLD patients 60 years of age and older and 41 healthy, age-matched controls. To differentiate the influence of age and depression, TMS measures of GABAergic and glutamatergic neurotransmission were also compared to those of 32 younger adults with depression and 32 younger healthy controls. For a subset of LLD patients ($n = 67$), TMS measures of cortical inhibition and excitation were then repeated following 12 weeks of open-label treatment with venlafaxine. Clinical response to treatment was defined as a $\geq$ 50% reduction in the Montgomery-Asberg Depression Scale score.
Results: (i) Baseline findings. Compared to younger healthy adults, we observed significant reductions in GABA-A receptor-mediated cortical inhibition in LLD patients and older healthy adults, and similar trending reductions in younger depressed patients. Measures of cortical excitation did not differ significantly between the four groups. Within depressed patients only, a weak correlation was observed between lower cortical inhibition and greater cortical excitation. (ii) Pre-post treatment findings. No significant changes in cortical inhibition or excitation were observed pre-post treatment within a subgroup of responders to

treatment ($n = 29$; mean ± SD MADRS score reduction = 80.8 ± 16.1%).
Conclusions: The similar reductions in cortical inhibition observed in old age and depression across the lifespan suggest that both healthy aging and depressive states involve diminished GABA-A receptor-mediated neurotransmission. Furthermore, deficits in cortical inhibition observed in depression may reflect an excitation-inhibition imbalance. The results of this study are consistent with the age-by-disease hypothesis of LLD, which posits that the molecular changes that occur with aging overlap with those that occur in age-gated diseases such as depression, and that these molecular shifts may contribute to a physiological vulnerability to develop psychopathology in old age. Given that cortical inhibition was not found to normalize following treatment in LLD despite considerable reductions in depressive symptoms, it is possible that aging has a more prominent effect than depression on cortical inhibition in late-life.
Keywords: Late-life Depression, Aging, Cortical Excitability, Transcranial Magnetic Stimulation, GABA transmission
Disclosure: Nothing to Disclose.

T177. Reduction in CSF sTREM2, a Biomarker of Microglia Activation, in Elderly Depressives

Nunzio Pomara*, Davide Bruno, Jay Nierenberg, Henrik Zetterberg, Kaj Blennow, Amanda Heslegrave, Martha Foiani

Nathan S Kline Institute/New York University School of Medicine, Orangeburg, New York, United States

Background: Both intense microglia activation and senescence, which respectively lead to marked and reduced inflammatory responses, have each been implicated in the etiology of depression. However, to date, results from post-mortem and PET studies of microglial status have been negative or inconclusive, perhaps reflecting the heterogeneity of microglia dysfunction in this disorder, and the lack of specificity of the neuroimaging ligands for microglia.
Triggering receptor expressed on myeloid cells 2 (TREM2) is a transmembrane innate immune receptor of the immunoglobulin family; in brain, it is found exclusively on microglia. TREM2 variants have been associated with increased risk for AD and other degenerative disorders. It undergoes proteolytic cleavage generating a soluble form (sTREM2), found in CSF, and emerging data suggest that its levels may reflect microglia activation.
These considerations prompted us to determine if elderly cognitively-intact individuals with a DSM-IV diagnosis of late-life major depression (LLMD) differ from healthy controls with respect to CSF sTREM2 and its relationship to depressive symptoms. Additionally, because of the association between depression and increased risk for AD, we also determined its relationship to CSF AD biomarkers.
Methods: CSF was obtained from 47 older subjects with intact cognition having an MMSE score of at least 28 and no gross MRI abnormalities other than white matter hyperintensities (28 LLMDs, 19 Healthy controls). CSF sTREM2 was determined using an electrochemiluminescence immunoassay. Severity of depression was determined at the baseline and screening visits using the HAM-D.
Results: CSF sTREM2 was significantly ($p < .05$) reduced in LLMD compared to controls. CSF sTREM2 was significantly (negatively) correlated with baseline HAM-D scores in the entire sample and in the depressed group separately. As previously reported in cognitively-intact populations, CSF sTREM2 was significantly (positively) correlated with CSF Aβ42, Aβ40, tau and p-tau. Antidepressant treatment and APOE-e4 status had no effect on CSF sTREM2.
Conclusions: The reduction in CSF sTREM2 level in LLMD and its association with more depressive symptoms suggest that decreased rather than increased microglia activation may contribute to the pathophysiology of this disorder. Future studies should determine how lower CSF sTREM2 levels may relate to activated M1 (pro-inflammatory) or M2 (anti-inflammatory) microglia phenotypes and to inflammatory and AD brain PET biomarkers.
Keywords: CSF Biomarkers, Late-life Depression, Microglia
Disclosure: Nothing to Disclose.

T178. Fear Extinction Deficits in Mice With Impaired Activity-Dependent BDNF Signaling are Associated With Hyperexcitability in Direct Hippocampal Inputs to the Medial Prefrontal Cortex

Henry Hallock*, Julia Hill, Yishan Mai, Keri Martinowich

Lieber Institute for Brain Development, Baltimore, Maryland, United States

Background: The brain-derived neurotrophic factor (BDNF) molecule is important for various forms of learning and memory. In particular, recall of an extinction memory increases BDNF transcription in the medial prefrontal cortex (mPFC) (Bredy et al., 2007), and BDNF infusion into the mPFC facilitates extinction learning (Peters et al., 2010). The ventral hippocampus (vHC) is also critical for extinction learning (Sierra-Mercado et al., 2011), and the CA1 subfield of the vHC sends efferent fibers that synapse directly onto mPFC neurons. Mutant mice with disrupted BDNF production from promoter IV transcripts (-e4 mice) also show impaired fear extinction that correlates with aberrant hippocampal-prefrontal oscillatory synchrony (Hill et al., 2016). These results led us to hypothesize that BDNF signaling in mPFC-projecting vHC neurons may regulate fear extinction. Understanding the neural mechanisms underlying fear extinction will be important for treating extinction deficits in post-traumatic stress disorder (PTSD).
Methods: In order to examine whether activity-dependent BDNF production is necessary for hippocampal to prefrontal communication during fear extinction, we simultaneously recorded local field potentials (LFP) from the mPFC and vHC of both wild-type (w/t) ($n = 9$) and –e4 mice ($n = 9$) during extinction sessions. We applied Granger causality to vHC and mPFC LFP data to assess directionality of information flow between the two structures. In order to causally link mPFC-projecting vHC neurons with fear extinction behavior, we used a dual viral-targeting strategy to express either an inhibitory DREADD receptor (hM4Di), or an excitatory DREADD receptor (hM3Dq) selectively in

these neurons in w/t (n = 5) and e4 mice (n = 4), respectively.

Results: We report that increased freezing during fear extinction in -e4 mice is associated with increased directionality between the vHC and mPFC in the slow gamma (30-50 Hz) frequency band, suggesting that increased hippocampal-to-prefrontal communication is implicated in extinction failure in these mice. We further report that inactivation of mPFC-projecting vHC neurons with injections of clozapine-N-oxide (CNO) 45 minutes prior to extinction caused a decrease in freezing in w/t mice (n = 5 per group: Experimental group = mice with Cre-dependent expression of hM4Di receptors; control group = mice with Cre-dependent expression of an mCherry reporter). Synthetic activation of these neurons in –e4 mice with CNO resulted in an increase in freezing during extinction sessions (n = 4 per group. Experimental group = mice with Cre-dependent expression of hM3Dq receptors; control group = mice with Cre-dependent expression of an mCherry reporter).

Conclusions: Our results suggest that extinction deficits in -e4 mice (i.e., sustained freezing) are caused, in part, by hyperexcitability in mPFC-projecting vHC neurons. This conclusion is supported by the observations that 1) hippocampal-to-prefrontal communication is strongest when extinction is impaired, 2) inactivation of neurons in this pathway facilitates extinction (decreases freezing), and 3) activation of neurons in this pathway impairs extinction (increases freezing). These data indicate that the vHC-mPFC pathway supports the expression of conditioned fear, which may interfere with the formation of an extinction memory in –e4 mice. Whether BDNF signaling directly drives the activation of these neurons during extinction remains unknown. One possibility is that promoter IV-derived BDNF influences interneuron excitability in the vHC, which could be important for maintaining excitatory/inhibitory balance in mPFC-projecting vHC neurons in CA1. Current experiments are underway to elucidate the mechanisms by which BDNF expression in this circuit affects neural activity that controls extinction-related behavior.

Keywords: BDNF, Fear Extinction, Hippocampal-Prefrontal, DREADDs

Disclosure: Nothing to Disclose.

T179. Resting-State Network Abnormalities in Unmedicated Individuals With Schizotypal Personality Disorder

Erin Hazlett*, Cheuk Tang, Edmund Wong, Caitlin Kelliher, Margaret McClure, M. Mercedes Perez-Rodriguez, Antonia New, Philip Szeszko, Monte Buchsbaum, Panos Roussos, Larry Siever

Icahn School of Medicine at Mount Sinai, Bronx, New York, United States

Background: Schizotypal personality disorder (SPD) bears a close resemblance to schizophrenia but with fewer and attenuated abnormalities, thus representing a potentially important and understudied intermediate schizophrenia spectrum phenotype. Examination of abnormalities in SPD can provide a better understanding of the etiology, genetics, treatment, and risk factors associated with psychosis. Although individuals with SPD demonstrate marked temporal lobe abnormalities that resemble schizophrenia, some models suggest relative "sparing" of frontal lobe regions that are typically abnormal in schizophrenia, which our group has supported empirically (e.g., Buchsbaum et al 2002; Koenigsberg et al 2005; Hazlett et al 2008). This "sparing" may serve to protect individuals from frank psychosis and the severe social and cognitive deficits typically observed in schizophrenia (Siever & Davis 2004; Hazlett et al 2008). Understanding brain abnormalities in SPD and their role in cognitive deficits and functional outcome is therefore a critical next step to: (1) identify risk biomarkers associated with schizophrenia; (2) establish plausible biological markers that can be used in early diagnosis and/or assessment of therapeutic outcomes; and (3) identify new therapeutic targets for the treatment of schizophrenia spectrum disorders.

The default mode network (DMN), also known as the task negative network, includes the posterior cingulate cortex and medial prefrontal cortex. Activity in these regions is greater in individuals while at rest than when engaged in goal-directed tasks and therefore, has been linked to internally-guided experiences. The DMN is anticorrelated (i.e. negatively correlated such that as one goes up, the other goes down) with activity in the dorsolateral prefrontal cortex, also known as the task positive network, which plays a primary role in goal-oriented activity. Several studies have identified DMN functional connectivity abnormalities in schizophrenia, but such studies in SPD are lacking. This is the first large-scale study to examine DMN functional connectivity in SPD.

Methods: The sample included 54 healthy control (HC) and 57 unmedicated SPD participants. All participants were recruited from the community using advertisements and received a rigorous clinical and diagnostic assessment including SCID-I for Axis I disorders and SIDP for personality disorders, as well as, resting-state 3T functional and structural MRI (2/3rds of the sample was scanned on Siemens Skyra and 1/3 on Siemens Allegra scanner in the Department of Radiology at Icahn School of Medicine at Mount Sinai). SPD patients were off all psychoactive medications for a minimum of 2 weeks (75% of the SPD participants were never previously medicated). Diagnostic groups were demographically matched (HC: mean age = 42, SD = 10, 33M, 14W; SPD: mean age = 46.1, SD = 11, 45M, 10W, p = ns).

FSL software (v5.0) was used for image processing and analysis. Using FSL FEAT, preprocessing steps included motion correction (MCFLIRT), slice-timing correction, BET brain extraction, spatial smoothing (5mm), high-pass temporal filtering for Skyra scans only, an additional B0 unwarping step was performed using associated B0 fieldmap images. Next, all the preprocessed BOLD images were coregistered to their associated T1 images, then coregistered into MNI space. FSL MELODIC was used to perform a group independent component analysis (ICA) using the temporal concatenation approach to derive spatial maps that represent resting state networks: https://fsl.fmrib.ox.ac.uk/fsl/fslwiki/MELODIC. Finally, the dual regression approach (from FSL) was used to derive "connectivity" maps to the network and additionally perform group differences/correlation/ covariate statistics: https://fsl.fmrib.ox.ac.uk/fsl/fslwiki/DualRegression. For the independent component analysis

used to analyze DMN connectivity, we specifically focused on the prefrontal cortex and superior temporal gyrus (STG), key regions known to play a role in the pathophysiology of schizophrenia.
Results: Compared with HCs, the SPD group had greater functional connectivity in regions comprising the DMN including the medial and lateral prefrontal cortex, as well as bilateral STG ($p < 0.05$, corrected). Among the SPD group, greater Schizotypal Personality Questionnaire (Raine 1991) scores were associated with greater DMN functional connectivity in prefrontal cortex regions.
Conclusions: Our findings are consistent with prior work reporting: (1) greater STG activation during auditory processing (e.g., Dickey et al 2008) and compensatory frontal lobe functioning in SPD (e.g., Hazlett et al 2008); and (2) hyperactivity in the DMN in schizophrenia (e.g., Whitfield-Gabrieli et al 2009) which is thought to contribute to negative symptoms and concomitant social impairments observed in schizophrenia. The pattern of findings is consistent with the hypothesis that DMN-related regions demonstrate greater-than-normal activation in SPD and may therefore, mitigate against the emergence of psychosis. Structural MRI and diffusion tensor imaging data will also be presented for this sample.
Keywords: fMRI, Resting State Functional Connectivity, Schizotypal Personality Disorder, Default Mode Network, Schizophrenia
Disclosure: Nothing to Disclose.

T180. Corticostriatal Connectivity in Antisocial Personality Disorder by Monoamine Oxidase-A Genotype and its Relationship to Aggressive Behavior

Nathan Kolla*, Katharine Dunlop, Jeffrey Meyer, Jonathan Downar

University of Toronto, Toronto, Canada

Background: The influence of genetic variation on resting-state neural networks represents a burgeoning line of inquiry in psychiatric research. Monoamine oxidase A (MAO-A), an X-linked gene, is one example of a molecular target linked to brain activity in psychiatric illness. MAO-A genetic variants, including the high and low variable nucleotide tandem repeat polymorphisms, have been shown to differentially affect brain functional connectivity in healthy humans. However, it is currently unknown whether these same polymorphisms influence resting-state brain activity in clinical conditions. Given its high burden on society and strong connection to violent behavior, antisocial personality disorder (ASPD) is a logical condition to study, since in vivo markers of MAO-A brain enzyme are reduced in key affect-modulating regions and striatal levels of MAO-A show a relation with the functional connectivity of this same region.
Methods: We utilized MAO-A genotyping and seed-to-voxel-based functional connectivity to investigate the relationship between genotype and corticostriatal connectivity in 21 male participants with severe ASPD and 19 male healthy controls.
Results: Dorsal striatal connectivity to the frontal pole and anterior cingulate gyrus differentiated ASPD subjects and healthy controls by MAO-A genotype. Furthermore, high dorsal striatum functional connectivity to the orbitofrontal cortex, anterior insula, and amygdala was associated with high proactive aggression in the ASPD participants carrying the low-activity MAO-A polymorphism.
Conclusions: These results suggest that MAO-A genotype may affect corticostriatal connectivity in ASPD and that these functional connections may also underlie use of proactive aggression in a genotype-specific manner.
Keywords: Magnetic Resonance Imaging, Antisocial Personality Disorder, Monoamine Oxidase-A
Disclosure: Nothing to Disclose.

T181. Exploring the Associations Between Scalp Recorded Electrophysiological Measures With in Vivo Measures of Brain Bioenergetics in Psychotic Disorders

Cagri Yuksel, Robert Rowledge, Sophie Brickman, Fei Du, Dost Ongur, Mei-Hua Hall*

McLean Hospital, Belmont, Massachusetts, United States

Background: Event-related potentials (ERPs) are synchronized neuronal responses to specific sensory or cognitive events. Investigators have used various ERPs phenotypes to probe cognitive and information processing deficits in major neuropsychiatric disorders. P50 sensory gating ERP measures inhibitory mechanisms thought to be crucial for protecting the brain from information overload. Auditory steady state responses at beta and gamma frequency (20-40Hz) index basic brain functions for integrating information within neural circuits and appear to play an important role in perceptual and cognitive processes. Early sensory processing at the level of auditory cortex is assessed with the N1 amplitude ERP. P3 amplitude is related to attention and working memory capacity, whereas its latency reflects stimulus processing speed. Evidence suggests that patients with psychotic disorders have robust deficits on these ERP components and that these ERP components are considered as endophenotypes (biological markers) for psychosis. It is plausible that intact bioenergetic mechanisms are crucial for these responses, as energy is required for sustaining ion gradients and neurotransmission. While there is evidence for impaired ERPs and brain bioenergetics in psychotic disorders, the link between these processes has not been studied. In this study, we investigate the associations of scalp recorded electrophysiological measures with in vivo measures of energy metabolism in psychotic disorders.
Methods: Study sample consisted of a cross-diagnostic psychosis group including eight patients with bipolar disorder with psychotic features, schizophrenia, schizoaffective disorder, and thirteen healthy controls. Electrophysiological measures including sensory gating (P50 ratio), N1 amplitude, P3 amplitude and latency, and auditory steady-state phase-locking response at 20, 30, & 40 Hz. The sensory gating ERP was elicited using the dual-Click paradigm (160 pairs of identical click stimuli, 5-ms duration; 2-ms rise/fall; 500-ms inter-click interval; 10-s inter-trial interval). P3 amplitude and latency ERPs were elicited by the auditory Oddball paradigm (400 binaural tones; 50-msec duration, 5 ms rise/fall times; 15% 1500 Hz target tones; 85% 1000 Hz standard tones). Auditory steady-state response (ASSR) of gamma oscillation was elicited by the auditory steady state

20-30-40-Hz click stimulation paradigm (150 trains of 1-ms white noise clicks, 500-ms duration, 1100-ms stimulus onset asynchrony, 20-30-40-Hz stimulation rate). We applied the same ERP processing procedures as described previously [Hall et al., 2013, 2015]. Phosphorus magnetic resonance spectroscopy and magnetization transfer (31P MT-MRS) at 4T was used to measure phosphocreatine/adenosine triphosphate ratio (PCr/ATP), pH and creatine kinase forward enzyme rate (CK kf) in the frontal lobe. Associations between EEG and 31P MT-MRS measures were tested using linear regression models in both patients and controls.

Results: Our preliminary analyses revealed that, as expected, patients exhibited significant sensory gating deficits (higher P50 ratio) compared to control subjects ($t = -2.28$, $p = 0.035$). In addition, there was a significant association between P50 ratio and CK rate in healthy controls ($r = 0.76$, $p = 0.024$) but not in patients. Higher sensory gating ratio was associated with higher CK rate.

Conclusions: Creatine kinase forward enzyme rate (CK kf) at frontal region is involved in generation of ATP from PCr, and reflects neuronal capacity to respond to increased energy demand. Our results suggest that this bioenergetic mechanism may be critical for sensory gating mechanism, and the coupling of these processes is compromised in psychotic disorders.

Keywords: EEG/ERP electrophysiology, Phosphorus Magnetic Resonance Spectroscopy, Psychosis

Disclosure: Nothing to Disclose.

T182. Guanfacine Augmentation of Cognitive Remediation Therapy in the Schizophrenia Spectrum

Margaret McClure*, Fionna Graff, M. Mercedes Perez-Rodriguez, Daniel Rosell, Erin Hazlett, Harold Koenigsberg, Larry Siever, Antonia New

Icahn School of Medicine at Mount Sinai, Fairfield, Connecticut, United States

Background: Impaired cognition is a hallmark feature of schizophrenia, as well as of schizophrenia spectrum disorders such as schizotypal personality disorder (SPD), and is the best predictor of functional outcome in this population. Cognitive remediation therapy (CRT) has demonstrated efficacy for improving cognition in schizophrenia patients, with generalized gains also made in real-world outcomes following this treatment. Trials of agents designed to enhance cognition, on the other hand, have been largely negative in the schizophrenia spectrum. We have demonstrated substantial positive effects on cognition in the schizophrenia spectrum with guanfacine, an agent that enhances alpha-2 adrenergic activity, although to date no trials have examined this agent's ability to facilitate and consolidate effectiveness of CRT. The current study is also the first to examine the benefits of guanfacine administration during CRT, as well as the first to study the efficacy of CRT in participants with SPD, a group with a similar pattern of cognitive and functional deficits, albeit less severe, to what is seen in schizophrenia.

Methods: We enrolled 28 participants with DSM-IV SPD in an 8-week, randomized, double-blind, placebo-controlled trial of guanfacine paired with CRT. Participants, who ranged in age from 22-62 (mean = 43.78, SD = 11.44), were recruited from the community in and around New York City and were free of psychotropic medication. Fifteen participants were randomized to guanfacine and 13 to placebo. CRT consisted of a combination of computer-based cognitive enhancement exercises (Psychological Software Services) and manualized-social skills training modified from the Cognitive Enhance Therapy program (Hogarty and Greenwald, 2006). Specifically, participants attended two 60-minute sessions of computer based cognitive enhancement exercises plus two 60-minute of social-skills training sessions each week (4 hours total per week) over 7.5 weeks for a total of 30 sessions (15 computer-based CRT + 15 social skills training). Cognitive performance was assessed using several subtest of the Matrics Clinical Consensus Battery (MCCB), the modified AX-CPT, the N-back, the Paced Auditory Serial Addition Test (PASAT), and the DOT test. Social cognition was assessing with the Movie for the Assessment of Social Cognition (MASC). Functional skills were assessed with the UCSD Performance Based Skills Assessment (UPSA). This battery was administered both pre- and post-treatment.

Results: Adherence to the protocol was excellent; all participants completed the trial with the exception of one who was withdrawn due to an elevated blood pressure at weekly check-in (this participant was later revealed to be on placebo), with an overall mean attendance of 83.33% of sessions. To evaluate the efficacy of the intervention, we conducted a series of repeated measures ANOVAs for each of our DVs with time (pre- and post) and medication status (guanfacine and placebo) as our IVs. We found statistically significant main effects for time (pre- versus post-) on MCCB speed of processing ($p = .015$), verbal learning ($p = .011$), visual learning ($p = .032$), and, as well as UPSA total score ($p = .025$), suggesting that overall SPD participants benefited from the intervention. In addition, there was a significant time x medication interaction for MCCB planning and organization ($p = .005$), and UPSA total score ($p = .026$), with individuals in our guanfacine group demonstrating greater improvement following treatment than those in our placebo group. There were no significant improvements on our working memory, context processing or social cognition measures (all $ps > .05$) in this trial.

Conclusions: Across groups, participants with SPD appeared to benefit from CRT both in terms of improved cognition and functional skills. In addition, participants with SPD who were treated with CRT plus guanfacine demonstrated statistically significant improvements in reasoning and problem solving and in their functional skills over and above those treated with CRT plus placebo. These results suggest that CRT may in fact have even broader efficacy in the larger schizophrenia spectrum, and may be an appropriate intervention for individuals with SPD. In addition, they indicate that guanfacine is an effective agent to augment CRT in the schizophrenia spectrum and this should be explored in future trials.

Keywords: Schizophrenia; Functional Capacity; Technology, Cognitive Functioning, Cognitive Enhancement, Guanfacine

Disclosure: Nothing to Disclose.

T183. DNA Methylation and Gene Transcription of BAIAP2 and DLG1, Regulators of Dendritic Spine Structure and Function, in the Superior Temporal Gyrus of Subjects With Schizophrenia

Brandon McKinney*, Ying Ding, Jennifer Kuflewski, David Lewis, Robert Sweet

University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania, United States

Background: Reduced dendritic spine density (DSD) in the cortex of the superior temporal gyrus (STG) is consistently observed in postmortem studies of schizophrenia (SZ). Elucidating the molecular mechanisms of this intermediate phenotype holds promise for understanding SZ pathophysiology and identifying SZ treatment targets. DNA methylation (DNAm), the addition of a methyl group to a cytosine nucleotide, regulates gene transcription and is a strong candidate for such a mechanism.
Methods: We tested the hypothesis that DNAm correlates with DSD in the human STG and that this relationship is disrupted in SZ. We used the Illumina Infinium HumanMethylation450 Beadchip Array to quantify DNAm on a genome-wide scale in the postmortem STG from 22 SZ subjects and matched non-psychiatric control (NPC) subjects; DSD measures were available for 17 of the 22 subject pairs.
Targeted bisulfite sequencing and quantitative PCR were used to further characterize DNAm-DSD correlations and investigate DNAm-gene transcription relationships for candidate gene.
Results: We found DNAm to correlate with DSD at more sites than expected by chance in NPC, but not SZ, subjects. Additionally, we show that the slopes of the linear DNAm-DSD correlations differed between SZ and NPC subjects at more sites than expected by chance. From these data, we identified 2 candidate genes for mediating DSD abnormalities in SZ: brain-specific angiogenesis inhibitor 1-associated protein 2 (BAIAP2) and discs large, drosophila, homolog of, 1 (DLG1).
Targeted bisulfite sequencing results for BAIAP2 and DLG1, and quantitative PCR results for each of the major BAIAP2 and DLG1 transcript variants, are pending.
Conclusions: Together, these data suggest that altered DNAm in SZ may be a mechanism for SZ-related DSD reductions and identify BAIAP2 and DLG1 as promising candidate genes through which this mechanism may act.
Keywords: Schizophrenia, DNA Methylation, Dendritic Spines, Epigenetics, Transcription
Disclosure: Nothing to Disclose.

T184. Treatment With Antipsychotic Combinations: Lessons Learned From Positron Emission Tomography (PET)

Gerhard Gründer*, Paul Cumming, Hartmut Lüddens, Michael Paulzen, Christoph Hiemke, Ingo Vernaleken

RWTH Aachen University, Aachen, Germany

Background: Binding of antipsychotics to D2-like dopamine receptors in the human brain has been extensively studied with positron emission tomography (PET). For most of the available antipsychotic compounds, the occupancy at brain D2 receptors occupancy can be calculated relative to the plasma concentration of drug. However, occupancy of neither the D2 nor any other receptor has ever been studied under treatment with a combination of (antipsychotic) drugs. We now present a model for calculating occupancy during combination therapy.
Methods: Values for the maximum attainable receptor occupancy (Emax) and the serum concentration predicted to provide 50% of the maximum attainable receptor occupancy (EC50) from our own series of [18F]fallypride PET studies with amisulpride, aripiprazole, clozapine, quetiapine and ziprasidone as well as published values for olanzapine and risperidone are used as inputs for a mathematical model, which is based on the law of mass action (Michaelis-Menten kinetics). Here, the occupancy [%] = (Emax x [C])/(EC50 + [C]) ([C] = plasma concentration) takes into consideration the competition of two (or more) different compounds with different affinities. Occupancy of striatal D2-like dopamine receptors under antipsychotic combination treatment is modelled according to a modification of the Michaelis-Menten equation, where occupancy [%] = 100 – 100 / (1 + AP1/IC501 + AP2/IC502), with AP1 and AP2 being the antipsychotic drug serum concentrations of two drugs, and IC501 and IC502 representing corresponding concentrations for 50% receptor occupancy as derived from our own and published PET studies.
Results: Calculations of D2 receptor occupancy values under treatment with clinically common antipsychotic combinations (e.g., clozapine/aripiprazole; aripiprazole/quetiapine; clozapine/amisulpride; amisulpride/olanzapine; clozapine/risperidone) will be presented in tabular form for a broad range of serum antipsychotic concentrations. As an example, we provide data for the combination of aripiprazole and clozapine. Assuming IC50 values of 9 ng/ml for aripiprazole and 580 ng/ml for clozapine (as derived from our own [18F] fallypride PET studies), combination treatment with these drugs at serum concentrations at the low end of the recommended therapeutic reference ranges (aripiprazole: 100 ng/ml; clozapine 350 ng/ml) will lead to 92% occupancy at striatal D2-like dopamine receptors. Monotherapy with aripiprazole at 100 ng/ml serum concentration leads to a negligibly rlowe D2 occupancy (91.6%). Elevation in serum clozapine concentration to a very high, clinically unrecommended, concentration of 1000 ng/ml, increases striatal dopamine D2-like occupancy to 93%. On the other hand, a decrease in serum aripiprazole concentration to 50 ng/ml, which is below the therapeutic reference range and an elevation of clozapine to 500 ng/ml is associated with a decrease in striatal dopamine D2-like occupancy to 87%.
The combination of amisulpride with olanzapine is a typical example for the combination of two drugs with similar affinities for D2-like dopamine receptors. A moderate daily dose of 600 mg amisulpride (EC50: 60 ng/ml, derived from our own PET studies), which is associated with a serum concentration of 200 ng/ml, leads to 77% striatal dopamine D2-like occupancy. Adding a daily dose of 10 mg olanzapine (EC50: 10 ng/ml, from Kapur et al., Am J Psychiatry 1998; 155: 921-28), which is associated with an olanzapine serum concentration of 20 ng/ml, increases striatal dopamine D2-

like occupancy to 84%. This is above the 80% threshold generally associated with high risk of extrapyramidal side-effects.
Conclusions: The model illustrates that striatal D2-like receptor occupancy readily exceeds the 80% threshold level for extrapyramidal side-effects when two compounds with moderate to high affinity are combined. We contend that the potential beneficial effects of combination treatment can hardly be attributed to (increased) D2-like receptor occupancy, but must rather be ascribed to mechanisms beyond D2 antagonism.
Keywords: Polypharmacy, Schizophrenia, Dopamine, Antipsychotics, Antipsychotic Treatment
Disclosure: Part 1: Boehringer Ingelheim, Consultant, Grant, Eli Lilly, Advisory Board, Honoraria, Lundbeck, Advisory Board, Consultant, Ono Pharmaceuticals, Consultant, Otsuka, Advisory Board, Honoraria, Roche, Advisory Board, Consultant, Honoraria, Grant, Neuraxpharma, Honoraria, Janssen-Cilag, Advisory Board, Honoraria, Servier, Advisory Board, Honoraria, Trommsdorf, Honoraria, Takeda, Advisory Board, Consultant, Brainfoods GmbH, Stock / Equity, Pharma Image GmbH, Stock / Equity, **Part 2:** Lundbeck, Consultant, Brainfoods GmbH, Stock / Equity, Pharma Image GmbH, Stock / Equity, **Part 3:** Lundbeck, Consultant, **Part 4:** Boehringer Ingelheim, Grant, Roche, Grant.

T185. Determining the Effects of a Missense Variant in SLC39A8 on Serum Metals and Protein Glycosylation in Schizophrenia

Robert Mealer*, Mark Daly, Richard Cummings, Ed Scolnick, Jordan Smoller

Harvard Medical School, Massachusetts General Hospital, Boston, Massachusetts, United States

Background: Genome-wide association studies have identified a large number of a genes associated with schizophrenia. One variant with high prevalence in the general population (~5%) and a strong correlation to schizophrenia ($p < 8x10-15$) is a missense mutation in the zinc/manganese transporter SLC39A8. Manganese (Mn) is a critical cofactor for certain enzymes regulating the post-translational addition of sugar polymers to proteins through glycosylation. Severe deficiency of SLC39A8 activity in humans leads to a profound neurologic phenotype with altered transferrin glycosylation and low Mn levels.
Methods: Using the Partners Biobank, a repository of biological samples linked to the electronic medical record (EMR) and genomic data, we developed a search algorithm to identify controls and patients with schizophrenia confirmed by chart review, and stratified these patients into groups based on the presence or absence of the SLC39A8 missense mutation. Serum obtained from the bio-repository was analyzed for Mn and Zn levels using Inductively Coupled Plasma Mass Spectrometry (ICP-MS), as well as protein N-glycosylation patterns using mass spectrometry.
Results: Search of the Partners Biobank identified controls and patients with schizophrenia with and without the missense SLC39A8 mutation, as well as small number of patients homozygous for the mutation. The SLC39A8 missense mutation showed a dose-dependent effect on the serum concentration of Mn with no effect Zn levels, consistent with prior studies suggesting the primary physiologic role of SLC39A8 as a Mn transporter. At the time of abstract submission, serum glycosylation patterns had been measured from ~40 patients and are pending analysis.
Conclusions: By comparing clinical characteristics (such as diagnosis, gender, age, race, treatment response, hospitalizations, duration of illness, substance use, and co-morbid psychiatric conditions) with SLC39A8 allele, Mn concentration, and protein glycosylation patterns, we hope to identify potential biomarkers, common molecular pathways, or endophenotypes for schizophrenia. Utilizing the Partners Biobank, a growing repository with over 40,000 stored serum samples linked to genomic and EMR data, we identified controls and patients with and without the SLC39A8 missense mutation linked to schizophrenia for our study. If data support a role of manganese and glycosylation in the etiology of schizophrenia, dietary supplementation with manganese or glycosylation precursors may represent novel treatment approach for this disorder.
Keywords: Schizophrenia, Manganese, Glycosylation
Disclosure: Nothing to Disclose.

T186. Direct Evidence for the Impaired Modulation of Hippocampal Glutamate During Memory Consolidation in Schizophrenia: A Novel Application of ^{1}H Functional Magnetic Resonance Spectroscopy

Jeffrey Stanley*, Patricia Thomas, Dalal Khatib, Asadur Chowdury, Usha Rajan, David Rosenberg, Luay Haddad, Amirsadri Alireza, Vaibhav Diwadkar

Wayne State University School of Medicine, Detroit, Michigan, United States

Background: Glutamate plays a central role in frontal-hippocampal mechanisms of learning and memory. The hippocampus is particularly rich in glutamatergic neurons and N-methyl-D-aspartate (NMDA) mediated synaptic plasticity is central for sub serving learning. Unsurprisingly, altered neuroplasticity related to glutamate has been proposed as a critical mechanism mediating learning and memory deficits in schizophrenia. Understanding glutamate-related hippocampal dysfunction in schizophrenia may therefore, elucidate not only mechanisms underlying the illness, but may subsequently help in tailoring intervention strategies. However in vivo studies have largely relied on hemodynamic signals based on fMRI to assess hippocampal function (and dysfunction) in schizophrenia, or on quantitating static levels of glutamate with ^{1}H MRS. These signals are not informative of functional modulation of hippocampal glutamate. Here we provide the first ever application of in vivo ^{1}H functional MRS (fMRS) to assess dysfunctional modulation of hippocampal glutamate in schizophrenia. fMRS is a highly novel method for quantitating functional neurochemistry and can provide direct evidence of normal and impaired hippocampal function (Stanley et al., 2017).
Methods: Fifteen early course DSM-V schizophrenia patients (10M + 5F; 25.9 ± 3.7yrs) and 13 healthy, young adults (8M + 5F; 25.8 ± 3.9yrs) participated in the ^{1}H fMRS study at 3T.

The associative learning and memory previously assessed in schizophrenia using fMRI (Wadehra et al., 2013) involved epochs of encoding (9 unique object-location pairs) and cued-retrieval (of those associated memoranda) and interspersed with rest epochs. Eight encoding-retrieval cycles were employed to allow learning to asymptote. Mixed rest epochs of either 27s (64% of data) or 9s were interspersed between task blocks. A visual flashing checkerboard task was introduced prior to the onset of the learning task to provide a non-hippocampal task as a non-memory baseline condition. A total of 19 consecutive single-voxel, short-TE ^{1}H spectra of 16 averages each were acquired from the right head/body of the hippocampus (voxel:1.7x3.0x1.2cm or 6.12 cm3, PRESS, TE = 23ms). The resulting ^{1}H spectra from the 8 encoding/rest epochs and 8 retrieval/rest epochs as well the 3 ^{1}H spectra from the baseline condition were quantified using LCModel. Performance on task was estimated by modeling the behavioral data (expressed as a proportion correct by epoch) using the three-parameter Gompertz function (learning rate, asymptote and inflection point). The statistical analyses assessed changes in glutamate across task conditions (encoding, retrieval) relative to baseline for each group as well as a group-by-task condition interaction for glutamate using the repeated measure GEE approach with task (encoding, retrieval) and time (epoch number) as within subject factors, and age and sex as covariates.

Results: Regarding performance, both groups showed negatively accelerated learning with no statistical group differences on the learning rate, asymptote or inflection point (all $p > .20$).

Independent of time, the task condition for glutamate was significant in controls ($p = 0.020$) with post-hoc tests demonstrating a significant increase in hippocampal glutamate during both encoding (5.9%; $p < .0001$) and retrieval (3.7%, $p = .028$) compared to baseline. In contrast, there were no significant differences in glutamate across conditions in schizophrenia patients. Additionally, the group-by-task interaction was not significant ($p = .23$); however, post-hoc analyses indicated a significantly higher glutamate modulation during encoding in controls compared to schizophrenia ($p = 0.025$), while glutamate modulation during retrieval was not significant between groups ($p = .64$).

Conclusions: The results indicate that even as schizophrenia subjects were engaged in the task, modulation of hippocampal glutamate was absent during encoding and retrieval. These results provide the first assessment, and evidence of impaired functional neurochemistry of the hippocampus in schizophrenia. In healthy controls, the increased levels of hippocampal glutamate observed during encoding and retrieval may be driven by the influx of oxidative metabolism related to increased neuronal activity (Schaller et al., 2014), but assessed independently of the confounds of hemodynamics. By inference, therefore, impaired glutamate modulation in schizophrenia may reflect a lack of neuronal engagement during learning. Though further studies are warranted (and are ongoing), our results provide the first direct evidence of a hippocampal dysfunction related to glutamatergic neurotransmission during processes of memory consolidation in schizophrenia.

Keywords: Glutamate, fMRS, Learning and Memory, Schizophrenia

Disclosure: Nothing to Disclose.

T187. A Proof-Of-Mechanism Study of the PDE10 Inhibitor RG7203 in Patients With Schizophrenia and Negative Symptoms Probing Reward Functions With Imaging and Behavioral Approaches

Daniel Umbricht*, Juergen Dukart, Markus Abt, Paul Tamburri, Christopher Chatham, Michael Frank, Anne Collins, David Walling, Rick Mofsen, Lev Gertsik, Lovingly Park, Jeff Sevigny

F. Hoffmann-La Roche, Ltd., Basel, Switzerland

Background: The enzyme phosphodiesterase 10 is highly expressed in the striatum where it modulates both dopamine D2 and D1 dependent signaling. Its inhibition leads to a suppression of D2 mediated signaling –similar to effects of D2 antagonists - and an enhancement of D1 dependent signaling. D1-dependent signaling has been implicated in reward based learning. Its deficient activation may be a key factor underlying deficient reward functions including reward anticipation and reward based learning that have been implicated as a major driver of negative symptoms of schizophrenia. Therefore, inhibition of PDE10 could be a way to ameliorate such a deficit. In healthy volunteers, the PDE10 inhibitor RG7203 indeed enhanced performance in tasks that probed reward functioning suggestive of its potential utility to enhance reward functions and by extension treat negative symptoms in schizophrenia. We therefore tested the hypothesis that it should enhance imaging and behavioral markers of reward functions in patients with moderate negative symptoms in order to establish mechanistic proof of its utility as treatment of negative symptoms.

Methods: In a three-way cross-over study we investigated the effects of two doses of RG7203 (5 mg and 15 mg) and placebo given as adjunctive treatment to stable background antipsychotic treatment on reward functioning and reward-based effortful behavior using the monetary incentive delay (MID) task during fMRI and the effort choice task in patients with chronic schizophrenia and moderate levels of negative symptoms. Patients had to show a score of at least 18 points on the PANSS negative symptom factor score at screening. Each treatment period lasted three weeks followed by a 2-week washout period. fMRI and behavioral tasks were administered at the end of each treatment period. Key outcome measures were the differential BOLD activity during reward anticipation versus control condition in the MID task, overall BOLD activity during the MID task and the percentage of high-effort high-reward choices when the probability of reward was 100% during the effort choice task.

Results: Thirty-three patients with schizophrenia (30 male; 21 B, 9 W, 3 A; mean age 36.6 ± 7 y) were recruited at three study centers in the US. At study entry PANSS NSFS was 22.8 ($\pm$1.4). Twenty-four subjects finished the entire study. Two patients dropped out due to adverse events (dystonic reactions), seven due to non-safety related issues. Patients were able to do the fMRI and behavioral tasks. RG7203 at 5 mg significantly increased differential BOLD activity during reward anticipation relative to control condition in the MID task. However, this enhancement occurred in the context of a significant overall decrease of BOLD activity during the MID task observed across all conditions. RG7203 significantly worsened reward-based effortful behavior in the effort choice

task, that is during treatment with PDE10 patients chose the high-effort high-reward significantly less often than during placebo treatment (67% for both doses of RG7203 versus 73% for placebo). A multiple regression revealed that the decrease in effortful behavior was significantly related to the decrease in overall BOLD activity during the MID task and not related to the relative increase of BOLD activity during reward anticipation observed at 5 mg.

Conclusions: In contrast to our expectation and previous results in healthy volunteers, RG7203 worsened indices of reward functions likely due to a further enhancement of D2 antagonistic activity. The results do not support the utility of a PDE10 inhibitor as adjunctive treatment for negative symptoms in patients with schizophrenia. Given the previous observation that RG7203 enhanced reward functions in healthy volunteers who were not treated with D2 antagonist, the results of our studies point to potentially deleterious effects of D2 blockade on reward functions and by extension on negative symptoms of schizophrenia and raise the question if the presence of D2 antagonistic treatment curtails the potential of any adjunctive treatment for negative symptoms.

Keywords: Schizophrenia Negative Symptoms, Experimental Medicine, Monetary Incentive Delay Task, BOLD Imaging, Effort-Cost Benefit Task

Disclosure: Part 1: F. Hoffmann - La Roche, Ltd., Employee, Novartis, Stock / Equity, **Part 2:** F. Hoffmann - La Roche, Ltd., Employee, Novartis, Stock / Equity, **Part 3:** F. Hoffmann - La Roche, Ltd., Employee, **Part 5:** F. Hoffmann - La Roche, Ltd., Employee.

T188. Further Insights Into the Neurobiological Basis of the Alterations of the Binocular Depth Inversion Illusion in Schizophrenia

F. Markus Leweke*, Juliane K. Mueller, Bettina Lange, Cristina E. Topor, Stefan Fritze, Dagmar Koethe, Cathrin Rohleder, Dusan Hirjak

The University of Sydney, Camperdown, Australia

Background: Binocular depth inversion illusion (BDII) represents a naturally occurring illusion of visual perception that involves higher-order visual and cognitive processes. Its impairment has been repeatedly linked to psychotic conditions (including delta-9-tetrahydrocannabinol induced altered states of consciousness) and identified as a potential marker for at risk mental states. Today, the neurobiological foundations of altered BDII in these clinical conditions are not well understood. We therefore investigated the association of BDII alterations to structural changes by respective structural MRI data.

Methods: After written informed consent, performed the binocular depth inversion illusion test (BDIIT) was performed in 22 antipsychotic-naive, right-handed, first-episode, antipsychotic-naïve schizophrenia patients (SZ) that also received a structural MRI scan (1.5 T). The BDIIT was performed as previously described (Leweke et al. 2009). The surface-based analysis via new version of Freesurfer (6.0) enabled calculation of cortical thickness and surface area. BDII total and faces scores were related to the two distinct cortical measurements.

Results: Higher BDII faces scores were positively associated with the right postcentral gyrus and pars triangularis of the right inferior frontal gyrus ($p < 0.01$, uncorr.). BDII faces right way up (FRWU) score was positively associated with cortical thickness in the right superior parietal, postcentral, supramarginal and precentral gyrus ($p < 0.05$, corr.). BDII faces upside down (FUSD) score was negatively associated with cortical thickness in the superior parietal and pericalcarine gyrus ($p < 0.01$, uncorr.).

Conclusions: The present study investigated for the first time the associations of cortical thickness to BDII scores in schizophrenia. BDIIT represents a test of impaired ability to mentally invert hollow (concave) objects to familiar convex percepts. BDII serves as a sensitive measure of perceptual alterations in ultra-high risk individuals for schizophrenia as well as acute psychotic states including schizophrenia (Koethe et al. 2009).

Recently, we reported a neurochemical investigation suggesting that frontal and cortical areas that rinse through the bridge veins rather than the spinal canal may play a relevant role in BDII alterations in schizophrenia (Reuter et al. 2016). Our current study provides first evidence that altered BDII performance may be linked to cortical thickness and surface area variations in frontal and parietal brain areas regions involved in "adaptive" or "top–down" modulation and stimulus processing. Our results suggest that cortical features of distinct evolutionary and genetic origin may differently contribute to BDII performance in schizophrenia.

Keywords: Visual Perception, Antipsychotic-Naïve First-Episode Schizophrenia, Cortical Thickness, Frontal Cortical

Disclosure: Part 1: Curantis UG (Ltd.), Stock / Equity, AstraZeneca, Honoraria, **Part 4:** Acerus Pharmaceuticals, Grant.

T189. Favorable Clinical Safety Profile for Lumateperone (ITI-007) - Switching From Standard-Of-Care Antipsychotic Therapy in Patients With Schizophrenia

Kimberly Vanover*, Cedric O'Gorman, Steven Glass, Jelena Saillard, Michal Weingart, Sharon Mates, Christoph Correll, Robert Davis

Intra-Cellular Therapies, Inc., New York, New York, United States

Background: Current standard-of-care antipsychotic therapy is often limited by safety issues that include weight gain, hyperlipidemia, extrapyramidal side effects, akathisia, hyperprolactinemia, and/or cardiovascular liability, such as QTc prolongation. It remains an unmet medical need to reduce the symptoms of schizophrenia without compromising safety.

Lumateperone (ITI-007) is an investigational drug that provides selective and simultaneous modulation of serotonin, dopamine and glutamate. Lumateperone is a potent serotonin 5-HT2A receptor antagonist, a dopamine receptor phosphoprotein modulator (DPPM) acting as a presynaptic partial agonist and postsynaptic antagonist at dopamine D2 receptors, a dopamine D1 receptor-dependent indirect modulator of glutamate (both NDMA and AMPA), and a serotonin reuptake inhibitor. Lumateperone demonstrated antipsychotic efficacy in two well-controlled clinical trials and demonstrated consistent improvements from baseline on the Positive and Negative Syndrome Scale (PANSS) total

score across three clinical trials in patients with acute schizophrenia. In these acutely ill patients, lumateperone was found to be well tolerated with a safety profile similar to placebo. The purpose of the present study was to evaluate the safety and tolerability of lumateperone administered to patients with stable schizophrenia switched from standard-of-care antipsychotic therapy.

Methods: In an open-label safety study, 302 patients with schizophrenia were enrolled and treated for 6 weeks with lumateperone (ITI-007 60 mg) administered orally once daily in the evening, with no dose titration necessary. Following treatment with lumateperone, patients were switched back to standard-of-care antipsychotic therapy and reassessed approximately 2 weeks after the last dose of lumateperone. To be eligible for inclusion in the study, patients must have had a clinical diagnosis of schizophrenia according to the Diagnostic and Statistical Manual of Mental Disorders (DSM-5) and be stable with respect to their schizophrenia symptoms. The primary objective was to determine the safety of lumateperone, assessed by adverse events, body weight, 12-lead electrocardiograms, vital signs, clinical laboratory tests, motor assessments [Barnes Akathisia Rating Scale (BARS), Simpson-Angus Scale (SAS), and Abnormal Involuntary Movement Scale (AIMS)], and the Columbia-Suicide Severity Rating Scale (C-SSRS). The secondary objectives were to determine the effectiveness of lumateperone to improve psychopathology as measured by the PANSS, social functioning as measured by the PANSS Pro-Social Factor and the Personal and Social Performance Scale (PSP), and depression as measured by the Calgary Depression Scale for Schizophrenia (CDSS).

Results: Lumateperone was well tolerated with a favorable safety profile. The most frequent drug-related treatment-emergent adverse event was somnolence. There was no drug related serious adverse event. Mean total scores on the BARS and SAS improved slightly with a switch to lumateperone treatment from standard-of-care, consistent with no clinical signal for akathisia or EPS, respectively. Mean scores on the AIMS showed no change, consistent with no emerging dyskinesia.

In contrast to many other antipsychotics that cause weight gain, mean body weight significantly decreased with lumateperone treatment. Lumateperone also demonstrated a favorable cardiometabolic and endocrine safety profile. Mean levels of cholesterol, triglycerides and prolactin significantly improved with lumateperone treatment and worsened again when patients returned to standard of care. The cardiovascular safety of lumateperone was also favorable with no change in multi-positional blood pressure or heart rate, no orthostasis, and no QTc interval prolongation. There was no emergence of suicidal ideation or behavior with lumateperone treatment.

Importantly, stable symptoms of schizophrenia did not worsen with lumateperone treatment. Although this open-label safety study was not designed to measure efficacy, statistically significant improvements were observed in change from baseline of the PANSS total scores in this stable patient population switched from standard of care antipsychotic therapy. Statistically significant improvements were also seen in the Positive symptom subscale scores, General Psychopathology subscale scores, Marder Negative Factor scores, and Prosocial Factor scores as well as in social function as measured by the Personal and Social Performance (PSP) scale in this stable patient population. Negative symptoms also improved significantly with lumateperone treatment in a subgroup of patients with prominent negative symptoms at baseline as measured by the PANSS negative symptom subscale and the Marder Negative Factor. Symptoms of depression improved significantly with lumateperone treatment in a subgroup of patients with schizophrenia and comorbid symptoms of depression.

Conclusions: Lumateperone represents a novel approach to the treatment of schizophrenia with a favorable safety profile in clinical trials. The lack of metabolic, motor and cardiovascular safety issues presents a safety profile differentiated from standard-of-care antipsychotic therapy. Moreover, patients with stable symptoms on other antipsychotics may further improve when switched to lumateperone, with no dose titration needed, especially patients with prominent negative symptoms and/or comorbid depression. These data may warrant further investigation in appropriately controlled trials in patients with prominent negative symptoms and, separately, in patients with comorbid depression to demonstrate efficacy in these populations. These data, taken together, are consistent with and extend data previously reported in placebo-controlled studies in patients with acute schizophrenia with lumateperone.

Keywords: Schizophrenia, Metabolic Safety, Antipsychotic

Disclosure: Part 1: Intra-Cellular Therapies, Employee, **Part 2:** Intra-Cellular Therapies, Employee, **Part 3:** Intra-Cellular Therapies, Employee, **Part 5:** Intra-Cellular Therapies, Employee.

T190. Anticipatory Pleasure Predicts Effort Expenditures and Persistence in Reward Motivated Behavior

Pamela DeRosse*, Anil Malhotra

Zucker Hillside Hospital, Glen Oaks, New York, United States

Background: Variations in motivational states and sensitivity to reward contingencies have long been associated with negative symptoms such as anhedonia and avolition. However, the introduction of the Research Domain Criteria (RDoC) has led to a substantial resurgence of interest in the examination of these relationships. Recent findings linking negative symptoms to variation in reward processing hold promise for the potential identification of much needed biomarkers for novel treatment development. Critically, however, these emergent findings are often derived from the study of patients with psychotic and affective disorders who are undergoing pharmacologic treatment with dopaminergic agents, which may be directly impacting reward circuitry. Thus, the present study sought to examine the relationship between negative symptoms and reward processing in a non-patient sample.

Methods: A demographically diverse sample of 77 adult participants with no history of psychiatric illness were assessed for attenuated negative symptoms using the Temporal Experiences of Pleasure Scale (TEPS) and the Schizotypal Personality Questionnaire-Brief (SPQ-B). We then sought to examine whether variation in these symptoms could predict variations in both willingness to expend effort for reward across different probabilities and magnitudes of

reward as well as the persistence of reward motivated behavior during the Effort Expenditures for Rewards Task (EEfRT).
Results: We found that lower scores on the anticipatory pleasure subscale (ANT) of the TEPS, reflecting the most problematic component of anhedonia for patients with negative symptoms, predicted lower effort expenditures on the EEfRT regardless of the probability of obtaining a reward or the magnitude of the potential reward ($F(2,74) = 3.34$; $p = .04$). Moreover, we found that relative to participants with high scores on the ANT, those with low scores showed a significant reduction in the persistence in effortful behavior across trials; behavior consistent with avolitional symptoms. However, this finding only applied to those trials in which there was a 50% chance of obtaining a reward ($F(2,70) = 10.91$; $p < .001$) perhaps suggesting that individuals with negative symptoms fail to persist in goal-directed behavior when the outcome of such behavior is not assured. Notably, we also found that although the negative symptom domain of the SPQ-B was very significantly correlated with the ANT scores ($r(76) = -.30$; $p = .009$), the overall negative symptom score was not significantly predictive of performance on the EEfRT. This finding provides further support for the utility of examining distinct components of the negative symptom construct.
Conclusions: These results provide converging evidence for the relationship between specific domains of negative symptoms and reward processing. Critically, these findings emerged in a sample exhibiting only attenuated negative symptoms and who were not undergoing any type of pharmacological treatment, suggesting that this relationship is not an epiphenomenon of psychiatric illness or medication effects. Additional work is currently underway to examine these relationships using task-based fMRI and may shed light on the neural mechanisms that underlie both the reductions in effort and lack of persistence associated with these symptoms.
Keywords: Negative Symptoms, Reward, Healthy Subjects
Disclosure: Nothing to Disclose.

T191. Effects of Schizophrenia Genetic Risk Variants on Brain Function in the GENUS Consortium Sample Collection

Gabriella Blokland, T GENUS Consortium, Tracey Petryshen*

Harvard Medical School, Massachusetts General Hospital, Boston, Massachusetts, United States

Background: The Genetics of Endophenotypes of Neurofunction to Understand Schizophrenia (GENUS) Consortium aims to understand the genetic mechanisms of impaired brain function underlying schizophrenia. The consortium is examining the influence of genetic variation on cognitive functions and brain structural measures that are abnormal in schizophrenia using a collection of 4,125 patients, 3,644 controls, and 327 familial high-risk participants from 16 international sites.
Methods: Data from each site were transferred to a central site for data harmonization and analyses. Genetic risk variants identified by previous well-powered schizophrenia GWAS, including individual SNPs, gene sets, and polygenic risk, were tested for association with cognitive performance and global/regional brain volume using linear regression.
Results: Analysis of 108 SNPs from the schizophrenia PGC2 GWAS mega-analysis identified significant associations in all GENUS participants between several SNPs and general cognitive ability (Spearman's 'g'), visuospatial ability, and verbal learning/memory (FWE corrected $p < 2.3x10\text{-}5$). Analysis of three gene sets identified by the PGC network and pathway analysis (synapse, histone methylation, and immune and neuronal/neurotrophic gene sets) detected nominal association between the synapse gene set and non-verbal learning/memory ($p < 0.001$). The SNPs and gene set each explain 0.5-1% of the variance in the associated cognitive functions. Nonsignificant genetic associations with brain volume measures are in line with previous studies, suggesting a weaker relationship between schizophrenia genetic risk and brain structure, or may be due to reduced sample power compared to the analyses of cognitive functions. Schizophrenia polygenic risk was associated with worse performance in several cognitive functions (verbal learning/memory, nonverbal working memory, visuospatial ability), as well as reduced gray matter volume of frontotemporal regions (inferior frontal, superior temporal gyrus) ($p < 0.0001$; 1-1.5% phenotypic variance explained).
Conclusions: These results suggest that schizophrenia polygenic risk may be associated with altered development of frontotemporal regions that mediate cognitive functions and psychotic symptoms central to schizophrenia. Overall, these results support the hypothesis that schizophrenia genetic risk influences brain abnormalities underlying the disorder.
Keywords: GWAS, Psychosis, Cognition, Structural Neuroimaging
Disclosure: Nothing to Disclose.

T192. Electrophysiological Assessment of Reward Processing to Near-Miss Events in Schizophrenia

Susanna Fryer*, Brian Roach, Judith Ford, Alison Boos, Daniel Mathalon

University of California, San Francisco, San Francisco, California, United States

Background: Alterations in the way the brain processes rewards are thought to contribute to negative symptom manifestation in schizophrenia, particularly anhedonia and amotivation. To date, most neuroimaging studies of reward processing in schizophrenia have relied on tasks that require response planning and execution to obtain rewards. However, less is known about the extent to which reward processing deficits endure in schizophrenia in the absence of response demands. We therefore used a slot machine reward-based paradigm to assess brain functioning during passive anticipation and receipt of monetary rewards in patients with schizophrenia ($N = 37$) in comparison to demographically-matched healthy controls ($N = 32$).
Methods: EEG was recorded while participants completed 288 trials of a slot machine task. To build expectancy, the display consisted of 3 slot reel positions each of which was initially blank and sequentially populated with fruit symbols from left to right. Reels were populated with one of 12 possible fruit symbols distributed equally among possible

outcomes, such that individual fruit symbols carried no predictive information about likelihood of a rewarding outcome. Trial initiation was self-paced via participant button press, after which timing of the slot reels was automated. The participant's button press triggered an audible animated coin drop and lever press to increase the face validity of the paradigm. The spin phase (reward anticipation phase) consisted of S1, S2, and S3 populating their respective slot reels each with a single fruit symbol. After the third reel populated, the outcome phase began (reward evaluation phase) and a 40Hz visual checkerboard boarder flicker appeared for 1000 ms duration followed by outcome text that depicted "WIN $1.25" or "LOSE," depending on the trial type. There were 3 types of trials: Wins ($P = .25$); Near Misses ($P = .25$); Total Misses ($P = .50$). Wins occurred when 3 identical fruit symbols were populated in the slot reels (AAA); Near Misses occurred when the first and second reel symbols were identical but the third reel symbol was incongruent (AAB). Total Misses occurred when S1 and S2 were incongruent (ABC, ABB, ABA). Near Miss and Total Miss trials were $0 payouts while Win Trials yielded a $1.25 payout. Event-related potentials reflecting reward anticipation (stimulus preceding negativity: SPN) and outcome evaluation (reward positivity: RewP) were assessed for task condition and group differences.

Results: Within the HC group, expected significant effects of condition were observed for the RewP, comparing amplitudes of Win to Near Miss trials ($p < .001$) and for the SPN comparing amplitudes of Win to Total Miss trials ($p = .02$). There were no significant group (HC vs. SZ) by trial type differences in either SPN or RewP amplitudes ($p > .05$).

Conclusions: Significant main effects of condition were observed for both reward anticipation and evaluation task manipulations indicating that the paradigm operated as expected. Preliminary group analyses indicated equivalent SPN and RewP amplitudes across groups, suggesting that, in the absence of response demands, these particular electrophysiological markers of anticipatory and evaluative aspects of reward processing are intact in patients with schizophrenia. These findings are consistent with models that predict the pathophysiology of reward processing anomalies in schizophrenia stem from higher-order cognitive processes required to use motivational information to guide behavior toward reward attainment. In contrast, more basic subcomponents of reward processing may be an area of preservation.

Keywords: Reward Processing, Anthranilic Acid; Kynureninc Acid; Schizophrenia; Parkinson's Disorder, EEG/ERP Electrophysiology

Disclosure: Nothing to Disclose.

T193. Decreased Muscarinic M1 Receptors in the Cortex, Striatum, and Hippocampus of Subjects With Schizophrenia: An Autoradiographic Study

Shaun Hopper*, Geoffrey Pavey, Madhara Udawela, Andrea Gogos, Brian Dean

Florey Institute for Neuroscience and Mental Health, Melbourne, Australia

Background: Positive allosteric modulators (PAM) of muscarinic M1 receptors (CHRM1) represent a promising potential treatment for schizophrenia (Conn et al, 2009). However, one possible limitation to the use of these drugs might be a lack of responsiveness in a sub-group of subjects with schizophrenia, termed muscarinic receptor-deficit schizophrenia (MRDS), who are defined by a marked loss of cortical muscarinic M1 receptors (Scarr et al, 2009). Having developed a novel methodology for measuring the potential efficacy of the CHRM1 PAM, BQCA, in human cortex (Dean et al, 2016), we reported a decreased responsiveness to that drug in cortical tissue from subjects with MRDS. Our methodology required homogenate membrane preparations which does not readily allow the study of small, discrete, cytoarchitectural regions such as are present in the hippocampus. This methodological deficiency is important as we have previously hypothesized that decreased [3H]pirenzepine binding in the hippocampus of subjects with schizophrenia is due to decreased CHRM4 rather than CHRM1 (Scarr et al, 2007). We therefore hypothesized that, unlike in the cortex, BQCA responsiveness would not be altered in the hippocampus of subjects with MRDS. To test this hypothesis, we adapted our novel assay of CHRM1 availability to an autoradiography method to allow measurement of both spatial localization and CHRM1 availability.

Methods: This study was conducted using sections (20μm) of hippocampus, striatum, and cortical region, BA6, from 40 subjects with schizophrenia, and 20 age- and sex-matched non-psychiatric control subjects. The 40 subjects with schizophrenia could be further-divided into 20 MRDS and 20 non-MRDS, as defined by their BA9 [3H]pirenzepine binding. We measured the binding of [3H]n-methylscopolamine ([3H]NMS; 0.4nM) in the absence (total binding), and presence (non-specific binding) of ACh (10mM); specific binding was taken to be the difference between total and non-specific binding. We also measured [3H]NMS (0.4nM) binding partially displaced by ACh (1mM) in the presence or absence of BQCA (3μM); the difference between these is the BQCA effect (measure of CHRM1 availability). All radioligand binding was incubated for 2 hours at room temperature and stopped by rapid dilution and cooling.

Results: Radioligand binding distribution was uniform across all cortical layers of BA6 and the striatum, and close to background in white matter. Specific binding of [3H]NMS and the BQCA effect were significantly lower in schizophrenia in BA6 ([3H]NMS: $p < 0.001$; BQCA: $p < 0.001$) and the striatum ([3H]NMS: $p < 0.01$; BQCA: $p < 0.05$). When analysing the data as three groups, ANOVA revealed significant variations in both measures in BA6 ([3H]NMS: $p < 0.0001$; BQCA: $F_{2,56} = 19.36$; $p < 0.0001$) and the striatum ([3H]NMS: $p < 0.0001$; BQCA: $p < 0.0008$). Post-hoc analysis showed that both measures were lower in MRDS ($p < 0.001$ for all four statistical tests), but not non-MRDS, compared to controls.

Within the hippocampus, [3H]NMS binding was lower in schizophrenia in all sub-fields (dentate gyrus (molecular and granular layers (M/G) and polymorphic layer (PL)); cornu ammonis (CA) 1-3 (alveus layer through to the pyramidal layer (a-p), lacunosum moleculare and stratum radiatum (lm-r)); and the subiculum (polymorphic and pyramidal layers (PP) and molecular layer (ML)); $p < 0.01$ for all regions). By contrast the BQCA effect was significantly lower only in M/G dentate gyrus ($p < 0.05$) and both layers of the subiculum (PP: $p < 0.05$; ML: $p < 0.05$). When split into

MRDS and non-MRDS, both [3H]NMS binding ($p < 0.01$ for all regions) and the BQCA effect ($p < 0.05$ for all regions) varied in all sub-fields due to lower [3H]NMS binding and a lower BQCA effect in the MRDS ($p < 0.05$ for regions), but not non-MRDS, when compared to controls.

Conclusions: Using our autoradiographic methodology, we reproduced our finding of decreased CHRM1 availability in BA6. In addition, our data in the striatum are the first evidence to suggest a loss of CHRM1 in that CNS region from subjects with MRDS. We also found decreased CHRM1 availability in the hippocampus from subjects with MRDS; these data suggest that the lower [3H]pirenzepine binding we have reported in the hippocampus of subjects with schizophrenia included, at least as a component, a decrease in CHRM1 in MRDS.

Conn PJ, Christopoulos A, Lindsley CW (2009). Allosteric modulators of GPCRs: a novel approach for the treatment of CNS disorders. Nature Reviews Drug Discovery 8(1): 41-54.

Dean B, Hopper S, Conn PJ, Scarr E (2016). Changes in BQCA Allosteric Modulation of [3H]NMS Binding to Human Cortex within Schizophrenia and by Divalent Cations. Neuropsychopharmacology 41(6): 1620-1628.

Scarr E, Cowie TF, Kanellakis S, Sundram S, Pantelis C, Dean B (2009). Decreased cortical muscarinic receptors define a subgroup of subjects with schizophrenia. Molecular Psychiatry 14(11): 1017-1023.

Scarr E, Sundram S, Keriakous D, Dean B (2007). Altered hippocampal muscarinic M4, but not M1, receptor expression from subjects with schizophrenia. Biological Psychiatry 61(10): 1161-1170.

Keywords: Post-Mortem, Muscarinic Acetylcholine Receptor, Schizophrenia Subtypes, Positive Allosteric Modulators, Autoradiography

Disclosure: Nothing to Disclose.

T194. Neuromodulation by Combined Oxytocin and Naturalistic Social Cognition

Prantik Kundu*, Derek Alexander Smith, Amanda Fisher, Sarah Rutter, Scott Moeller, M. Mercedes Perez-Rodriguez

Icahn School of Medicine at Mount Sinai, New York, New York, United States

Background: Oxytocin (OXT) is a neuropeptide hormone that may promote social cognition. For instance, in schizophrenia spectrum disorders such schizotypal personal disorder (SPD), OXT increases attention to social cues and modulates social gaze. However, the effects of OXT are context-dependent, individually variable, and may occur via several neural mechanisms. To evaluate this variability, pharmaco-functional MRI (phfMRI) with OXT has been used in task-based designs, revealing modulation of amygdala activity. Recently, resting-state phfMRI has also shown that OXT administration in women increases connectivity between striatal and insula networks. Naturalistic experimental contexts such as narratives and movies may activate a wider array of OXT-ergic neural systems than simple tasks or baseline. We present pilot results from a phfMRI experiment during naturalistic viewing of the Movie for the Assessment of Social Cognition (MASC) after treating participants with OXT or placebo in a randomized control trial. Our hypothesis is that modulation of social gaze by OXT correlates to changes in functional connectivity in the regions of OXT receptor expression, specifically amygdala and nucleus accumbens (NAcc).

Methods: This study was approved the Institutional Review Board of the Icahn School of Medicine at Mount Sinai. Results from an unmedicated patient with SPD are presented here (male, age 34), involving a placebo and drug condition, separated by a 2-week washout period. In a double-blind trial design, 40 IU of OXT or placebo were administered intranasally via atomizer at the beginning of the experiment. Brain imaging began immediately after OXT administration. Structural (T1-weighted) scans were done during an uptake period of approximately 25 minutes. Then, 8 minutes of resting state-fMRI with fixation on a cross-hair was acquired. Next, the full MASC video was presented during fMRI, elapsing approximately 18 minutes. Briefly, the MASC is a live-action movie with four actors portraying characters in social settings. Eye-tracking during MASC playback was acquired to evaluate the effects of OXT on social gaze. An SR Research Eyelink 1000 MRI-compatible eye-tracker was used, after calibration. The standard deviation of gaze from the center of faces in the MASC (detected with OpenCV) was used to measure social gaze. A 3 Tesla Siemens Skyra MRI (Erlangen, Germany) was used for imaging with a 32-channel receive-only head coil and body transmit coil. T1-weighted MPRAGE was acquired with 0.9mm isotropic resolution and whole-brain coverage. Multi-echo multi-band functional MRI was acquired with parameters: 3.0mm isotropic resolution, TR = 1.12s, multi-band factor 5, in-plane acceleration factor 2, whole-brain coverage, 3 TEs = 11.,28.46,45.92 ms. Functional and structural MRI data were preprocessed using multi-echo independent components analysis (ME-ICA). This approach directly separates BOLD and non-BOLD signals based on quantitative T2* decay measures without arbitrary preprocessing. Resting state and MASC data were processed equivalently. Functional data were co-registered to anatomical images, which were in turn normalized to MNI standard space. Seed-based connectivity (6mm radius) of the NAcc and amygdala were compared for MASC and resting state data in the drug and placebo conditions.

Results: Eye-tracking and functional connectivity data were compared between sessions. In the placebo condition, the SPD patient showed alert attention to the MASC video, but gaze was away from faces ($p < 0.01$). In the drug condition, gaze was normalized to view faces. Functional connectivity using a right NAcc seed during MASC viewing in the placebo condition showed bilateral NAcc connectivity, but no significant connectivity to cortical areas ($R > 0.5$). In contrast, in the OXT condition, NAcc showed high positive connectivity ($R \sim 0.8$) to ventro-medial prefrontal cortex (vmPFC) and with amygdala ($R \sim 0.7$). Functional connectivity of the amygdala under placebo during the MASC was found to temporal and visual cortex areas, and negative correlation was found to insular cortex and middle frontal gyrus ($R \sim$ -0.6). In the OXT condition, the amygdala showed broadly reduced connectivity, with significant connectivity only to sensory areas. Comparing rs-fMRI connectivity in drug conditions, seed based connectivity with the NAcc under OXT showed substantially increased connectivity (delta-$R \sim 0.4$) to ventromedial prefrontal cortex. Amygdala rs-fMRI connectivity between placebo and drug conditions did not differ significantly.

Conclusions: Results from this pilot study support the hypothesis that the functional connectivity of two key areas expressing OXT receptor, NAcc and amygdala, is modulated by OXT in a context dependent way. MASC viewing under OXT enhanced NAcc connectivity and reduced amygdala connectivity to regions associated with social cognition and theory of mind, especially vmPFC. The change in functional connectivity during OXT may also be linked to the significant enhancement of social gaze in OXT in the SPD patient. Notably, gaze was quantifiable here due to presentation of the MASC with eye-tracking. In our ongoing case-control study of OXT in SPD, we will further evaluate how OXT modulates the functional interaction of NAcc and amygdala to normalize baseline psychopathology of social cognitive domains.

Keywords: Oxytocin, Amygdala, Fear, fMRI, Schizotypy, Social Cognition

Disclosure: Nothing to Disclose.

T195. TNF and IL-6 are Associated With the Deficit Syndrome and Negative Symptoms in Patients With Chronic Schizophrenia

David Goldsmith*, Ebrahim Haroon, Andrew Miller, Gregory P. Strauss, Peter Buckley, Brian Miller

Emory University School of Medicine, Atlanta, Georgia, United States

Background: Increased inflammatory markers have been found in patients with chronic schizophrenia, and have been associated with negative symptoms. The deficit syndrome is a distinct subtype of schizophrenia, characterized by primary and enduring negative symptoms. Only one study (Garcia-Rizo et al., 2012) has compared inflammatory markers in patients with and without deficit schizophrenia. This study found higher concentrations of C-reactive protein (CRP) and interleukin (IL)-6 in the deficit subgroup, who were all newly diagnosed and antipsychotic naïve. Nevertheless, whether a broader array of inflammatory markers are related to deficit schizophrenia and whether antipsychotic treatment may contribute to this relationship in patients more characteristic of the syndrome (i.e. individuals with chronic illness on medication) remains unknown. We thus measured inflammatory markers in patients with and without deficit schizophrenia and controls. We hypothesized that there would be significant increases in inflammatory marker concentrations in patients with deficit schizophrenia and that inflammatory markers would be associated with symptoms that characterize the deficit syndrome.

Methods: Fifty-six patients with schizophrenia and twenty-eight healthy controls were included in the study. Deficit schizophrenia classification was determined using the Proxy for the Deficit Syndrome employing the Positive and Negative Symptom Scale. Inflammatory markers were measured based on a recent meta-analysis (Goldsmith et al., 2016) that found the following inflammatory markers to be elevated in individuals with chronic schizophrenia on antipsychotic medications: interferon (IFN)-gamma, IL--1beta, IL-6, soluble IL-2 receptor (sIL-2R), and tumor necrosis factor (TNF). To examine associations between inflammatory markers and group assignment, multivariate general linear models (GLM) were performed with the inflammatory markers as dependent variables and diagnostic group as a fixed factor. After establishing the overall significance of the model (Roy's Largest Root), the relative significance of each inflammatory marker was examined. In the initial GLM, patients versus controls was the fixed factor. In subsequent models, group assignment was defined as either deficit versus non-deficit patients versus controls or deficit versus non-deficit patients. Stepwise, backward linear regression models were used to determine the relationship between relevant inflammatory markers and symptoms that characterize deficit schizophrenia (blunted affect and alogia), as well as total PANSS negative and positive symptom subscale scores.

Results: Seventeen patients met criteria for deficit schizophrenia. A significant group by inflammatory marker profile interaction was seen for the whole group [$F(5,54) = 2.44$, $p = 0.046$], with TNF [$F(2,57) = 3.51$, $p = 0.036$] and IL-6 [$F(2,57) = 3.89$, $p = 0.026$] accounting for group differences. TNF was significantly higher in patients with deficit schizophrenia compared to both healthy controls and patients without deficit schizophrenia using Least Square Difference (LSD) post hoc analyses (deficit vs. control, $p < 0.005$; deficit vs. non-deficit, $p < 0.01$). IL-6 was also significantly higher in patients with deficit schizophrenia compared to both healthy controls and non-deficit patients in LSD post hoc analyses (deficit vs. control, $p = 0.044$; deficit vs. nondeficit, $p = 0.036$).]. GLM models comparing the two patient groups, including chlorpromazine (CPZ) equivalents and race as covariates, revealed an overall group effect [$F(5,38) = 2.48$, $p = 0.049$], with TNF [$F(1,42) = 6.41$, $p = 0.015$], and IL-6 [$F(1,42) = 6.57$, $p = 0.0214$] showing a significant association with group status, both being higher in the deficit group (both $p < 0.05$). In linear regression models, increases in plasma TNF were significantly associated with increased blunted affect (beta = 0.31, $p = 0.018$) and alogia (beta = 0.30, $p = 0.024$) as well as increased total PANSS Negative scores (beta = 0.31, $p = 0.012$), controlling for CPZ equivalents and race. No associations were seen between TNF and PANSS Positive or General scores. No associations were found with IL-6 concentrations and negative symptoms.

Conclusions: TNF and IL-6 were elevated in patients with deficit schizophrenia compared to nondeficit patients and healthy controls, indicating that these inflammatory markers are related to the deficit subtype in patients with chronic schizophrenia prescribed antipsychotics. TNF was also found to be associated with deficit schizophrenia in multivariate models, while controlling for significant clinical variables, including medication. These findings support the notion that deficit schizophrenia may represent a distinct pathophysiology involving the immune system irrespective of antipsychotic medication and other relevant variables including race, education, and smoking status. Finally, TNF was associated with the severity of negative symptoms on the PANSS, and specifically with items on the PANSS that are included in deficit schizophrenia categorization. The association of increased inflammation and negative symptoms of the disorder suggests that inflammation may represent a novel treatment target in deficit schizophrenia.

Keywords: Schizophrenia, Inflammatory Cytokines, Deficit Syndome, Negative Symptoms

Disclosure: Nothing to Disclose.

T196. Theta-Gamma Coupling in the Prefrontal-Hippocampal System During Performance of a Spatial Working Memory Task

Makoto Tamura*, Timothy Spellman, Andrew Rosen, Joseph Gogos, Joshua Gordon

Mitsubishi Tanabe Pharma Corporation, Yokohama, Japan

Background: Cross-frequency coupling supports the organization of brain rhythms and is present during a range of cognitive functions. However, little is known about whether and how long-range cross-frequency coupling across distant brain regions subserves spatial working memory. Many single neurons in the medial prefrontal cortex (mPFC) are modulated by theta-frequency oscillations in the hippocampus. This theta-frequency synchrony between the hippocampus and mPFC is associated with successful spatial working memory performance. Additionally, recent evidence suggests that gamma-frequency synchrony between the hippocampus and other connected brain regions also contributes to spatial working memory. In this study, the role of cross-frequency coupling in spatial working memory is examined using several approaches.
Methods: We have previously shown that mice carrying a heterozygous deletion of Zdhhc8 (Zdhhc8+/– mice), a gene within the 22q11.2 microdeletion region that contributes to cognitive dysfunction and schizophrenia, demonstrate impaired acquisition of a T-maze delayed non-match-to-place (DNMTP) task. Zdhhc8+/– mice and wild type littermates were implanted with microelectrodes targeting the mPFC, ventral hippocampus (vHPC) and dorsal hippocampus (dHPC). Local field potential (LFP) and multiple single-unit recordings were obtained during performance of the DNMTP task. We have previously demonstrated that performance on a spatial working memory task is impaired by optogenetic inhibition of vHPC-to-mPFC terminals. Specifically, inhibition of these terminals during the sample phase impairs performance on the subsequent choice phase. Here data from the choice phase of correct trials was re-analyzed for theta-gamma coupling strength.
Results: Slow gamma (30-70) Hz oscillations in the mPFC were coupled to theta oscillations both locally and in the vHPC. This theta-gamma coupling was enhanced in Zdhhc8 +/– mice during performance of a DNMTP task. This enhancement was inversely correlated with choice accuracy of the task, and apparent only in choice phases of correct trials. Optogenetic and behavioral manipulations revealed that theta-slow gamma coupling was enhanced with increased task difficulty, regardless of the cause of this increase. Finally, enhanced vHPC theta-mPFC slow gamma coupling drove synchronous firing in mPFC neurons, suggesting that theta-slow gamma coupling can improve behavioral performance by modulating activity and synchrony within the mPFC.
Conclusions: This study suggests that increases in vHPC theta-mPFC slow gamma synchrony underlie successful behavioral performance in the setting of increases in task difficulty, reflecting a potential circuit-based compensatory mechanism important for effective spatial working memory behavior.

Keywords: Functional Connectivity, Systems Neurobiology, Prefrontal Circuit, Schizophrenia, 22q11.2 CNV
Disclosure: Part 5: Mitsubishi Tanabe Pharma Corporation, Employee.

T197. Dietary Omega 3 and Erythrocyte Omega 3 are Associated With Symptoms, Functioning and Psychotic Conversion in a Clinical High Risk Population

Kristin Cadenhead*, Jean Addington, Carrie Bearden, Ty Cannon, Barbara Cornblatt, Daniel Mathalon, Thomas McGlashan, Diana Perkins, Larry Seidman, Elaine Walker, Scott Woods, Jeff Yao

University of California, San Diego, La Jolla, California, United States

Background: Omega-3 Fatty Acids (FAs), EPA (eicosapentaenoic acid) and DHA (Docosahexaenoic acid), are essential for normal brain development and may also have neuroprotective properties. Abnormal FA metabolism may play a role in the etiology of psychiatric illness. Reductions in Red Blood Cell (RBC) membrane Polyunsaturated Fatty Acids (PUFAs) have been reported in both chronic patients and unmedicated first episode patients with schizophrenia. Dietary supplementation of EPA and DHA may have beneficial effects in both somatic and mental illness. Studies of Omega 3 supplementation in Clinical High Risk (CHR) populations have had mixed results. One study (Amminger et al) found reduced conversion to psychosis along with improvement in symptoms and functioning in the Omega 3 versus the Placebo group while two studies (McGorry et al and Cadenhead et al unpublished) have failed to replicate these findings. In the current study, we assessed the relationship of dietary Omega 3 and RBC PUFA levels to symptoms, functioning and conversion to psychosis in a sample of CHR subjects participating in a trial of Omega 3 FA versus Placebo.
Methods: As part of a 24-week, randomized, double-blind, placebo, fixed dose-controlled study of Omega-3FA versus placebo in 127 CHR subjects, baseline diet was assessed using a systematic checklist of Omega-3FA foods and fasting RBC PUFA composition was assessed. Regular assessments of symptoms and functioning were performed throughout the study. We investigated the relationship between reported Omega 3 FA consumption, RBC PUFA levels and outcome measures as part of this trial.
Results: Of the 127 CHR subjects recruited into the trial, 118 completed baseline assessment and 70 completed the 6 month trial. Ten percent of subjects converted to psychosis during the 24 months. The rate of psychotic conversion did not differ in the Omega-3FA (13%) versus Placebo (8%) samples. However, conversion to psychosis was predicted by lower levels of EPA (24-month conversion 16.2% low EPA vs 2.6% high EPA) in the RBC membrane (Wald Statistic 3.72, $p<0.05$). Greater levels of negative symptoms and low GAF scores were associated with a diet consisting of few Omega 3 fatty acid rich foods and corresponding low EPA in the RBC membrane. The self-reported Omega 3 fatty acids in the diet were significantly correlated with a number of the RBC PUFA composition variables and measures of oxidative stress.
Conclusions: The finding of a significant association between baseline diet low in Omega-3FA rich foods and

outcome raises the question of whether it is possible to influence both physical and mental health with lifestyle choices including diet. These findings reinforce that early detection of food insecurity is crucial since this factor is modifiable with the potential for significant gains in terms of quality of life, physical and mental health. Longitudinal follow-up will provide insight into whether these indices are risk factors for future psychosis and functional outcome.
Keywords: Omega-3 Fatty Acid, Clinical High Risk, First Episode Psychosis
Disclosure: Nothing to Disclose.

T198. Development of an Olfactory 5-Choice Continuous Performance Task for Mice: Elevated Motoric Impulsivity With MK-801

David MacQueen*, Vishal Bobba, Jared Young

University of California, San Diego, La Jolla, California, United States

Background: Cognitive deficits are a hallmark feature of numerous psychiatric disorders, often appear during adolescence (prior to diagnosis), and contribute to global functional outcome. Tasks that can quantify these cognitive domains across species are required to determine the neurobiological substrates of such deficits and evaluate putative therapeutics. At present, cognitive tasks for rodents often require extensive training periods and provide limited comparability to human cognitive testing. Leveraging the robust olfactory acuity of rodents (their preferred sensory modality as vision is to humans), we developed an odor-based 5-choice continuous performance task (o5C-CPT) for mice which can be rapidly trained (10 days) yielding high levels of engagement and accuracy.
Methods: In fully automated olfactometers, 13 male C57/BL6 mice were trained to stability on a procedure in which subjects initiate trials by entering their head into an odor sampling port. On each trial, 1 of 6 odors was presented and responses on a lick port were recorded. For each subject, 1 of 6 odors was randomly selected as a non-target odor. Responses to target odors (5/6) yielded delivery of sucrose solution, while responses to non-target odors were punished with a time-out. After reaching stability, the effects of the NMDA antagonist MK-801 (0.10, 0.17, & 0.30 mg/kg) on performance was assessed in a within-subjects design.
Results: MK-801 significantly slowed the latency of hit responses [$F(3,36) = 6.1$, $p = 0.002$] and decreased both trials completed [$F(3,36) = 8.6$, $p = 0.002$], and reinforcers collected [$F(3,36) = 9.0$, $p < 0.001$], with differences apparent at the highest dose relative to placebo. Trials in which the subject exited the sampling port before odor presentation (short samples) were increased [$F(3,36) = 19.973$, $p < 0.001$] at all doses relative to placebo. A trend towards decreased accuracy on target trials (miss rate) was observed [$F(3,36) = 2.9$, $p = 0.102$), but no effect on overall sensitivity was detected [$F(3,36) < 1$, $p = 0.425$).
Conclusions: The present study demonstrates the feasibility of the o5C-CPT which can be rapidly trained and is sensitive to drug effects on multiple aspects of performance. MK-801 elevated motoric impulsivity (short sampling) while high doses slowed overall performance. Refinement of training procedures will enable a direct comparison of drug effects with the well-established visual based 5C-CPT paradigm. Such speed of training will enable testing to be conducted in rodents during adolescence, a previously ponderous challenge in operant tasks requiring extended training periods.
Keywords: olfactory, Attention, Impulsivity, NMDA Antagonists, 5C-CPT
Disclosure: Nothing to Disclose.

T199. History of Cannabis Use is Associated With Greater Impairment in Neurophysiological, Clinical and Functional Measures in Schizophrenia Patients

Juan Molina*, Neal R. Swerdlow, Yash B. Joshi, Michael L. Thomas, Joyce Sprock, Monica E. Calkins, Michael F. Green, Tiffany A. Greenwood, Raquel E. Gur, Ruben C. Gur, Laura C. Lazzeroni, Keith H. Nuechterlein, Allen D. Radant, Larry J. Seidman, Larry J. Siever, Jeremy M. Silverman, William S. Stone, Catherine A. Sugar, Debby W. Tsuang, Ming T. Tsuang, Bruce I. Turetsky, David L. Braff, Gregory A. Light

University of California, San Diego, San Diego, California, United States

Background: Cannabis use is common in patients with schizophrenia, but the impact of a history of cannabis use on neurophysiologic, clinical, and psychosocial domains is not well understood. Chronic cannabis use has been associated with persistent changes to neurophysiologic functioning, including deficits in mismatch negativity (MMN) and sensory gating in otherwise healthy individuals. In schizophrenia, robust deficits in MMN and P3a amplitude have been demonstrated. However, one study reported that chronic cannabis use was associated with increased MMN in schizophrenia patients. Therefore, we sought to clarify the relationship between a history of cannabis use and neurophysiological measures of early auditory information processing – believed to mediate functional outcome – as well as measures of clinical symptoms and psychosocial functioning in schizophrenia. We analyzed data from a well-characterized case-control sample of schizophrenia patients and healthy subjects, collected by the Consortium on the Genetics of Schizophrenia (COGS).
Methods: Schizophrenia patients ($N = 901$) who had data available on neurophysiologic measures of early auditory information processing (MMN, P3a), symptomatology (Scales of Positive and Negative Symptoms (SANS, SAPS)), and function (Global Assessment of Function Scale (GAF), Scale of Functioning (SOF)) were queried from the larger COGS-2 database. After excluding patients with other substance use disorders, the remaining sample was stratified based on a history of a cannabis use disorder ($n = 74$) without history of other substance use disorders, and a comparison group of schizophrenia patients without a history of substance abuse ($n = 265$). Subjects were excluded from COGS-2 if they met criteria for substance abuse within 30 days of testing, dependence within 6 months of testing, or screened positive on urine toxicology.
Results: Relative to patients with no history of substance use disorders, patients with a history of a cannabis use disorder showed greater impairment, i.e. less negativity, in MMN

($F = 6.3$, $p < 0.015$) and reduced P3a amplitude ($F = 3.9$, $p < 0.05$). Patients with a history of a cannabis use disorder also had greater levels of negative (SANS, $F = 11.4$, $p = 0.001$) and positive symptoms (SAPS, $F = 11.0$, $p = 0.001$) compared to patients with no substance abuse history. Similarly, past cannabis use disorder was associated with worse psychosocial functioning (GAF, $F = 10.5$, $p = 0.001$; SOF, $F = 8.8$, $p = 0.003$).

Conclusions: Among COGS subjects with a chronic schizophrenia diagnosis, a history of cannabis use disorder was associated with greater deficits in MMN and P3a amplitude. Past cannabis use disorder was also associated with greater clinical severity and worse functional outcome in these patients. These findings suggest that biological factors leading to cannabis abuse, or other factors associated with that abuse (including, for example, the modulation of endocannabinoid systems), may have persistent effects on neurophysiological measures of early auditory information processing and on clinical domains. Future prospective studies are needed to establish how history of cannabis use contributes to the variance in measures of neurophysiologic performance and impacts clinical and functional outcome in schizophrenia.

Keywords: Cannabinoids, NMDA Receptors, MMN, ERP, Auditory, Training, Early Auditory Information Processing, Relevance for Outcomes

Disclosure: Nothing to Disclose.

T200. Elucidation of mGlu1 Receptor Neurophysiology Through Use of Allosteric Modulators: Implications for the Treatment of Schizophrenia

P. Jeffrey Conn*, Samantha Yohn, Daniel Foster, Daniel Covey, Pedro Garcia-Barrantes, Joseph Cheer, Craig Lindsley

Vanderbilt University, Nashville, Tennessee, United States

Background: Given the shortcomings of current antipsychotics, novel therapeutic approaches are needed that exhibit clinical efficacy with reduced adverse effect liability. Activation of the muscarinic M4 receptor has previously been demonstrated to have antipsychotic-like efficacy in several preclinical models. Recently our laboratory reported that these antipsychotic effects are mediated through a novel mechanism wherein activation of M4 receptors reduces dopamine release through a CB2 cannabinoid-dependent mechanism (Foster et al. 2016). This provides a mechanism which allows selective inhibition of midbrain neurons implicated in psychosis, without inhibiting dopamine release in the cortex or other regions which may lead to adverse effects. However, the underlying neuronal effects by which M4 induce endocannabinoid release is not well understood. The metabotropic glutamate subtype 1 receptor (mGlu1) has been extensively characterized to release endocannabinoids in various brain regions; therefore, we investigated whether mGlu1 mediates M4-induced reductions in striatal DA and subsequent antipsychotic-like effect. In addition to its role in modulating M4 effects, it is conceivable that mGlu1 may also inhibit dopamine release independently of M4. Substantial preclinical and clinical evidence has implicated mGlu1 in the pathophysiology of schizophrenia (Ayoub et al. 2012) and modulation of striatal dopamine release (Zhang and Sultzer, 2003). We hypothesize that selective activation of mGlu1 will reduce striatal DA release and have antipsychotic efficacy in rodent models of psychosis.

Methods: To assess striatal dopamine release, acute coronal brain slices containing the striatum (300mm) were obtained via a variation of a reported brain slice methodology for adult mice (Foster et al. 2014). When monitoring electrically-evoked dopamine transients, stimulating electrodes were placed 75mm deep into the dorsolateral striatum and slices were electrically stimulated (250-500μA) every 2.5 minutes via a bipolar stimulating electrode. The effect of selective mGlu1 activation on striatal dopamine release was conducted in acute coronal brain slices and through in vivo fast scan cyclic voltammetry. A stimulating electrode was implanted into the medial forebrain bundle and a carbon fiber working electrode into the dorsal striatum. Biphasic current pulse (±450μA, 60Hz) were applied for 2s to the dopamine axons to evoke DA release. The regulatory role of mGlu1 on the antipsychotic-efficacy of M4 activation and antipsychotic-effect of selective activation of mGlu1 was assessed using the prepulse inhibition of acoustic startle reflex assay (PPI) and amphetamine-induced hyper-locomotion in 6-week-old C57Bl/6 wild-type mice (Jackson Laboratories).

Results: The results from our experiments are two-fold. First, we found that M4-mediated inhibition of striatal dopamine release and antipsychotic-like effects were inhibited by the highly selective mGlu1 negative allosteric modulator VU0469650. Prior experiments have found that the group 1 agonist DHPG reduces striatal DA release, an effect independent on mGlu5 activation. Through use of selective mGlu1 compounds, we found that mGlu1 is responsible for the effects of an EC80 DHPG concentration on striatal dopamine release and co-application of a mGlu1 PAM can potentiate the effects of EC20 DHPG on striatal DA release. In addition, we found that selective activation of mGlu1 causes a 25% reduction in DA release in vivo and possess antipsychotic-like efficacy in preclinical models of psychosis.

Conclusions: These results highlight the regulatory role of mGlu1 activation in the antipsychotic-like effect of M4 PAMs. Moreover, it suggests that there is a cross communication between mGlu1 and M4 which facilitates endocannabinoid release. Future studies are needed to assess how this is occurring. In addition, these findings demonstrate a novel intervention strategy utilizing mGlu1 selective compounds.

Keywords: Glutamate, Schizophrenia, Dopamine

Disclosure: Part 1: Vanderbilt University, Patent, **Part 4:** Astra Zeneca, Grant.

T201. Schizophrenia Risk Variation in DRD2 and Striatal D2 Receptor Availability in Vivo

Daniel Eisenberg*, Philip Kohn, Catherine Hegarty, Angela Ianni, Michael Gregory, Jasmin Czarapata, Joseph Masdeu, Bhaskar Kolachana, Karen Berman

National Institute of Mental Health, Bethesda, Maryland, United States

Background: Pharmacological treatment of schizophrenia has long relied upon blockade of D2 dopamine receptors, though whether D2 systems are involved in the etiology of

the illness remains an open question. Prior work using positron emission tomography has suggested that individuals with schizophrenia may have elevations in striatal D2 dopamine receptor availability (Kegeles et al, 2010), though not all studies have been corroborative (Kessler et al, 2009), and effects of medication exposure in patients have been difficult to eliminate. In a large genome-wide association study, the Schizophrenia Working Group of the Psychiatric Genomics Consortium (2014) identified rs2514218, a single nucleotide polymorphism (SNP) upstream of DRD2, as significantly linked to schizophrenia risk. Subsequent reports have found possible effects of this SNP on striatal activation measured with functional magnetic resonance imaging (Vink et al, 2016) and on clinical response to antipsychotic treatment (Huang et al, 2016; Zhang et al, 2015), which were attributed to D2 receptor mechanisms. However, it is unknown whether this variant in fact affects D2 receptor function in the living human brain.

Methods: Seventy-four healthy individuals (mean age 36.1 +9.7, 36 female) genotyped for rs2514218 using TaqMan assay underwent positron emission tomography (PET) to measure D2/3 receptor availability. Caffeine and nicotine were restricted for 4 hours prior to imaging sessions. Images of the brain were collected on a Siemens HRRT PET scanner in list mode in three segments over four hours immediately following intravenous injection of [18F]-fallypride. Continuous optical head position tracking throughout the scan session was used for motion correction, and transmission images were acquired for each imaging segment to correct for attenuation. A separately acquired T1-weighted structural MRI scan was used to define hand-edited striatal (bilateral caudate, putamen, and ventral striatum) regions of interest and a cerebellar reference region for each individual, as well as to guide spatial normalization of the dynamically binned, corrected PET frames for use in voxelwise analyses within the striatum. A simplified reference tissue model (Lammertsma and Hume, 1996) was used to calculate tracer binding potential (BPND). Between-groups analysis was carried out with general linear modeling in order to test for effects of risk allele homozygosity, controlling for age.

Results: Genotyping revealed 31 risk homozygotes and 43 minor allele carriers with allelic frequencies found to be in Hardy Weinberg equilibrium. Multivariate testing identified a significant effect of genotype on BPND ($p = 0.037$) across all striatal regions. Post-hoc univariate tests identified caudate ($p = 0.020$) and putamen ($p = 0.042$) but not ventral striatal effects. Voxelwise analyses further localized these effects to the dorsal caudate head ($p < 0.05$, FWE corrected), with minor allele carriers showing greater BPND.

Conclusions: These results in healthy controls suggest that the GWAS-implicated DRD2 schizophrenia risk SNP rs2514218 may be associated with differential D2 dopamine receptor availability in the living human brain, particularly in the caudate nuclei, where prior work using [18F]-fallypride has identified abnormalities in schizophrenia (Kegeles et al, 2010). Although these data provide novel evidence of cis-acting functionality for this statistically identified genetic marker, the direction of the findings does not align with the expected risk phenotype, indicating that putative mechanisms underlying this variant's reported illness association may be complex and merit further investigation.

Huang E et al (2016). Pharmacogenomics 17(2): 103-109.
Kegeles LS et al (2010). Biol Psychiatry 68(7): 634-641.
Kessler RM et al (2009). Biol Psychiatry 65(12): 1024-1031.
Lammertsma AA, Hume SP (1996). Neuroimage 4(3.1): 153-158.
Schizophrenia Working Group of the Psychiatric Genomics Consortium (2014). Nature 511(7510): 421-427.
Vink M et al (2016). Schizophr Bull 42(3): 843-850.
Zhang JP et al (2015). Schizophr Bull 41(6): 1248-1255.

Keywords: Dopamine (D2, D3) Receptors, Schizophrenia, Genetics

Disclosure: Nothing to Disclose.

T202. Genomic Dissection of Bipolar Disorder and Schizophrenia Including 28 Subphenotypes

Douglas Ruderfer*, PGC Bipolar Disorder and Schizophrenia Working Group, Pamela Sklar, Kenneth Kendler

Vanderbilt University Medical Center, Nashville, Tennessee, United States

Background: Schizophrenia (SCZ) and bipolar disorder (BD) are highly heritable disorders (h2 ~ 0.8) that share a significant proportion of common genetic factors, which is evidenced from both family and recent large-scale genetic studies (genetic correlation (rg) ~ 0.6). Understanding the genetic factors underlying the specific symptoms of these disorders will be crucial for improving diagnosis, intervention and treatment. Our previous work demonstrated that a polygenic score could significantly distinguish these two disorders. However, no particular locus reached genome-wide significance in a direct case-case genome-wide association (GWA) study. Since that study, sample size and number of subphenotypes have greatly increased, providing improved power to identify disease and subphenotype specific genomic factors.

Methods: In case-control data consisting of 52,555 cases (20,129 BD, 32,541 SCZ) and 54,065 controls we performed GWA on a combined phenotype (BD+SCZ vs controls), disease specific phenotypes and a comparison phenotype (SCZ vs BD) across ~15 million imputed variants controlling for site, chip and ancestry. We incorporated transcriptional data from both blood and brain to identify significantly associated regulatory variants using Summary-data-based Mendelian Randomization (SMR). Disease specific genomic regions were assessed using a regional joint association approach and regional heritability estimation. Finally, we tested if there was significant correlation between 28 subphenotypes (4 in SCZ, 24 in BD) and estimated polygenic risk scores from both BD and SCZ.

Results: We identified 114 genome-wide significant loci (GWS) when combining all cases vs controls of which 37 represented novel findings. Two genome-wide significant loci were identified when comparing SCZ vs BD with a third locus discovered when including expression information using SMR. We performed regional joint association and identified a genomic region of overlapping association in BD and SCZ, which was, however, contributed from independent variants. Regional heritability analyses demonstrated that the estimated heritability of BD based on the SCZ GWS

regions was significantly higher than that based on the average genomic region ($N = 82$, p = 1.1x10-6). However, the heritability of SCZ based on the BD GWS regions was not significantly different from that based on the average ($N = 10$, p = 0.56). We identified several significant correlations between BD and SCZ polygenic risk scores: 1) SCZ subphenotypes: negative symptoms (SCZ, 3.6x10-6) and manic symptoms (BD, $p = 2$x10-5), 2) BD subphenotypes: psychotic features (SCZ 1.2x10-10, BD 5.3x10-5) and age of onset (SCZ 7.9x10-4). Finally, we show that psychotic features in BD has significant heritability (h2 = 0.15, SE = 0.06), significant genetic correlation with SCZ (rg = 0.34) and significant sign test result between SCZ GWAS and a GWAS of BD with and without psychotic features ($p = 0.0038$, one-side binomial test).

Conclusions: For the first time, we have identified specific loci pointing to a potential role of 4 genes (DARS2, ARFGEF2, DCAKD and GATAD2A) that distinguish between BD and SCZ providing an opportunity to understand the biology contributing to differing symptoms of these disorders. Our results suggest a direction of genomic sharing with SCZ GWS loci contributing to BD but not the other way around. Finally, we present polygenic analyses of 28 different subphenotypes demonstrating underlying genomic components contributing to 1) several symptom dimensions including psychotic features in BD, negative and manic symptoms in SCZ as well as to 2) measures of severity including number of hospitalizations and age of onset. We further show that existence of psychotic features in BD has a strong genetic component shared with SCZ for both genome-wide and at the most significant SCZ loci. In conclusion, our results provide the best evidence so far of genomic components divergent between BD and SCZ contributing directly to specific symptom dimensions.

Keywords: Schizophrenia, Bipolar Disorder, Genetics, Genomics, Subphenotypes

Disclosure: Nothing to Disclose.

T203. Mapping the 22q11.2 Locus and Risk for Psychosis and Autism: An Integrative Functional Genomics Approach

Jennifer Forsyth*, Daniel Nachun, Michael Gandal, Giovanni Coppola, Carrie Bearden

University of California, Los Angeles, Los Angeles, California, United States

Background: 22q11.2 deletion syndrome (22q11DS) is a recurrent copy number variant (CNV) that occurs in ~1 in 4000 live births and involves the deletion of 1.5 – 3 megabases (Mb) on the long arm of chromosome 22. 22q11DS is the greatest known single genetic risk factor for psychosis and also confers significantly heightened risk for autism spectrum disorder (ASD). As the typical deletion spans ~50 protein coding genes, it remains unclear which genes within the 22q11.2 locus confer risk for these disorders. Notably, large scale genomic studies have begun to unravel the genetic architecture of schizophrenia and ASD, with both disorders showing a complex and highly polygenic architecture that may converge on partially overlapping and partially distinct biological pathways.

Methods: To clarify the functional pathways conferring risk for schizophrenia and ASD, and potential genes within the 22q11.2 locus that might contribute risk for these disorders, we: 1) utilized BrainSpan whole-genome transcriptomic data (http://www.brainspan.org/) to identify modules of co-expressed genes and corresponding biological pathways enriched for schizophrenia and ASD genetic risk; and 2) mapped 22q11.2 genes across these modules to identify genes likely to confer risk for each disorder. Modules of co-expressed genes were identified from BrainSpan samples spanning the full period of human brain development (i.e. early prenatal through 30 years of age) using weighted gene co-expression network analysis; common and rare genetic variants associated with schizophrenia and ASD were derived from the literature. Module enrichment for polygenic risk was tested using MAGMA when single nucleotide variant level summary statistics were available; otherwise, hypergeometric overlap testing was used.

Results: Polygenic risk for schizophrenia loaded highly onto a neuronally-enriched module (M5) involved in excitatory synaptic transmission and synaptic plasticity that increased steadily in expression through development until adolescence, as well as onto an early prenatal module (M2) involved in gene expression, RNA processing, and chromatin modification. Genetic risk for ASD was enriched within these same two modules, as well as within two additional modules. One of the additional ASD-enriched modules (M3) was also involved in early chromatin modification, RNA processing, and gene expression regulation, whereas the other unique ASD-associated module (M7) was a neuronally-enriched module that showed peak expression during mid-late prenatal development and was involved in brain development, neuronal migration and axon guidance, and synapse formation. Of 36 genes within the 22q11.2 locus expressed in the BrainSpan data, 7 genes were found in each of the 2 modules enriched for both schizophrenia and ASD genetic risk (i.e. M5 and M2). Top 22q11.2 genes in the neuronally-enriched M5 module, ranked according to scaled connectivity scores, included PI4KA, a catalytic enzyme involved in the regulation of signal transduction and synaptic transmission, SEPT5, a cytoskeletal protein involved in axon growth and vesicle targeting, and SLC7A4. Top 22q11.2 genes in M2 included UFD1L, a key component of the ubiquitinated protein degradation pathway involved in controlling protein stability, activity, and synapse localization, SNAP29, a syntaxin-binding protein that modulates the presynaptic machinery involved in vesicle-membrane fusion, and HIRA, a histone chaperone and epigenetic regulator found to impact neural progenitor cell proliferation and differentiation. Seven additional 22q11.2 genes were found in the ASD-only associated module M3; top 22q11.2 genes within this module included ZNF74, a zinc finger protein involved in transcriptional and post-transcriptional regulation of gene expression, and DGCR8, a key component of the microRNA microprocessor complex involved in regulating the translation of messenger RNA to proteins.

Conclusions: These results suggest that 22q11.2 genes contribute to risk for schizophrenia and ASD through mechanisms that overlap with other common and rare variants conferring risk for these disorders, and highlight potential genes within the 22q11.2 locus that may be most critical for conferring risk for these neurodevelopmental disorders.

Keywords: Functional Genomics, Schizophrenia, Autism Spectrum Disorders, 22q11.2 CNV
Disclosure: Nothing to Disclose.

T204. Mitochondrial Complex I Deficiency in Schizophrenia and Bipolar Disorder and Medication Influence

Marquis Vawter*, Brandi Rollins, Ling Morgan, Brooke Hjelm, Adolfo Sequeira, Richard Myers, Jack Barchas, Francis Lee, Alan Schatzberg, Huda Akil, Stanley Watson, William Bunney

University of California, Irvine, Irvine, California, United States

Background: Previous reports of mitochondrial dysfunction in the brain from subjects with schizophrenia (SZ) and bipolar disorder (BD) have shown decreased protein and transcript levels for mitochondrial genes, primarily those related to Complex I, the first protein assembly of the electron transport chain. However, in vitro results have suggested the antipsychotic and antidepressant drugs used to treat these psychiatric disorders may be fully or partially responsible for these mitochondrial dysfunctions.
Methods: We measured mitochondrial Complex I activity in postmortem brain tissue in parallel with measures of antipsychotic and antidepressant medications to gain a clearer understanding of these observed alterations. We additionally measured mitochondria DNA (mtDNA) copy number and the heteroplasmy rate of the 4,977 bp "common deletion" in the same samples. The aggregated protein concentration of Complex I was evaluated in brain tissue homogenate from the dorsolateral prefrontal cortex (DLPFC) of three groups (SZ, BD, and controls) and was used to standardize concentrations in the subsequent Complex I activity assay. Finally, regression analyses were performed to test for the effects of disease and psychiatric medications on Complex I activity while controlling for important covariates such as age, sex, and pH.
Results: Complex I activity was significantly decreased by 45% in SZ compared to controls ($p = 0.02$) after adjusting for relevant covariates while no significant difference was found in BD. Additionally, two analyses of case only (SZ and BD) were conducted to evaluate the effect of drugs and age at disease onset. First, Complex I activity was significantly decreased ($p = 0.01$) in pooled cases that had detectable psychotropic medications and drugs compared to pooled cases that did not have detectable levels in toxicological assays. Second, subjects with an age of onset in teens (with no detectable psychotropic medications) showed significantly decreased ($p < 0.05$) Complex I activity compared to subjects with an adult age of onset. Additionally, both SZ and BD groups displayed significant increases ($p < 0.05$) of mtDNA copy number in the DLPFC compared to controls. Despite the observed differences in mtDNA copy number, the mitochondria common deletion burden was not significantly altered in SZ or BD, but this metric did have a significant and positive correlation with age in the brain ($p < 0.01$).
Conclusions: The present study suggests that in chronic patients with SZ and BD there is a significant reduction in mitochondrial activity particularly in subjects that had taken recent antipsychotic or antidepressant medications. One proxy indicator of illness severity, age at onset, showed that subjects with a younger age of onset showed more reduction in Complex I activity than adult onset cases. Present data show an increase in copies of mtDNA in SZ and BD, a surrogate for the actual numbers of mitochondria in the brain. These results support prior studies showing that antipsychotic or antidepressant treatments can lead to mitochondrial hypofunction and suggest that drugs which target improvement in mitochondrial function may be a beneficial therapeutic strategy for some SZ and BD patients with an early age of onset. The negative toxicology results of postmortem brain is an indicator of the absence of acute treatment, we cannot conclude that those patients were never exposed to psychiatric medications. Further studies of medication-free first-episode psychosis patients are needed to elucidate whether mitochondrial pathophysiology occurs independently of longer term medication effects in subjects that later develop a psychiatric illness.
Keywords: Mitochondrial Function, mtDNA, Psychotropic Medications, Teen Age Onset, Adult Onset
Disclosure: Nothing to Disclose.

T205. Using Ketamine to Test the Link Between Auditory Evoked Potential Amplitude and the Formation/Decay of Auditory Sensory Memory in Awake Behaving Monkeys

Tobias Teichert*

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: For several seconds, auditory information about past sounds is passively stored in auditory sensory memory. Despite the importance of auditory sensory memory for many aspects of auditory function, its neural mechanisms are still a matter of debate. However, it has been noted that the amplitude of the auditory evoked N1, which is reduced immediately after a tone has been processed, recovers back to baseline at the same rate at which information decays from auditory sensory memory. Reduced N1 amplitude may thus indicate that the previous sound is still encoded in auditory sensory memory. Individuals with schizophrenia (SZ) show decreased performance in simple auditory tasks that have been linked to impaired encoding of information into auditory sensory memory. In addition, SZ show a blunted modulation of the N1 as a function of previous tones. This blunting can be mimicked in healthy controls by systemic injection of sub-anesthetic doses of ketamine. Here we tested the hypothesis that the administration of ketamine in the non-human primate affects both N1 amplitude and sensory memory encoding in parallel, and thus mimics the behavioral/electrophysiological phenotype in SZ.
Methods: We recorded AEPs from 32 cranial EEG electrodes while macaque monkeys performed a novel delayed pitch-discrimination task designed to track the dynamic formation and decay of auditory sensory memory. In the task, animals listened to sequences of standard tones and released a lever when they identified a pitch-deviant target tone. The stimulus-onset asynchrony (SOA) of consecutive tones varied randomly between 0.250 and 12 sec. The target could

occur between sequence positions 2 and 13. The frequency-difference between standard and target (ΔF) varied between 0 and 1.2 octaves. On catch trials (ΔF = 0) animals were rewarded for not releasing the lever. On randomly assigned days animals performed the task either after the injection of a sub-anesthetic dose of ketamine or vehicle.
Results: As expected, target detection rate increased with ΔF. The slope of the corresponding psychometric function was used to quantify discrimination performance as a function of SOA and the number of preceding standards. Preliminary data showed that discrimination performance gradually increased with repetition number, and decreased with SOA. Several AEPs such as the P31 and the N85 –the presumed monkey homolog of the N1– increased with SOA (while discrimination performance decreased). In contrast, AEP amplitudes generally increased with stimulus repetition number (while discrimination performance increased). Both effects shared similar timing and topography. Preliminary data from 2 recording sessions in one animal shows that the task can be performed after the injection of a sub-anesthetic dose of ketamine that was previously shown to substantially blunt the scaling of AEP amplitude with SOA in a passive listening task. In clear contrast, no obvious blunting of AEP amplitude was observed when the tones were played in the context of the delayed pitch-discrimination task. However, ketamine seemed to eliminate the robust increase of N1 amplitude with repetition number that was observed on days with saline injection. The poster will provide detailed comparisons between AEP amplitude and discrimination performance on and off ketamine based on a full data set.
Conclusions: Findings in the saline condition reveal a complex yet systematic relationship between the experimental manipulations (SOA and repetition number), and the two dependent variables: AEP amplitude and pitch discrimination performance. A small set of preliminary recordings suggest that AEP amplitudes are much less affected by ketamine during active pitch discrimination than during passive listening. This preliminary finding raises the interesting question of how attention and/or task engagement counteracts the strong effect of ketamine on AEP amplitude observed during passive listening. Furthermore, the preliminary observation that ketamine abolished the increase of AEP amplitude with repetition number may be related to the role of NMDA receptors in strengthening the memory trace of the standard tone across successive yet temporally isolated presentations. To validate this preliminary interpretation of the AEP data, additional recordings and analyses will need to establish a parallel effect on discrimination performance.
Keywords: Auditory Deficits in Schizophrenia, Ketamine, Rhesus, EEG/ERP Electrophysiology, Auditory Short-Term Memory
Disclosure: Nothing to Disclose.

T206. Uncorrelated PANSS Score Matrix to Reduce Pseudospecificity of PANSS Factors: Application to Baseline Data

Seth Hopkins*, Ajay Ogirala, Antony Loebel, Kenneth Koblan

Sunovion Pharmaceuticals, Inc., Marlborough, Massachusetts, United States

Background: Interpretation of the efficacy of antipsychotic agents in treating schizophrenia using standard (Marder) Positive and Negative Syndrome Scale (PANSS) factors is confounded by moderate-to-high between-factor correlations. In a previous pooled analysis of short-term, placebo-controlled lurasidone clinical trials, clustering and factor analysis were used to identify an uncorrelated PANSS score matrix (UPSM) that generated transformed PANSS factor scores that were found to have high face validity (good correlation with standard [Marder] PANSS factors), and high specificity/orthogonality (low levels of between-factor correlation) in measuring change. The UPSM was derived from analysis of PANSS change scores. In a validation analysis using 12 separate clinical trials, we confirmed that the weighted UPSM coefficients had generalizable utility, yielding transformed PANSS factors with high specificity while retaining good levels of correlation with standard PANSS factors. In contrast, standard factor analyses in schizophrenia derive their symptom structures by analyzing severity of baseline scores. We wished to determine to what extent an UPSM transform of PANSS severity scores at study baseline would correlate with standard Marder PANSS factors, and with well-validated assessment scales, including the Montgomery Åsberg Depression Rating Scale (MADRS) and the Negative Syndrome Assessment Scale (NSA).
Methods: Baseline PANSS factor data were analyzed from a randomized, double-blind, placebo-controlled, 6-week study of lurasidone 80 mg/d, lurasidone 160 mg/d, and quetiapine-XR 600 mg/d in the treatment of schizophrenia in patients ($N = 486$) with a baseline PANSS total score ≥ 80. Weighted UPSM coefficients, derived from previous analyses of PANSS change scores, were applied to the baseline PANSS factor scores to generate transformed PANSS factors. Correlation (Pearson's) and cluster analyses were then performed among PANSS factors together with other independent measures, including MADRS and NSA scales.
Results: UPSM transformed PANSS factor scores at Baseline were found to correlate well with their respective standard PANSS factors. The baseline correlation between NSA score, and standard PANSS negative symptom score was 0.68. The baseline correlation between NSA score and transformed PANSS negative-apathy/avolition was 0.53; the correlation at study baseline between NSA score and transformed PANSS negative-deficit of expression subscore was 0.55. At baseline, the correlation between MADRS total score, and standard PANSS depression/anxiety factor and transformed PANSS depression factor, respectively, was 0.56 and 0.53. Cluster analyses revealed co-clustering of positive, negative, cognitive, and movement symptom measures with specific UPSM-transformed PANSS factors. At baseline, as was observed with change during treatment, there was a greater degree of mutual orthogonality among UPSM-transformed PANSS factors relative to standard (Marder) PANSS factors.
Conclusions: These results, based on an analysis of data from a placebo-controlled schizophrenia trial, confirm and extend our UPSM change score analysis, and show that the UPSM transform can also be applied to PANSS factor scores at baseline. These findings provide further evidence for a consistent underlying schizophrenia symptom structure that is present in both exacerbated and stable (treated) states.
Sponsored by Sunovion Pharmaceuticals, Inc.

Keywords: Schizophrenia, Atypical Antipsychotics, Assessment, Positive and Negative Syndrome Scale
Disclosure: Part 1: Sunovion Pharmaceuticals, Inc., Employee

T207. Childhood Trauma is Associated With Decreased Gray Matter Volume of Language Areas in Psychosis Patients With Auditory Hallucinations

Ann Shinn*, Eunkyoung Kang, Valerie Sydnor, Jessica Talero, Melissa Hwang, Milissa Kaufman, Sylvain Bouix, Martha Shenton, Dost Ongur

Harvard Medical School, McLean Hospital, Belmont, Massachusetts, United States

Background: Abnormalities of brain areas involved in auditory and speech/language processing have been implicated in the pathophysiology of auditory hallucinations (AH). A growing literature also suggests that trauma, especially early in life, increases the risk of AH. We investigated gray matter volume (GMV) of language-related brain areas in psychosis patients with lifetime history of AH (AH) compared to psychosis patients without AH (NAH) and healthy controls (HC). We predicted that AH patients would show decreased GMV in language-related brain areas, and that the degree of GMV reduction would be related to the severity of childhood trauma.
Methods: We collected the childhood trauma questionnaire (CTQ) (Bernstein et al., 1997) and high-resolution structural MRI (Siemens 3T TIM Trio, multiecho MPRAGE, voxel size 1.0mm3) in 36 AH, 29 NAH, and 26 HC male and female participants 18-50 years old. Patients were all-comers with psychosis, including schizophrenia (SZ), schizoaffective disorder (SZA), and psychotic bipolar disorder (BP). Patients scoring 3 (threshold) on SCID item B16 were categorized AH (24 SZ/SZA, 12 BP); all others NAH (12 SZ/SZA, 17 BP).
Image analysis included visual quality control, production of brain masks using multi-atlas brain segmentation, and Free Surfer anatomic parcellation (http://surfer.nmr.mgh.harvard.edu/) based on the Desikan-Killiany atlas. We selected, a priori, 13 brain regions in each hemisphere for analysis. Eleven correspond to well-defined language-sensitive areas (Fedorenko et al., 2010): temporal pole (TP), inferior temporal (ITG), middle temporal (MTG), superior temporal (STG), pars orbitalis (IFGorb), pars triangularis (IFGtri), superior frontal (SFG), caudal middle frontal (cMFG), inferior parietal (IPL), supramarginal (SMG), and cerebellum (CER). We also included transverse temporal (TT), corresponding to the primary auditory cortex, and caudal/dorsal anterior cingulate (dACC), a cognitive control region frequently implicated in AH pathophysiology. GMVs were divided by the estimated total intracranial volume. We used SPSS to perform ANOVA and calculate Pearson's correlation coefficients. In this preliminary analysis, we report results meeting a threshold of $p < 0.05$, uncorrected.
Results: AH and trauma:
We found an effect of AH status on all 5 CTQ domains: emotional abuse (mean in AH 11.3, NAH 10.8, HC 6.2; $F2,86 = 7.9$, $p = 0.001$), physical abuse (7.5, 7.0, 5.4; $F2,86 = 3.4$, $p = 0.04$), sexual abuse (7.9, 6.1, 5.2; $F2,95 = 4.6$, $p = 0.01$), emotional neglect (11.0, 10.3, 6.8; $F2,86 = 7.2$, $p = 0.001$), and physical neglect (7.4, 7.0, 5.3; $F2,86 = 4.7$, $p = 0.01$). Post-hoc pair-wise comparisons showed the findings to be driven primarily by differences between AH and HC. AH reported greater sexual abuse than NAH; otherwise, AH and NAH did not differ significantly in trauma history.
AH and gray matter volume of language areas:
We found an effect of AH status on GMV in right (R) SFG ($F2,88 = 3.5$, $p = 0.04$), left (L) TP ($F2,88 = 4.9$, $p = 0.009$), LITG ($F2,88 = 4.5$, $p = 0.01$), LMTG ($F2,88 = 6.8$, $p = 0.002$), LSTG ($F2,88 = 3.6$, $p = 0.03$), RSTG ($F2,88 = 3.5$, $p = 0.03$), RTT ($F2,88 = 3.1$, $p = 0.049$), LSMG ($F2,88 = 3.3$, $p = 0.04$), and RSMG ($F2,88 = 3.7$, $p = 0.03$). Post-hoc comparisons showed the findings were due to differences between AH and HC (LMTG, LSTG, LSMG, RSFG), or NAH and HC (LTP, LITG, LMTG, RSTG, RTT, RSMG). There were no significant GMV differences between AH and NAH.
AH, trauma, and gray matter volume of language areas:
The relationship between trauma and GMV varied considerably by AH group. Among AH patients, emotional abuse was negatively correlated with GMV in LIFGtri ($R = -.38$, $p = 0.02$), RIFGorb ($R = -.43$, $p = 0.01$)], and RCER ($R = -.35$, $p = 0.04$). Physical abuse was negatively correlated with GMV in LIFGorb ($R = -.43$, $p = 0.01$), RIFGorb ($R = -.42$, $p = 0.01$), LIFGtri ($R = -.51$, $p = 0.002$), RIFGtri ($R = -.34$, $p = 0.048$), RSFG ($R = -.43$, $p = 0.01$), LITG ($R = -.43$, $p = 0.01$), RITG ($R = -.35$, $p = 0.04$), LMTG ($R = -.40$, $p = 0.02$), RTT ($R = -.37$, $p = 0.03$), RSMG ($R = -.38$, $p = 0.02$), LCER ($R = -.42$, $p = 0.01$) and RCER ($R = -.49$, $p = 0.003$). Physical neglect was negatively correlated with GMV in RIFGorb ($R = -.39$, $p = 0.02$), LTP ($R = -0.38$, $p = 0.02$), and LITG ($R = -0.38$, $p = 0.02$). We found no significant correlations between GMV and sexual abuse or emotional neglect in the AH group.
In NAH patients, sexual abuse was positively correlated with LIPL GMV ($R = .50$, $p = 0.006$). There were no significant correlations between GMV and emotional abuse or neglect, or physical abuse or neglect.
In HC, we found positive correlations between emotional abuse and RCER ($R = .44$, $p = 0.03$) and between sexual abuse and RSFG ($R = .51$, $p = 0.01$). Physical abuse ($R = -.43$, $p = 0.03$), emotional neglect ($R = -.53$, $p = 0.006$), and physical neglect ($R = -.49$, $p = 0.01$) were negatively correlated with RMTG. Emotional neglect was also negatively correlated with RcMFG ($R = -.47$, $p = 0.02$).
Conclusions: Among patients with AH, childhood emotional abuse, physical abuse, and physical neglect negatively correlated with GMV in multiple language-related areas; i.e., the greater the trauma exposure, the lower the GMV in these regions. This pattern was observed only in the AH group. We found no negative correlations between trauma and GMV in the NAH group. Furthermore, the NAH and HC groups were found to have brain regions that correlated positively with trauma, possibly suggestive of compensatory changes. While this study is cross-sectional, and the results preliminary, the data suggest that trauma may increase AH risk in part through loss of GMV in language-related brain areas.
Keywords: Auditory Hallucinations, Childhood Trauma, Gray Matter Volumes, Dimensions of Psychosis
Disclosure: Nothing to Disclose.

T208. Suicide in Prodromal Psychosis: Examining Predictors of Suicide Attempts in Individuals With Attenuated Psychosis Syndrome

Zachary Zuschlag*, Mark Hamner

Department of Veterans Affairs, Charleston, South Carolina, United States

Background: A strong association has been shown to exist between schizophrenia and suicide; however, research examining suicidality in the prodromal phase of psychotic disorders is limited. Although the prevalence of suicidality in the prodromal psychosis population has been observed, relatively little is known about predictive factors for suicide attempts or completed suicides in this population. The aim of the current study was to examine suicidal behaviors and potential risks factors in order to identify predictors of suicide attempts in a population of prodromal individuals, specifically those identified as having the DSM-5-defined Attenuated Psychosis Syndrome.
Methods: A retrospective review was conducted to identify individuals with APS during a 5-year period across a large medical university's inpatient and outpatient settings. A secondary analysis of APS individuals was conducted to examine a number of covariates in an effort to identify potential predictors of suicide attempts. Descriptive statistics were performed to determine statistical significance. Chi-square analysis was used to analyze dichotomous variables. Non-dichotomous variables were analyzed using a t-test for comparison of means. Final analysis consisted of fitting a multivariable logistic regression model to control for socio-demographic factors.
Results: Individuals meeting diagnostic criteria for the Attenuated Psychosis Syndrome ($N = 152$) were included in the data analysis. Of these 152 individuals, 40 had at least 1 lifetime suicide attempt, 26.3% of our study population. A number of covariates were analyzed to determine if their presence increased the rates of suicide attempts. Six covariates were found to be statistically significant predictors of suicide attempts in our population: Axis II disorder ($p = 0.006$, OR = 3.353 with a 95% CI at 1.408-7.985); personal history of trauma as a whole ($p = 0.022$, OR = 2.487, CI = 1.408-7.985); the sub category of sexual trauma ($p = 0.005$, OR = 3.968, CI = 1.512-10.411); tobacco use ($p = 0.039$, OR = 2.266, CI = 1.042-4.929); family history of non-psychotic Axis I disorder ($p = 0.042$, OR = 2.370, CI = 1.032-5.444); and number of hospitalizations ($p = 0.001$, OR = 1.318, CI = 1.113-1.560) .
Conclusions: Our results show that suicidality is a prominent feature of prodromal individuals with APS. 26.3% of individuals in our study had at least 1 lifetime suicide attempt. Six risks factors were found to be statistically significant predictors of lifetime suicide attempts: Axis II disorder, history of trauma as a whole, the sub-category of sexual trauma, tobacco use, family history of non-psychotic Axis I disorder, and number of inpatient psychiatric hospitalizations.
Keywords: Schizophrenia Prodrome, Suicide, Early Psychosis, DSM-5
Disclosure: Nothing to Disclose.

T209. Long-Term Safety and Tolerability of a New Monthly Extended-Release Formulation of Risperidone (RBP-7000) in the Treatment of Subjects With Schizophrenia

James Graham*, John Csernansky, Maurizio Fava, John Newcomer, Anne Andorn, Sunita Shinde, Gilbert Muma, Christian Heidbreder

Indivior, Inc., Richmond, Virginia, United States

Background: RBP-7000 (risperidone in ATRIGEL® Delivery System) is a once-monthly, extended-release depot formulation in development for the treatment of schizophrenia. RBP-7000 delivers therapeutically relevant plasma concentrations of risperidone on the first day of dosing, with no required loading dosing or supplemental dosing. RBP-7000 was designed to optimise D2 receptor occupancy and to achieve rapid therapeutic concentrations with the aim of preventing early treatment failure, which is often due to partial or complete nonadherence. RBP-7000 simplifies adherence by allowing continuous treatment (monthly physician-administered dosing) instead of daily oral treatment (patient-administered) or twice-monthly treatment (physician-administered), with no need for supplemental oral dosing. These conveniences, both at treatment initiation and during maintenance therapy, may contribute to better patient outcomes by improving adherence and continuity of treatment. The efficacy, safety and tolerability of RBP-7000 were previously examined in an 8-week double-blind placebo-controlled trial (NCT02109562) that included adults (18-55 years) with a DSM-IV-TR diagnosis of schizophrenia. The primary objective of the current study (NCT02203838) was to evaluate the long-term safety and tolerability of RBP-7000.
Methods: This phase 3 open-label study (53 US sites) included completers from the prior 8-week study (rollover subjects) and new patients (de novo subjects). De novo subjects were required to have a DSM-IV-TR diagnosis of schizophrenia and a Positive and Negative Syndrome Scale (PANSS) total score ≤ 70. Rollover subjects, who received 2 injections of placebo or RBP-7000 (90 mg or 120 mg) in the prior study, received 11 additional injections of open-label RBP-7000 (120 mg) in the current study. De novo subjects received 13 injections of open-label RBP-7000 (120 mg) after being titrated or converted to oral risperidone (3 or 4 mg/day). All subjects were allowed a single down-titration to 90 mg and a single up-titration back to 120 mg based on tolerability. Safety assessments included treatment-emergent adverse events (TEAEs), clinical laboratory tests, vital signs, electrocardiogram (ECG), Abnormal Involuntary Movement Scale (AIMS), Simpson-Angus Scale (SAS), Barnes Akathisia Rating Scale (BARS) and Columbia-Suicide Severity Rating Scale (C-SSRS). Efficacy measures included the mean change from baseline by visit in PANSS total score. All analyses were conducted in the safety population, defined as all subjects who received ≥ 1 dose of open-label RBP-7000.
Results: The safety population included 500 subjects (rollover, $n = 92$; de novo, $n = 408$); 234 (46.8%) who completed the study. The majority of subjects were male (67.8%) and black/African-American (70.8%); mean age was 45.1 years. Mean baseline PANSS total scores ($\pm$ SD) were: 58.0 ($\pm$8.33)

for de novo subjects; 72.9 (±20.99), 76.4 (±15.99) and 71.0 (±13.93) for rollover subjects who previously received placebo, RBP-7000 90 mg and RBP-7000 120 mg, respectively. Overall, 73.4% of subjects had ≥ 1 TEAE, 11.6% had a TEAE that led to study discontinuation, and 8.6% had a TEAE leading to dose modification. Most TEAEs were mild or moderate; approximately 5% of subjects had a severe TEAE. TEAEs reported in > 5% of all subjects were injection site pain (13.0%), weight increase (12.8%), schizophrenia (7.8%), insomnia (7.0%), injection site nodule (6.8%), akathisia (6.0%), injection site induration (5.8%), and upper respiratory tract infection (5.2%). Most subjects (> 80%) did not experience injection site reactions (pain, tenderness, erythema/redness or induration/swelling), either immediately after dosing or at 3 hours postdose. Only 2 subjects discontinued the study due to an injection site reaction. Serious TEAEs were reported in 6.8% of subjects, including 4 that resulted in death (cardiac arrest, psychotic disorder, pulmonary embolism, cause unknown). However, no serious TEAE was considered related to study drug. There were no clinically meaningful changes, patterns or trends observed in laboratory parameters, vital signs or ECG parameters. No neuroleptic malignant syndromes or seizures were reported. Mean BARS, AIMS and SAS total scores remained relatively stable across study visits. There was no evidence of increased risk of suicidality based on C-SSRS assessments. The study was not powered for efficacy, but PANSS total score decreased (improved) from baseline to end of treatment in rollover subjects who previously received RBP-7000 90 mg (-12.5 [± 15.53]) or 120 mg (-10.9 [± 13.16]), indicating continued improvement in acute schizophrenia following months of exposure to RBP-7000. A larger mean decrease (improvement) in PANSS total score was found in subjects who had previously received placebo (-20.2 [± 15.59]), but the number of evaluable subjects was small. Among de novo subjects who were stable at enrollment, PANSS total score generally remained stable with a minimal mean change from baseline to end of treatment (-0.4 [± 8.67]).

Conclusions: Except for injection site tolerability, which is expected with all subcutaneously administered medications, there were no notable safety signals. The safety profile seen with 12 monthly injections of RBP-7000 was consistent with that of oral risperidone. Importantly, schizophrenia symptoms continued to improve in previously exposed subjects with acute schizophrenia who rolled over into the longer-term study.

Keywords: Antipsychotic, Schizophrenia, Extended-Release Depot, Long-Term Safety

Disclosure: Part 2: Indivior, Inc., Employee, **Part 5:** Indivior, Inc., Employee.

T210. D2 Dopamine Receptors Alter Circadian Rhythms in Pancreatic Islet Cells: Implications for the Metabolic Side Effects of Antipsychotic Drugs

Danielle Chipchura, Heather Wei, Zachary Freyberg, Michael McCarthy*

University of California, San Diego, San Diego, California, United States

Background: Antipsychotic drugs (APDs) are used widely in the treatment of psychiatric disorders. However, these drugs also cause unwanted side effects including weight gain and metabolic abnormalities. Accordingly, patients receiving APDs are at elevated risk of cardiac disease, type 2 diabetes and shortened life span. The pharmacological mechanisms for APD-induced metabolic dysfunction are poorly understood, but may involve the blockade of D2 dopamine receptors (D2R). In addition to their expression in brain, D2R is located in pancreatic beta-cells where they negatively regulate insulin release. As a critical regulator of blood glucose, insulin release follows a circadian rhythm to anticipate food intake and prolonged periods of fasting. Under healthy conditions, insulin release peaks during the day and is low at night. However, disruptions of circadian rhythms, and the insulin rhythm in particular may cause hyperglycemia, weight gain and lipid abnormalities. Many APDs are sedating, and dosed at night to promote sleep. We hypothesize that APDs, particularly when taken at night, may disrupt 24 h rhythms in beta-cells, and interfere with the regulation of insulin by dopamine through a circadian mechanism.

Methods: We examined laboratory and anthropometric data from patients treated over a 12-month period with a partial D2 agonist (aripiprazole) either in the morning ($n = 90$) or at bedtime ($n = 53$). Differences in metabolic parameters and BMI were examined in the two groups who got the medication at different times of day. We then established an in vitro model of pancreatic beta-cell circadian rhythms using beta cell-derived INS-1E cells. To measure circadian gene expression rhythms, INS-1E cells were transduced with a lentivirus containing the Per2-luc circadian reporter and a blasticidin resistance element. Per2-luc rhythms were measured for 3-4 days under constant conditions in cell lines stably expressing Per2-luc. To measure circadian rhythms in insulin release, we used a lentiviral construct to deliver a proinsulin-luc reporter. Insulin release was then measured for 1-2 days under constant conditions.

Results: Patients treated with aripiprazole in the evening had significantly higher levels of serum triglycerides and lower levels of HDL cholesterol compared to patients who took the drug in the morning. There were no changes in hemoglobin A1c, fasting glucose levels, or BMI. In vitro, INS-1E cells showed circadian rhythms in gene expression. Stimulation of D2R with agonists including bromocriptine or L-DOPA decreased the rhythm amplitude, whereas dopamine D1 receptor stimulation did not have a major effect. In contrast, sulpiride, a D2R-selective antagonist, reversed the effects of D2R stimulation by bromocriptine or L-DOPA on rhythms. Insulin release was rhythmic and in anti-phase with Per2-luc expression, with highest levels of insulin release occurring at times of low levels of Per2-luc (corresponding to daytime). D2R antagonism by sulpiride significantly altered the rhythm of insulin release, causing higher levels of insulin during the nadir.

Conclusions: Dopamine signaling in pancreatic beta-cells alters circadian rhythms in gene expression and insulin release. APDs that block D2R interfere with these rhythms and may lead to increased insulin secretion over 24 hr cycles, especially at night when insulin levels are typically low. This can translate clinically in patients treated with aripiprazole (or similar APDs), where time of dosing affects likelihood of metabolic risk. Patients taking APDs at night may have additional metabolic risk factors compared to those who take the medication during the day. Circadian interventions may therefore reduce the adverse consequences on metabolism of APDs.

Keywords: Antipsychotic Unduced Weight Gain, Circadian Rhythm, Dopamine 2 Receptor, Metabolic Side Effects
Disclosure: Nothing to Disclose.

T211. Connecting NMDA Receptor Disruption to a Specific Aberration in Prefrontal Cortical Neurophysiology Essential for Executive Function

Mary Binko, Catharine Mielnik, Amy Ramsey, Evelyn Lambe*

University of Toronto, Toronto, Canada

Background: The cognitive deficits of schizophrenia include impaired executive function, attention problems and distractibility, indicative of prefrontal cortical disruption. Converging evidence suggests that such cognitive symptoms arise from glutamatergic abnormalities, specifically from N-methyl-D-aspartate (NMDA) receptor hypofunction. Indeed, prominent deficits in executive function are found in transgenic mice with reduced expression of the obligate GluN1 subunit of the NMDA receptor.
Methods: Here, we examine functional brain changes in the prefrontal cortex of GluN1 knockdown mice relative to wild-type littermate controls. In a tripartite approach, we examined pyramidal neurons in acute prefrontal brain slices: (1) measuring basic electrophysiological properties, (2) probing the integrity of synaptic and extra-synaptic NMDA receptor responses, and (3) evoking network activity as an integrated measure of regional brain function.
Results: Overall, these experiments paint a striking portrait of the prefrontal cortex in the GluN1 knockdown mouse. Many aspects of cellular electrophysiology are largely preserved in the face of lifelong NMDA receptor knockdown, including near-normal NMDA currents to synaptic stimulation. Yet, responses to extra-synaptic NMDA stimulation are severely disrupted, and electrically-evoked network activity is absent under our experimental conditions in the GluN1 prefrontal cortex. Ongoing work examines mechanisms to restore network activity and executive function.
Conclusions: Since network activity is essential for executive function, its specific disruption in the GluN1 knockdown mice would account for the cognitive deficits observed in this model of schizophrenia.
Keywords: Medial Prefrontal Cortex, NMDA Receptor, Electrophysiology, Genetic Mouse Model, Schizophrenia
Disclosure: Nothing to Disclose.

T212. Circuit Level Link Between Disruption of Parvalbumin+ Interneuron Function and Gamma Band Oscillations in a Rodent Model of Schizophrenia

Anitha Manohar*, Michael Marino

Merck Research Laboratories, West Point, Pennsylvania, United States

Background: Schizophrenia is a complex disorder that includes positive, negative, and cognitive symptom domains. The clinical efficacy of most available treatments is limited to the positive symptom domain (i.e. reduction of hallucinations). Cognitive impairment associated with schizophrenia remains a significant unmet medical need and is highly correlated with quality of life concerns such as addiction, unemployment, and suicide. Cortical parvalbumin positive (PV+) interneurons have been implicated in the pathophysiology of schizophrenia. These fast spiking neurons are thought to be essential for the generation of high frequency oscillations in the gamma range (30-80 Hz) which are correlated with cognitive task performance. More importantly, schizophrenia patients show dysfunctional gamma oscillations during cognitive tasks and in response to auditory stimuli. This evidence taken together with post-mortem reports that show a decrease in a variety of markers of PV+ interneuron function suggest that disruption in the PV+ system may underlie the gamma deficit and cognitive impairment associated with schizophrenia. Thus, human biology indicates that reversing the hypo-function of cortical PV+ interneurons and restoring gamma oscillations may be a viable means of treating the cognitive symptoms of the disease. In our study, we seek to understand the role of PV+ interneurons in modulation of cortical information and the network level generation of gamma oscillations. We use a 40 Hz click train of auditory pulses as stimuli to entrain the network rhythm to gamma frequency. This clinically translatable paradigm can then be used to study the cortical network at both the level of single neurons as well as activity generated by neuronal ensembles recorded in vivo from awake behaving rats. Furthermore, we will employ NMDA receptor antagonists, which are known to closely mimic clinical symptoms of schizophrenia (positive, negative and cognitive), to disrupt gamma oscillations and evaluate the ability of current and putative novel treatment to restore function. As a first step, we have measured the 40 Hz entrainment in the prefrontal local field, and have begun studies to disrupt this activity with ketamine.
Methods: Male Sprague Dawley rats were implanted with electrodes in the prefrontal cortex (PFC, layer 5, pre-limbic and infra-limbic regions). In-vivo extracellular activity was recorded from freely moving rats in their home cages in a sound attenuated recording box using tethered cables connected through a commutator. Rats were injected with an acute dose of NMDA receptor antagonist Ketamine (10 mg/kg) or vehicle control (saline). We recorded spontaneous activity as well as auditory click-train evoked steady state responses (ASSR). Wavelet based spectral analysis was used to obtain time frequency decomposition of the signal to calculate average power and phase locking to click trains across trials (inter-trial coherence, ITC). The power and ITC were averaged between 100-500 ms after stimulus presentation in order to isolate the steady state response from the initial evoked response in the signal. The peak values at 40 Hz was used to compare the ASSR across animals.
Results: A click train of repeated pulses presented at 40 Hz induces a corresponding oscillation at 40Hz in the local field potentials recorded from the PFC. This is measured as a peak in the mean power spectrum averaged across trials. An acute injection of ketamine (10 mg/kg, s.c.) does not produce a change in the average power in the induced gamma band oscillations when compared to vehicle administration in the same cohort of animals (paired t-test, $t = 0.3239$, $df = 5$, $p = 0.75$). Further there was no significant disruption in the phase coherence of gamma oscillations induced by the stimuli across trials as measured by the ITC (paired t-test,

$t = 1.057$, $df = 5$, $p = 0.33$). Current efforts are aimed at exploring the dose-effect function of ketamine to determine if a robust disruption of ASSR can be achieved.
Conclusions: In summary, we can say that NMDAR antagonist ketamine at 10 mg/kg does not decrease the mean power and phase coherence of auditory steady state response at 40Hz. The effect of NMDA antagonism by ketamine on entrained gamma oscillations may be dose dependent and/or frequency dependent. Further evaluation of ketamine dose, entrainment frequencies and time points are required before investigating the role of PV+ interneurons in the network level generation of gamma oscillations.
Keywords: Parvalbumin Neurons, Schizophrenia, NMDA Receptor, Gamma Oscillation, Ketamine
Disclosure: Part 5: Merck & Co., Employee.

T213. Genome-Wide Association of Endophenotypes for Schizophrenia

Tiffany Greenwood*, Laura Lazzeroni, Monica Calkins, Robert Freedman, Michael Green, Raquel Gur, Ruben Gur, Gregory Light, Keith Nuechterlein, Ann Olincy, Allen Radant, Larry Seidman, Larry Siever, Jeremy Silverman, William Stone, Catherine Sugar, Neal Swerdlow, Debby Tsuang, Ming Tsuang, Bruce Turetsky, David Braff

University of California, San Diego, La Jolla, California, United States

Background: As part of the Consortium on the Genetics of Schizophrenia (COGS), we have previously reported the results of our efforts to characterize the genetic architecture of 12 endophenotypes for schizophrenia. We have previously reported substantial heritability for all endophenotypes in the COGS-1 family sample. Subsequent studies of these families implicated genomic regions of interest through candidate gene association and linkage methods, most of which converge on a single gene network related to glutamate signaling. Here we report our initial efforts to capture association for these same endophenotypes on a genome-wide scale in the COGS-2 cohort of schizophrenia cases and controls.
Methods: A sample of 1352 schizophrenia cases and healthy control subjects, collected as part of phase 2 of COGS, was genotyped as part of the Psychiatric Genomics Consortium (PGC) for the PsychChip. This array contains a 240,000 genome-wide marker grid, 240,000 exome markers, and 50,000 custom content markers for psychiatric illness and population ancestry and allows for accurate imputation of variants with minor allele frequencies of 1%. Standard quality control filters were applied, removing subjects for gender discrepancies, invariant markers and those with minor allele frequencies <0.01, and SNPs with call rates <0.98 or Hardy-Weinberg equilibrium p values $<10\text{-}6$ among controls. Ancestry was confirmed via comparison with the HapMap3 reference, and population outliers were removed. The two largest groups, reflecting European and Latino ancestry, were selected for further analysis. Each group was imputed separately to the 1000 genomes reference using the PGC pipeline, retaining only markers with high confidence scores. A final round of quality control filters were applied as above to each sample separately to generate final datasets including >6.2M variants with a total genotyping rate >0.99. The final datasets included 523 cases and 506 controls of European ancestry and 100 cases and 83 controls of Latino ancestry. Multidimensional scaling was used to calculate principal components for each dataset for use as covariates to correct for residual population stratification and genotype pool discrepancies. Association was performed using linear regression, adjusting for age and sex as needed, as well as the first five principal components. We evaluated all primary endophenotypes that overlap between COGS-1 and COGS-2. These include prepulse inhibition of startle (PPI) at 60ms, antisaccade, California Verbal Learning Test (CVLT) trials 1-5 total correct responses, degraded stimulus Continuous Performance Test (DSCPT) discrimination (d'), and Letter-Number Span (LNS) correct reordering, as well as abstraction and mental flexibility (ABF), face memory (FMEM), spatial memory (SMEM), verbal memory (VMEM), spatial processing (SPA), sensori-motor dexterity (SM), and emotion recognition (EMO) from the University of Pennsylvania Computerized Neurocognitive Battery. Association results from the European and Latino analyses were combined through meta-analysis, weighted for sample size.
Results: Many independent regions exceeding a genome-wide suggestive threshold of 10-5 were identified in the COGS-2 European case-control sample that were strengthened when combined with the results in the Latino case-control sample. Replication of the most prominent regions was assessed in 276 schizophrenia probands and 321 healthy control subjects of European ancestry from the COGS-1 families, who were also genotyped for the PsychChip and underwent identical analytic procedures.
Conclusions: These analyses have identified many genomic regions of interest that require further exploration and validation. We note that the case-control sample presented here represents only a portion of the complete COGS-2 sample, with 1034 additional COGS-2 subjects of African American ancestry and 1093 COGS-1 family members still pending genotyping. Whole genome sequencing is also pending for both the COGS-1 and COGS-2 samples that will allow for a deeper investigation of the genomic regions identified here though association. It is important to note that we are investigating the genetic architecture of heritable neurocognitive and neurophysiological endophenotypes associated with schizophrenia risk. Understanding the molecular basis of these endophenotypes, many of which are recognized as treatment targets by the FDA, will pave the way for precision based medicine.
Keywords: Schizophrenia Genetics, Endophenotype, Genome-Wide Association Study, Cognition
Disclosure: Nothing to Disclose.

T214. A Phase 3 Study to Determine the Antipsychotic Efficacy and Safety of ALKS 3831 in Adult Patients With Acute Exacerbation of Schizophrenia

Steven Potkin*, Bernard L. Silverman, Adam Simmons, Ying Jiang, Lauren DiPetrillo, David McDonnell

University of California, Irvine, Irvine, California, United States

Background: ALKS 3831 is composed of a flexible dose of the antipsychotic olanzapine (OLZ) and a fixed dose of samidorphan (a μ-opioid receptor antagonist) and is under development for the treatment of schizophrenia. ALKS 3831 has been studied in phase 1 (healthy volunteers) and phase 2 (stable schizophrenia) studies. In the phase 2 study, ALKS 3831 mitigated OLZ-associated weight gain and exhibited an antipsychotic efficacy similar to OLZ. This phase 3 study aimed to assess antipsychotic efficacy and safety of ALKS 3831 in patients with acute exacerbation of schizophrenia.
Methods: This was a 4-week, phase 3, randomized, double-blind, active (OLZ monotherapy) and placebo-controlled study of ALKS 3831 in patients experiencing acute exacerbation of schizophrenia (ClinicalTrials.gov: NCT02634346). Following screening, eligible patients ($N = 403$) were randomized 1:1:1 to one of three groups: ALKS 3831, OLZ monotherapy, or placebo. Patients were treated in an inpatient setting for the first 2 weeks of the study period and could be treated as inpatients or outpatients for the remaining 2 weeks. Patients were excluded if they received OLZ within 6 months prior to screening. Antipsychotic efficacy was assessed using the Positive and Negative Syndrome Scale (PANSS), Clinical Global Impression–Severity (CGI-S), and CGI–Improvement (CGI-I) scales. The primary endpoint, change from baseline in PANSS total score at Week 4, was analyzed using a mixed model with repeated measurements (MMRM). The key secondary endpoint, change from baseline in CGI-S at Week 4, was also analyzed by MMRM. PANSS response and CGI-I response were analyzed by a logistic regression model. Additionally, safety and tolerability were assessed as adverse events (AEs) and were analyzed using descriptive statistics.
Results: Of 401 patients randomized and dosed to ALKS 3831 ($n = 134$), OLZ ($n = 133$), and placebo ($n = 134$), 91.0%, 88.7%, and 82.8% of patients, respectively, completed treatment. The most common reason for discontinuation was withdrawal by patient (6.0% in the ALKS 3831 group, 6.8% in the OLZ group, and 6.0% in the placebo group). Baseline characteristics were similar between groups: the mean age was 41 years and 61% were male. Baseline mean body mass index was higher in the OLZ group compared with the ALKS 3831 group. Baseline mean ± standard deviation scores were 101.7 ± 11.9 for PANSS total score and 5.1 ± 0.7 for CGI-S score. The mean OLZ dose was 18.4 mg/day in both active treatment arms. Least squares (LS) mean difference ± standard error (SE) versus placebo from baseline to Week 4 in PANSS total score was −6.4 ± 1.8 ($P < 0.001$) and −5.3 ± 1.8 ($P = 0.004$) for the ALKS 3831 and OLZ groups, respectively. LS mean difference ± SE versus placebo from baseline to Week 4 in CGI-S score was −0.38 ± 0.12 ($P = 0.002$) and −0.44 ± 0.12 ($P < 0.001$) for the ALKS 3831 and OLZ groups, respectively. The percentage of patients with an improvement in PANSS response (defined as ≥30% improvement from baseline) at Week 4 was 59.8%, 53.8%, and 38.3% in the ALKS 3831, OLZ, and placebo groups, respectively. The percentage of patients with an improvement in CGI-I response (defined as score of ≤2) at Week 4 was 57.6%, 50.8%, and 33.1% in the ALKS 3831, OLZ, and placebo groups, respectively. Discontinuation due to AEs was low in all groups: 1.5%, 2.3%, and 5.2% in ALKS 3831, OLZ, and placebo groups, respectively. Common AEs (≥4%) included weight gain, somnolence, dry mouth, anxiety, headache, schizophrenia, and agitation. The number of patients in the two active treatment groups who experienced movement disorders was generally low (≤6.0%).
Conclusions: ALKS 3831 demonstrated greater antipsychotic efficacy compared with placebo, as measured by the PANSS and CGI-S scale, and was similar to the active control, OLZ. ALKS 3831 was generally well tolerated, with a similar safety profile to OLZ.
Keywords: Antipsychotic Treatment, Schizophrenia, Samidorphan, Olanzapine
Disclosure: Part 1: Alkermes, Advisory Board, Grant Otsuka, Advisory Board, Grant, Honoraria, Sunovion, Advisory Board, Honoraria, Roche, Advisory Board, Lundbeck, Advisory Board, Grant, FORUM, Advisory Board, Grant, Allergan, Advisory Board, Honoraria, Eli Lilly, Grant, Toyama, Grant, Eisai, Grant, Novartis, Honoraria, **Part 4:** Eli Lilly, Grant, Toyama, Grant, Otsuka, Grant, FORUM, Grant, Alkermes, Grant, Eisai, Grant, Lundbeck, Grant.

T215. Meta-Analysis on the Association Between Putative Schizophrenia Risk Genes and Prepulse Inhibition of the Acoustic Startle Response

Kenechi Ejebe*, Boris Quednow, Michael Wagner, Veena Kumar, Panagiotis Roussos

Icahn School of Medicine at Mount Sinai, New York, New York, United States

Background: Sensorimotor gating measured by prepulse inhibition (PPI) of the acoustic startle response (ASR) has been proposed as one of the most promising electrophysiological endophenotypes of schizophrenia. PPI involves a weaker stimulus (prepulse) inhibiting the reaction to a stronger stimulus (pulse). Stimuli include acoustic, tactile and light. During the past decade, a number of publications have reported significant associations between schizophrenia risk polymorphisms and PPI in samples of schizophrenia patients and healthy volunteers. However, an overall evaluation of the robustness of these results has not been published so far. We aimed to perform the first p-value-based meta-analysis of published and so far unpublished associations between gene polymorphisms and PPI of ASR.
Methods: We performed meta-analysis for published SNPs associated with PPI and schizophrenia by querying PubMed using the search term that covered the most relevant published association studies:(((gene OR genetics OR polymorphism OR polymorphisms OR SNP) AND (prepulse OR "pre-pulse") AND startle AND (humans OR volunteers OR patients)) NOT rats NOT mice NOT review [Publication Type]). This search was conducted on 12/05/2016 and received 61 publication results. 3 unpublished publications were also included. Each of the 64 publications was systematically reviewed for data results showing SNPs associated with PPI in patients with schizophrenia and healthy controls. 19 publications contained SNPs significantly associated with PPI in 722 patients with schizophrenia and 1,029 healthy controls. 41 SNPS were reported in two or more independent publications to be associated with PPI. For the meta-analysis, only SNPs with p-values that were found in 2 or more studies were included. A weighted Z

score approach was used, with all the p-value converted to Z-score.
Results: After Benjamini and Hochberg correction for the number of analyzed polymorphisms, significant association were shown for 16 SNPs including rs4680 (5 x 10-5) and rs1027599(4x 10-3) which were the most significant SNPs. However, none of these associations survived a whole-genome-correction.
Conclusions: These results imply that sensorimotor gating might be modulated by xyz genotype indicating a role of these gene variations in the development of early information processing deficits in schizophrenia. However, although gene PPI-gene associations seem to be stronger compared to the genetic associations with the complex phenotype of schizophrenia, the overall impact of single genes on PPI is still rather small suggesting that PPI is – like the disease phenotype – highly polygenic. Further studies should investigate gene-gene interactions within the group of polymorphisms with the strongest association related to PPI
Keywords: Prepulse Inhibition, Acoustic Startle Response, Sensorimotor Gating, Schizophrenia, SNP
Disclosure: Nothing to Disclose.

T216. Aberrant Activity in Conceptual and Sensorimotor Networks Underlie N400 Deficits in Schizophrenia: An ERP-FMRI "Fusion" Study

Michael Jacob*, Judith Ford, Brian Roach, Vince Calhoun, Daniel Mathalon

San Francisco Veterans Affairs Medical Center, San Francisco, California, United States

Background: The mapping of meaning onto everyday objects results from an interaction between perceptual and conceptual/semantic functions. In schizophrenia (SZ), the failure to effectively coordinate perceptual and conceptual systems might underlie thought disorganization or unusual thought content. Language processing impairments in SZ have been linked to disorganization within conceptual networks and/or deficits in the integration of perceptual features. Semantic priming tasks are used to map associations within semantic networks, revealed experimentally by speeded reaction times to primed stimuli. Event related-potentials (ERPs) are used to probe the neural substrates of semantic networks, triggering the N400 ERP component to unprimed, semantic violations. Reductions in N400 amplitudes have been observed in SZ, consistent with overly broad associations in semantic networks. Language paradigms using fMRI with SZ patients have revealed abnormal modulation across a range of cortical structures, while the underlying neuroanatomical substrates of the N400 remain unknown.
Methods: SZ patients ($n = 24$) and healthy control subjects (HC; $n = 25$) performed a picture-word semantic priming task in separate ERP and fMRI sessions. In this task, a line drawing of an object is presented (250 ms), and following a short delay (75 ms), a word appears. Subjects are instructed to respond by indicating whether the word was a semantic match (primed, 50% of trials) or non-match (unprimed, 50% of trials) to the preceding picture. Pictures were drawn from 10 natural categories, with half of the non-match trials comprising picture-word pairs from a related semantic category (e.g., banana-apple) and the other half comprising picture-word pairs from unrelated semantic categories (e.g., banana-cow). We focused our analyses on the effects of group (HC vs. SZ), priming (unprimed vs. primed) and category relatedness among unprimed words (related vs. unrelated). Three analyses were conducted on the data: (1) a standard, event-related general linear model (GLM) to determine the location and extent of brain activation clusters from the fMRI dataset, (2) a focused region of interest (ROI) analysis based on the conceptual/semantic network identified in a meta-analysis by Binder et al. 2009, and (3), a joint independent components analysis (JICA) to identify brain regions associated with the ERP N400 component. Correlations between brain activations and clinical measures of unusual thought content and thought disorganization were also assessed.
Results: Using a conventional, event-related GLM approach, we found no difference between unprimed and primed activation maps across groups (and no group x priming interaction effect) despite robust N400 priming effects and N400 amplitude deficits in SZ patients relative to HC. Focusing only on the unprimed, but semantically related, picture-word trials, we found one significant ($p = 0.012$, FDR-corrected) cluster in the cingulate cortex (BA31) with larger activation for related unprimed trials relative to primed trials across groups (but no group x priming interaction). No significant brain activation differences were found between unrelated unprimed trials and primed trials for either group. The analysis of category relatedness among unprimed trials showed significantly ($p < 0.05$, FDR-corrected) suppressed activity in response to unrelated, relative to related, non-match picture-word pairs in four regional clusters: visual cortex, right superior temporal gyrus, precuneus and anterior cingulate. We suspected that effects of category relatedness might be mediated by semantic interference in conceptual networks, as indicated by increased reaction times for related, relative to unrelated, non-match trials. Accordingly, we extracted activity for unrelated and related contrasts from ROIs within the conceptual network identified by Binder et al. Similar to the GLM analysis, we found suppressed activation in unrelated, relative to related, non-match trials, in the anterior cingulate and precuneus (condition x ROI interaction, $p = 0.0001$). This analysis also revealed a significant group x condition interaction ($p = 0.004$): the reduction of activity in the unrelated, relative to the related, condition was only significant in SZ patients. JICA identified an ERP-fMRI "fused" component that captured the N400 component and yielded associated fMRI maps for the related and unrelated non-match conditions. The JICA map for the related condition showed concordance with the conceptual network, particularly for midline cingulate structures, whereas the JICA map from the unrelated condition showed little concordance with the conceptual network, linked primarily to visual sensory and motor areas. The component scores associated with this fused JICA component were reduced in SZ relative to HC ($p < 0.001$). In SZ patients, activity within the precuneus from the related JICA map was inversely correlated with unusual thought content ($R2 = 0.378$, $p = 0.0014$).
Conclusions: These findings suggest that the neural networks associated with the N400 response to semantic violations

depends on conceptual relatedness. Unprimed but semantically related stimuli show increased activation in conceptual networks, particularly midline cingulate areas linked to the N400. Unprimed, semantically unrelated stimuli show decreased activation in conceptual networks, with N400 responses linked to sensorimotor areas. These results are consistent with semantic models of 'embodied abstraction,' where the contribution of sensorimotor systems to language processing depends on context. SZ patients show excessive deactivation of the conceptual network in addition to reduced sensorimotor activation, suggesting impaired sensorimotor embedding in semantic processing. Impaired embedding could reflect impoverished conceptual representation and overly broad semantic networks, possibly contributing to unusual thought content.

Keywords: Semantic Priming, EEG/ERP Electrophysiology, Schizophrenia, Joint Independent Component Analysis, fMRI

Disclosure: Nothing to Disclose.

T217. Chronic Stress Regulation of Sustained Attention and Cholinergic Dendritic Morphology in Rats

Debra Bangasser*, Alexander Telenson, Brittany Wicks, Joy Bergmann, Marni Shore, Natalie Newcamp, Attilio Ceretti, Samantha Eck, Arron Hall, Kimberly Wiersielis, Jessica Tucci

Temple University, Philadelphia, Pennsylvania, United States

Background: Attention impairments are found in patients with disorders including schizophrenia, attention deficit hyperactivity disorder, bipolar disorder, and Alzheimer's disease. Another shared feature of these disorders is that chronic stress worsens their symptoms and progression. However, the mechanisms by which chronic stress alters attention remain largely unknown. In this study, we examined in rats whether chronic variable stress alters sustained attention, the ability to monitor situations for intermittent and unpredictable events. Appropriately sustaining attention requires the engagement of the basal forebrain cholinergic system, so we are also beginning to explore whether chronic variable stress alters structural plasticity of cholinergic neurons.

Methods: To assess the effects of chronic stress on sustained attention, male and female rats were trained on a touchscreen, operant, sustained attention task in which they had to discriminate signal trials from non-signaled trials. After attaining criterion (70% correct responses on signal and non-signal trials), rats were exposed to a 6-day chronic variable stress procedure. Sustained attention was assessed 30 min following each daily stressor. Previous studies have shown that, during the sustained attention task, accuracy on signal trials is mediated by cholinergic neurons in the nucleus basalis of Meynert (NBM) in the basal forebrain. In other brain regions, such as the amygdala and hippocampus, chronic stress is thought to affect cognition by altering the morphology of dendrites on neurons within these regions. We wanted to determine whether chronic stress alters the morphology of dendrites on NMB cholinergic neurons. To address this question, we developed an innovative approach to label cholinergic neurons in rats by injecting a virus with Cre-dependent fluorescent marker into the NBM region of rats genetically modified to express Cre-recombinase under the control of the choline acetyltransferase promoter. Ongoing studies are imaging neurons from stressed and unstressed rats and then evaluating whether chronic variable stress induces any morphological changes.

Results: The effects of chronic variable stress on sustained attention were assessed in rats and we found that stressor exposure did not significantly alter performance on non-signal trials [$F(6,84) = 1.9$, $p = .17$]. However, exposure to chronic variable stress did disrupt performance on signal trials [$F(6,84) = 2.83$, $p = .02$]. Given that cholinergic neurons in the NBM mediate accuracy on signal trials, this result suggests that cholinergic neurons are affected by chronic stress. To determine whether chronic variable stress induces structural plasticity in cholinergic neurons, we performed a Sholl analysis by counting the number of dendrites that intersect with concentric circles that radiate from the cell body in 20μm increments. Preliminary results are revealing that chronic stress increases intersections with circles at a distance of 140μm and greater from the cell body ($p < .05$). These finding suggest that chronic variable stress induces dendritic hypertrophy.

Conclusions: The results of the present study suggest that chronic variable stress impairs aspects of sustained attention mediated by the basal forebrain cholinergic system. Our ongoing studies suggest that chronic stress can induce hypertrophy of cholinergic dendrites. If supported with more subjects, this result would suggest that chronic stress impairs attention by affecting inputs into the NBM via the regulation of the morphology of cholinergic dendrites. Importantly, understanding the processes by which chronic stress impairs attention may lead to the identification of new targets that can be manipulated therapeutically to improve attention in stressed patients with a variety of psychiatric and neurodegenerative disorders.

Keywords: Sustained Attention, Acetylcholine, Dendritic Remodeling, Chronic Stress

Disclosure: Nothing to Disclose.

T218. Combined Oxytocin and CBSST for Social Function in People With Schizophrenia

Robert Buchanan*, Deanna L. Kelly, Gregory P. Strauss, James M. Gold, Elaine Weiner, Robert P. McMahon, Jason Holden, Eric Granholm

Maryland Psychiatric Research Center, University of Maryland School of Medicine, Baltimore, Maryland, United States

Background: A significant proportion of people with schizophrenia are characterized by impaired ability to socially engage with others, which may reflect social aversion secondary to defeatist beliefs; decreased motivation for social interactions; and/or impairment in the normal reinforcement value of social interactions. Unfortunately, pharmacological interventions have limited benefits for impaired social function, whereas psychosocial interventions provide only partial benefit for this critical aspect of the illness. The development of an effective intervention for functional outcomes remains a central therapeutic challenge. The current study assessed whether the addition of intranasal

oxytocin to Cognitive Behavioral Social Skills Training (CBSST) improved social function.
Methods: Participants with DSM-IV-TR schizophrenia or schizoaffective disorder entered a 24-week, double-blind, placebo-controlled, randomized clinical trial with a 3-month follow-up evaluation. The study was conducted at two sites: Maryland and California. Participants were randomized to either: intranasal oxytocin 36 I.U. (3 sprays) BID ($n = 31$) or intranasal placebo-oxytocin 3 sprays BID ($n = 31$). All participants received CBSST, which was delivered in four 6-session modules (cognitive, social and problem-solving CBSST modules plus a module adapted from Social Cognition and Interaction Training). The modules were delivered in twice weekly 1-hour sessions, for a total of 48 sessions over 24 weeks. Birchwood Social Functioning Scale (BSFS) was used to assess social function; the Defeatist Performance Attitude Scale (DPAS) was used to assess defeatist beliefs; the Asocial Beliefs Scale (ABS) was used to assess social disinterest and aversion. The Brief Psychiatric Rating Scale (BPRS) total score was used to assess global psychopathology and the BPRS positive symptom items was used to assess positive symptoms; the Scale for the Assessment of Negative Symptoms (SANS) total score was used to assess negative symptoms; and the Calgary Depression Scale (CDS) total score was used to assess depressive symptoms. The primary analytic approach for the efficacy measures was a mixed model for repeated measures ANCOVA, adjusting for the baseline score.
Results: The primary outcome measure was the BSFS total score. There were no significant time ($F = 1.11$; $df = 1,49.3$; $p = 0.30$), treatment ($F = 1,44$; $df = 1,49.3$; $p = 0.24$), or treatment x time ($F = 1.44$; $df = 1,49.3$; $p = 0.24$) effects. The DPAS and ABS were used to assess the effect of CBSST on defeatist attitudes and asocial beliefs; there was a trend effect of time for the DPAS ($F = 3.17$; $df = 1,41.4$; $p = 0.08$). There were no significant effects for treatment ($F = 0.00$; $df = 1,43.9$; $p = 0.95$) or treatment x time ($F = 1.78$; $df = 1,41.4$; $p = 0.19$). There was also a trend effect of time for the ABS ($F = 3.16$; $df = 1,44.1$; $p = 0.08$). There were no significant effects for treatment ($F = 0.20$; $df = 1,47.2$; $p = 0.66$) or treatment x time ($F = 0.17$; $df = 1,44.1$; $p = 0.68$). The time effect for the SANS total score was not significant ($F = 0.86$; $df = 5,41.4$; $p = 0.52$). However, when the SANS items were divided into those related to emotional expression and avolition/asociality, there was a trend time effect for the avolition/asociality composite measure ($F = 2.44$; $df = 5,41.4$; $p = 0.05$). There were no significant treatment ($F = 0.11$; $df = 1,50.3$; $p = 0.75$) or treatment x time ($F = 1.10$; $df = 5,41.4$; $p = 0.37$) effects for SANS total score. There were significant time ($F = 3.61$; $df = 5,35.4$; $p = 0.01$) and treatment ($F = 5.06$; df+1,47.7; $p = 0.03$) effects for BPRS total score. The treatment x time effect was not significant ($F = 0.93$; $df = 5,35.4$; $p = 0.48$). There was also a significant treatment effect for BPRS positive symptom item score ($F = 4.45$; $df = 1,51.0$; $p = 0.04$). There was a trend for the time effect ($F = 2.31$; $df = 5,37.1$; $p = 0.06$); the treatment x time effect was not significant ($F = 2.26$; $df = 5,37.1$; $p = 0.48$). The time ($F = 0.69$; $df = 5,43.0$; $p = 0.63$) and treatment x time ($F = 0.50$; $df = 5,43.0$; $p = 0.77$) effects for CDS total score were not significant. However, there was a trend for the treatment effect ($F = 3.09$; $df = 1,51.8$; $p = 0.08$).
Conclusions: The study results suggest that there is no significant benefit of oxytocin for social function, as measured by BSFS total score. The observed time effects for the DPAS, ABS, and SANS avolition/asociality are consistent with the results from previous CBSST studies and suggest that CBSST was effective in the current study. The addition of oxytocin to CBSST did result in a significant reduction of global psychopathology and positive symptoms. There was no effect of oxytocin on negative symptoms; there was a trend effect for depressive symptoms. The observed effect on global psychopathology and positive symptoms is consistent with previous studies, which used high dose oxytocin. (clinicaltrials.gov, trial number: NCT01752712)
Keywords: Oxytocin, Cognitive Behavioral Therapy, Social Skills Training, Social Functioning, Schizophrenia
Disclosure: Part 1: Astellas, Advisory Board, Boehringer-Ingelheim-RCV, Advisory Board, ITI, Inc., Advisory Board, Lundbeck, Advisory Board, Takeda, Advisory Board, Upsher-Smith Laboratories, Inc., Consultant, Pfizer, Honoraria, Takeda, Consultant.

T219. FXR1-GSK3β Mediates Homeostatic Regulation of Neuronal Activity

Jivan Khlghatyan, Alesya Evstratova, Aleksandra Marakhovskaia, Valerie Mongrain, Katalin Toth, Jean-Martin Beaulieu*

University of Toronto, Toronto, Canada

Background: The fragile X mental retardation syndrome-related protein 1 (Fxr1) is a member of a small family of RNA binding proteins that also comprises the fragile X mental retardation protein 1 (Fmr1) and Fxr2. The neuronal functions of this family have mostly been studied in the context of autism spectrum disorders. However, recent GWAS have linked FXR1 to schizophrenia and bipolar disorders, therefore indicating its possible wider roles in mental illnesses. Furthermore, interaction between polymorphisms affecting cortical expression of the FXR1 and GSK3B genes, regulates emotion processing in healthy humans and affects depressive and manic dimensions in bipolar patients.
Methods: We used somatic CRISPR/Cas9 mediated knockout and overexpression to investigate the impact of Fxr1-Gsk3β mediated signaling on neuronal functions directly in the adult mouse brain.
Results: Suppression of Gsk3b and increase of Fxr1 expression in prefrontal cortex neurons lead to anxiolytic responses associated with decrease in AMPA mediated excitatory postsynaptic currents. Furthermore, Gsk3β-Fxr1 signaling contributes to the homeostatic regulation of synaptic strength in response to activity suppression in vitro and sleep deprivation in vivo.
Conclusions: These results underscore a potential mechanism underlying the action of Gsk3β-Fxr1 signaling on neuronal activity and plasticity. Association between homeostatic plasticity and Gsk3β-Fxr1 functions also suggests how this pathway can contribute to emotional regulation in response to environmental conditions, like sleep deprivation, or in illnesses like mood disorders and schizophrenia.
Keywords: Schizophrenia, Bipolar Disorder, Fragile X, Homeostatic, Sleep Deprivation
Disclosure: Nothing to Disclose.

T220. Differential Deficits in Early Visual Processing Across Neuropsychiatric Populations: Implications for Screening and Intervention

Antigona Martinez*, Pablo Gaspar, Steven Hillyard, Cheryl Corcoran, Yaakov Stern, Daniel Javitt

Nathan S Kline Institute for Psychiatric Research, Orangeburg, New York, United States

Background: Abnormal visual processing is a feature of several psychiatric disorders including schizophrenia, autism and neurodegenerative disorders. However, no standardized visual stimulation paradigms are available to evaluate cross-diagnostic differences. Here we report on results from a highly efficient visual stimulation paradigm that can assess three different components of visual processing in parallel: 1) the stimulus-onset visual P1 potential, 2) motion-induced N2m potential and 3) the steady state visual evoked response (ssVEP). To avoid habituation, the onset and motion stimuli are presented at long intervals but the intervening time period is used for steady-state stimulation. To evaluate differential deficits in magnocellular versus parvocellular pathway function, the gratings are composed of either low (LSF) or high (HSF) spatial frequencies and are of high and low luminance contrast. Time-frequency analyses and parallel fMRI studies were used to evaluate potential underlying pathophysiological mechanisms.
Methods: The paradigm was used to acquire electrophysiological indices of early-stage visual and motion processing in 250 subjects comprising eight different cohorts: 1) patients with diagnosed schizophrenia (SzP) 2) clinically high-risk subjects (CHR) 3) individuals with autism spectrum disorder (ASD) 4) healthy elders with negative cortical beta amyloid deposition (amyl-) 5) healthy elders positive for amyloid deposition (amyl+) 6) subjects diagnosed with mild cognitive impairment (MCI) 7 and 8) middle-age healthy controls (HCm; matched to SzP) and young healthy controls (HCy; matched to CHR). Stimuli were grayscale vertical sinusoidal gratings, either LSF (0.8 cpd) or HSF (5 cpd), with either a low (4%) or high (75%) luminance contrast presented against an isoluminant gray background and subtending 2 x 2 degrees of visual angle. The lower edge of all stimuli occurred 1 of visual angle above a central fixation cross. Each trial lasted 4900 ms and began with a 500 ms presentation of the central cross. Following, one of the three stimuli was presented randomly and remained static for 400 ms. The grating then drifted rightward at a velocity of 14 degrees/s for 200 ms. After an 800 ms delay during which the grating was static, the grating counterphase reversed at a rate of 10 Hz for 3s. During the entire trial, subjects' task was to fixate on a central cross and press a button when the crosshair dimmed slightly (every 3-12 s).
Results: As expected, SzP showed robust deficits in response to the onset of all stimuli, especially LSF stimuli. Likewise, relative to similar-aged controls, SzP showed significantly reduced responses to both the motion onset and steady-state flicker of the stimuli. CHR subjects, on the other hand, had normal responses to the onset and steady-state of all stimuli but had highly impaired motion-onset responses. A different pattern was observed in ASD subjects, who had highly increased responses to stimulus onset and steady-state together with reduced motion responses. Finally, all 3 visual processing measures were reduced in amyloid+ healthy elders and MCI subjects compared to amyloid-negative comparison subjects. The magnitude of the responses elicited by the onset and motion of all stimuli correlated with subjects' amyloid status.
Conclusions: Deficits in early visual processing are observed across neuropsychiatric populations, with patterns selective to each population. In general, SzP show reduced visual responses across all visual conditions, with greater deficits to magnocellular-biased LSF stimuli, while CHR individuals show reduced responses only to motion stimuli. In contrast to the psychosis-spectrum populations, similar-age high-functioning ASD subjects show enhanced responses within the same experimental paradigm. In aging, reductions in visual processing are observed equally across LSF and HSF stimuli and correlate with amyloid status, suggesting potential utility in screening, early detection, and early intervention.
Keywords: Visual Information Processing, Event-Related Potentials, Schizophrenia, Neuropsychiatric Disorders
Disclosure: Nothing to Disclose.

T221. Discrepancies Between Self-Assessment of Disability and Objective Measures of Everyday Functioning in Schizophrenia and Bipolar Disorder

Marc Kalin*, Martin Strassnig, Laura Fochtmann, Kotov Roman, Evelyn Bromet, Phil Harvey

School of Medicine and Public Health University of Wisconsin, Miami, Florida, United States

Background: Self-assessments are frequently used to index everyday functioning in individuals with severe mental illnesses, but these reports may not coincide with objective measures of functioning. The purpose of this study is to understand the relationship between subjective and objective assessments of disability, as well as cognitive and physical performance, in individuals with schizophrenia and bipolar disorder with psychotic features.
Methods: The sample consisted of participants from the Suffolk County Mental Health Project, a cohort with first-admission psychosis. We examined 20-year follow-up of 146 participants with schizophrenia and 87 with bipolar disorders. We compared self-assessment of disability (WHODAS 2.0) with interviewer-ascertained everyday functioning indexed by achieving independence in residence and gainful employment. We also measured symptoms and indexed cognitive performance and physical capacity with performance-based measures.
Results: Compared to schizophrenia, bipolar disorder participants evaluated themselves as globally less disabled and they were more likely to have achieved current milestones. Unemployed patients reported more impairment on the WHODAS, but there were no differences in WHODAS scores as a function of independence in residence. WHODAS scores were correlated with clinical symptoms and physical functioning, but not cognitive test performance. Everyday functioning was associated with cognitive test performance and physical functioning but not with clinical symptoms.

Conclusions: Patients with schizophrenia and bipolar disorder did not differ in their ratings of everyday disability as a function of living independently, and their self-reports of disability were more linked to symptoms than to objective predictors of the skills required for everyday independence. These data suggest that self-reports of everyday functioning may be recapturing symptoms more than the factors actually predictive of reduced everyday functioning.
Keywords: Schizophrenia, Bipolar, Functional Outcomes
Disclosure: Nothing to Disclose.

T222. A Schizophrenia-Associated Missense Mutation in Kalirin Alters Pyramidal Cell Morphology in a Mouse Model

Melanie Grubisha*, Gregg Homanics, Peter Penzes, Robert Sweet

University of Pittsburgh Medical Center, Pittsburgh, Pennsylvania, United States

Background: Kalirin (KAL) is a Rho GEF that is highly involved in regulation of cytoskeletal morphology within dendrites. A missense mutation (P2255T, KAL-PT) in the KAL9 and KAL12 isoforms has been associated with schizophrenia. KAL coordinates RhoA activation downstream of p75 when p75 interacts with Nogo receptor (NGR). This NGR/p75/KAL complex acts to restrict dendritic morphogenesis. I have found that KAL-PT acts as a gain of function mutation, increasing RhoA activity when expressed in vitro and impairing dendritic morphogenesis. We hypothesize that enhanced activation of the NGR/p75/KAL pathway during early development downregulates the expression of microtubule transport proteins and subsequently impairs dendritic morphogenesis across development.
Methods: A humanized mouse model of the KAL-PT mutation was created using CRISPR/Cas9 genome editing. Frontal pole homogenate was collected from wild-type (KAL-WT) and KAL-PT mice and RNAseq was performed on an Illumina HiSeq platform. Differential gene expression was calculated and pathway analysis was performed using BaseSpace Correlation engine. In vitro morphological studies were performed on DIV8 neurons grown from dissociated cortical cultures derived from KAL-WT, KAL9 heterozygous (KAL-het), and KAL-PT P0 mouse pups. Sholl analysis was performed using NeuroLucida software.
Results: Frontal pole cortex from KAL-PT CRISPR/Cas9 mice shows significant overlap with an existing dataset generated from Nogo-A overexpression, and this overlap is enriched for downregulation of molecules involved in microtubule transport. In vitro studies demonstrate decreased dendritic branching and length in pyramidal neurons from KAL-PT DIV8 cortical cultures compared to KAL-WT. KAL-het show an intermediate phenotype.
Conclusions: The PT mutation results in perturbation of signaling to the cytoskeleton, with enrichment for disruptions in microtubule transport proteins. These signaling pathway perturbations may underlie some of the observed functional impairments seen in pyramidal cells in schizophrenia, specifically decreased dendritic length and complexity. Using a disease-associated mutation to model convergent pathway perturbations involved in cytoskeleton remodeling may aid the development of novel pharmacotherapeutics for schizophrenia.
Keywords: Schizophrenia, Animal Model, Kalirin
Disclosure: Nothing to Disclose.

T223. 18-Year Course of Cognitive Functioning in Psychotic Disorders

Eva Velthorst*, Anne-Kathrin Fett, Avi Reichenberg, Camilo Ruggero, Jennifer Callahan, Evelyn Bromet, Roman Kotov

Icahn School of Medicine at Mount Sinai, New York, New York, United States

Background: There is an ongoing debate about the progressive nature of cognitive deficits in psychotic disorders. This study therefore aimed to examine the long-term changes in cognitive functioning after first hospitalization with psychosis.
Methods: Data came from the Suffolk County Mental Health Project, a prospective study of first-admission patients with psychotic disorders. Cognitive tests were administered 2 years (n = 399; schizophrenia spectrum: 285, affective psychoses: 226, and other psychoses: 117) and 20 years (n = 240; 115, 92, and 34, respectively) after first admission, with 195 individuals completing cognitive tests both times. A never psychotic comparison group ($N = 260$) was assessed at year 20.
Results: Cases with schizophrenia spectrum disorders generally showed lower cognitive functioning than cases with affective and other psychoses at 2- and 20-year points. Almost all test indicated a decline in cognitive performance over time (~0.50 standard deviation), and it was comparable in magnitude across diagnostic groups. The most consistent predictors of larger decline were lower childhood IQ and SES. At 20-year, cases showed greater cognitive impairment than age-matched never psychotic participants and this difference was especially pronounced in older participants.
Conclusions: Our findings indicate that cognitive functioning in psychotic disorders continues to decline after the illness begins and that this decline is not specific to schizophrenia but present across psychotic disorders. The results tentatively suggest premature aging in some cognitive domains and call attention to persistent and, in fact, worsening cognitive problems in psychotic disorders. Thus, public policy goals for these conditions should not only include cognitive remediation but also prevention of premature cognitive decline.
Keywords: Cognition, Psychotic Disorders, Longitudinal
Disclosure: Nothing to Disclose.

T224. Can Presynaptic and Postsynaptic Monoaminergic Mechanisms Work Synergistically? Current Findings and Future Directions

Dimitri Grigoriadis*, Sam R.J. Hoare, Andrea Kudwa, Steve Boikess

Neurocrine Biosciences, Inc., San Diego, California, United States

Background: Most antipsychotic medications primarily target the dopaminergic system through inhibition of

postsynaptic D2 dopamine receptors, but decreasing the activities of other monoamines (e.g., serotonin and its receptor network) may also play a role in reducing psychosis or other psychiatric symptoms. The vesicular monoamine transporter 2 (VMAT2) is an integral membrane protein that packages presynaptic cytosolic monoamines, including dopamine, into vesicles for synaptic release. Despite having different mechanisms of action, antipsychotics and VMAT2 inhibitors can both act to decrease activity in central dopaminergic systems. Therefore, under conditions where hypermonoaminergic mechanisms play a major role and antipsychotic treatment is indicated (e.g., patients with schizophrenia who require ongoing antipsychotic treatment to maintain psychiatric stability), concomitant treatment with a VMAT2 inhibitor may offer the possibility of using lower antipsychotic doses while maintaining full efficacy. Selective VMAT2 inhibitors may offer additional benefit in patients with psychiatric illness who require the use of an antipsychotic for psychiatric treatment.
Methods: The primary active metabolite of valbenazine, a selective VMAT2 inhibitor recently approved for the treatment of tardive dyskinesia in adults, [+]-α-dihydrotetrabenazine ([+]-α-HTBZ), was evaluated using two well-established in vivo animal models to assess effects of [+]-α-HTBZ on sensorimotor gating and antipsychotic-like behavior when administered either independently or concomitant with typical (haloperidol) or atypical (risperidone and olanzapine) antipsychotics. Prepulse inhibition in mouse assessed effects on sensorimotor gating while the conditioned avoidance task in rats (CAR) was used to evaluate potential antipsychotic efficacy directly related to dopaminergic neuromodulation.
Results: At doses equivalent to therapeutic plasma concentrations in humans, both [+]-α-HTBZ (10 mg/kg) and haloperidol (1 mg/kg) similarly enhanced prepulse inhibition: haloperidol (1 mg/kg), 53%; [+]-α-HTBZ (10 mg/kg), 53%; both $P<0.001$ vs. DMSO and sterile water controls, respectively. The CAR task revealed that [+]-α-HTBZ significantly reduced the conditioned avoidance response ($P<0.001$ vs. vehicle) in a manner similar to both risperidone and olanzapine; at subthreshold doses, all 3 drugs demonstrated minimal efficacy (risperidone 0.10 mg/kg, olanzapine 0.60 mg/kg, [+]-α-HTBZ 0.15 mg/kg). However, when [+]-α-HTBZ (0.15 mg/kg) co-administered with risperidone (0.10 mg/kg), the combined effect was approximately 5-10 times greater than that of either drug alone ($P<0.001$ versus vehicle); a comparable synergistic effect was found when [+]-α-HTBZ (0.15 mg/kg) was dosed with olanzapine (0.60 mg/kg). Moreover, the combined effects of two subthreshold [+]-α-HTBZ doses (0.15 mg/kg and 0.34 mg/kg) administered across a range of risperidone doses (0.03 to 1 mg/kg) resulted in a leftward shift of the risperidone dose-response curve thus indicating that the synergistic effect of these two drugs on the avoidance response was dose-dependent.
Conclusions: In vivo studies using classical rodent models sensitive to typical and atypical antipsychotics demonstrated that a selective VMAT2 inhibitor alone can elicit antipsychotic-like behavioral profiles. Furthermore, co-administration of subthreshold doses of the selective VMAT2 inhibitor, [+]-α-HTBZ, with subthreshold doses of commonly prescribed atypical antipsychotics such as risperidone and olanzapine also resulted in full antipsychotic efficacy, thereby indicating a synergistic effect between the 2 different drug classes. These findings suggest that concomitant modulation of presynaptic and postsynaptic monoaminergic function may have important implications in clinical settings where first-line or adjunctive psychiatric therapies require the use of typical or atypical antipsychotics and may reduce the incidence of known dose-related adverse side-effects while maintaining their therapeutic potential.
Keywords: VMAT2, in vivo, Metabolites
Disclosure: Part 2: Neurocrine Biosciences, Inc., Employee, **Part 5:** Neurocrine Biosciences, Inc., Employee.

T225. Evidence for Sleep as a Modifiable Risk Factor for PTSD

Dewleen Baker*, Dean Acheson, Victoria Risbrough, Caroline Nievergelt, Brian Kwan, Adam Maihofer, Xin Tu

University of California, San Diego, La Jolla, California, United States

Background: The Marine Resiliency Study (MRS) is a prospective, longitudinal study of approximately 2,600 Marines deploying to Iraq or Afghanistan, implemented to assess risk/resilience in a healthy, young cohort of service members exposed to deployment stress. Previously published MRS findings show pre-deployment physiology, e.g., autonomic tone and immune status to be post-deployment PTSD risk predictors. Given that autonomic tone and immune status are associated with sleep which is a potentially modifiable behavior, we sought to determine if pre-deployment sleep quality was a risk indicator for development of trauma-related PTSD after a combat deployment.
Methods: Marines and accompanying Navy personnel were assessed during training at pre-deployment, and followed-up after a 7-month combat deployment, at three and six months post-deployment. Measurement of a broad array of behavioral, psychosocial, physiological and blood-based biomarker data were obtained from all consenting participants at all three time points. We employed structural equation models to examine longitudinal relationships between self-reported indicators of pre-deployment sleep quality in participants without PTSD and post-deployment PTSD re-experiencing symptoms.
Results: Pre-deployment sleep quality consistently predicted levels of PTSD re-experiencing symptoms at the 3 and 6-month post-deployment timeframes. In contrast, PTSD re-experiencing symptoms were not predictive of future sleep quality. This pattern of results suggests a potential causal effect of poor sleep on development of PTSD symptoms following deployment.
Conclusions: Based on the prospective, longitudinal study design of MRS, insights into pre-existing physiology predictive of mental health risk/resilience were obtained. We now show that a modifiable behavior, sleep may contribute to post-deployment risk/resilience.
Keywords: Sleep Disturbance, PTSD, Early Identification of Risk
Disclosure: Nothing to Disclose.

T226. Sexually Dimorphic Response in Corticosterone and Brain Kynurenic Acid After Acute Sleep Deprivation in Rats

Annalisa Baratta, Austin Buchla, Silas Buck, Ana Pocivavsek*

Maryland Psychiatric Research Center, University of Maryland School of Medicine, Catonsville, Maryland, United States

Background: Inadequate sleep is a prevalent problem within our society. Cognitive dysfunction is one of the recurring consequences of sleep loss that impacts daily function and there is a compelling need to further understand the underlying molecular mechanisms linking sleep loss and cognitive impairments. Sleep loss-induced stress has been shown to activate the hypothalamic-pituitary-adrenal (HPA) axis and influence several biological systems. As elevations in the HPA axis secreted hormone corticosterone (CORT) can initiate tryptophan metabolism via the kynurenine pathway (KP), our current experiments were designed to test the hypothesis that kynurenic acid (KYNA), an astrocyte-derived metabolite of the KP, is elevated in the brain after sleep deprivation (SD). KYNA is an antagonist of α7 nicotinic acetylcholine (α7nACh) and NMDA receptors, and elevations in KYNA negatively impact learning and memory.
Methods: As sleep patterns are sexually dimorphic, we investigated the effect of sleep deprivation (SD) on KP metabolism and CORT in both male and female adult Wistar rats. To examine the role of circulating gonadal hormones, separate cohorts of adult male and female rats were gonadectomized (GDX), respectively removing the testes and ovaries at least 10 days prior to experiments. Animals were sleep deprived by gentle handling for 6 h from Zeitgeber time (ZT) 0 to ZT 6, where ZT 0 is the start of the light-phase. Plasma and brains were collected from control and SD animals the day of the experiment at ZT 6.
Results: Levels of the stress response hormone CORT were significantly impacted by SD ($F_{1,32} = 11.4$, $P < 0.01$) and sex ($F_{1,32} = 9.2$, $P < 0.01$). CORT was elevated by 130% in SD females compared to control females. Male animals, however, did not have significantly higher CORT after SD. KYNA levels in the hippocampus, a region critically involved in learning and memory, were impacted by both SD ($F_{1,26} = 8.4$, $P < 0.01$) and sex ($F_{1,26} = 7.4$, $P < 0.05$). There was a 140% increase in hippocampal KYNA levels in male rats after SD ($P < 0.05$), but there was no significant elevation in female rats. Irrespective of sex, brain levels of the KP metabolite 3-hydroxykynurenine (3-HK) remained unchanged after SD. In the plasma, no significant changes in KYNA or its bioprecursor kynurenine were observed after SD. As our results demonstrate a striking sexual dimorphism in the response to SD, we hypothesized a role of circulating gonadal hormones and conducted subsequent experiments in adult GDX or sham-operated male and female rats. In male animals, CORT was significantly impacted by SD ($F_{1,39} = 6.6$, $P < 0.05$) and surgery ($F_{1,39} = 18.1$, $P < 0.0001$), and post-hoc analysis revealed a significant 85% elevation in plasma CORT only in GDX males after SD. In females, SD significantly elevated CORT ($F_{1,33} = 29.1$, $P < 0.0001$) in both sham-operated (+88%) and GDX (+162%) rats. Analysis of KYNA in the hippocampus revealed that upon removal of circulating androgens, KYNA levels were not elevated in GDX male animals after SD and only sham-operated males had significantly higher hippocampal KYNA compared to counterpart controls. SD did not significantly impact brain KYNA levels in GDX or sham-operated females. Interestingly, we found a sexually dimorphic relationship between peripheral CORT and brain KYNA after SD. In female SD animals, plasma CORT correlated negatively with hippocampal KYNA ($r = -0.39$, $P < 0.05$), while the relationship was not significant in male SD animals ($r = -0.08$, $P = 0.69$).
Conclusions: Taken together, we demonstrate a sexually dimorphic response to SD and an inverse relationship between plasma CORT and hippocampal KYNA in female animals. The interplay of this relationship may reveal sex-specific mechanisms that impact hippocampal learning and memory in response to sleep loss. Our futures studies are investigating the impact of acute SD on cognitive performance in rats and testing our hypothesis that KYNA elevations mediate memory impairments after SD. We are using a KYNA synthesis inhibitor, BFF-816, that targets the enzyme kynurenine aminotransferase (KAT) II and prevents de novo KYNA production. Unraveling the contribution of KYNA, a novel molecular target that is influenced by SD, may lead to the development of new therapeutic agents to improve cognitive outcomes after sleep loss.
Keywords: Sleep Deprivation, Kynurenine Pathway, Sex Difference, Hippocampus, Corticosterone
Disclosure: Nothing to Disclose.

T227. The Effects of Poor Sleep Quality on Brain Structure and Cognition are Greater in HIV+ Individuals

April Thames*, Zanjbeel Mahmood, Andrea Hammond, Michael Irwin

University of California, Los Angeles, Los Angeles, California, United States

Background: The adverse effects of sleep disturbances have been well-documented in healthy populations, with studies associating it with greater likelihood of developing cognitive impairment (Fortier-Brochu et al. 2012; Joo, et al., 2014) and dementia in older adults (Chun, et al., 2012). Cognitive impairments are supported by findings from morphometric brain imaging studies, although findings are mixed with regard to positive and negative associations with regional volume of particular structures (Altena et al., 2010; Begre et al., 2008; Joo et al., 2013; Riemann et al., 2007; Winkleman et al., 2013). An estimated 30-70% of people living with HIV (PLWH) report sleep disturbances (Rubinstein et al., 1998; Prenzlauer et al., 1993) compared to 10% - 30% of those in the general population (National Sleep Foundation, 2005). Specifically, PLWH report greater daytime sleepiness, fatigue and overall poor sleep quality (Low et al., 2011; Wibbeler et al., 2012). Given the prevalence of cognitive impairment in individuals with HIV (Heaton et al., 2011), there are legitimate concerns regarding the cognitive consequences of sleep disturbance in this population.
The purpose of the current study was to characterize HIV+ and HIV- individuals based upon their self-reported sleep

quality (SQ) and to examine the effects of SQ on cognitive functioning. We hypothesized that the HIV status groups would significantly differ on self-reported SQ, with HIV+ individuals reporting worse SQ as compared to controls. Furthermore, we expected that the adverse effects of poor SQ on cognitive performance and brain structure would be greater in the HIV+ group.
Methods: Ninety-seven HIV+ (n = 58) and HIV- (n = 39) participants were recruited from HIV clinics and the local community through local advertisements. Questionnaires and screeners about medical, neurological and psychiatric history were used to screen for potential confounds (see Thames et al., 2017). Two-step cluster analysis was used to classify participants into distinct HIV/SQ groups: HIV-positive/good sleep quality (HIV+/SQ+; n = 34), HIV-negative/good sleep quality (HIV-/SQ+; n = 24), HIV-negative/poor sleep quality (HIV-/SQ- ;n = 13), and HIV-positive/poor sleep quality (HIV+/SQ- ; n = 26), based upon their responses to the SATED scale (Buysse, 2014), which measures sleeping patterns within the last 30 days. Participants were administered a brief cognitive test battery used in prior studies (Thames et al., 2017; 2015). Cognitive domain-specific composite score was calculated by averaging t scores from individual cognitive tests (Heaton et al., 1991; Miller and Rohling, 2001). All participants underwent MRI to examine secondary outcomes of brain structure.
Results: HIV status groups did not significantly differ in age, education, estimated premorbid IQ, depression scores and race/ethnicity (p's $>$ 0.10). However, there were significant differences in gender ($\chi 2$ (2) = 8.53, p = .01) with significantly greater proportion of males and transgendered (male-to-female) participants in the HIV+ group compared to controls. We did not observe that gender was significantly related to SQ and cognitive outcome variables of interests, and it did not significantly differ between the HIV/SQ groups (all p's $\geq$ 0.10). No other significant HIV/SQ group differences were found. All HIV+ participants were on a stable regimen of cART and were clinically stable based upon current plasma CD4 count, and 76.8% had undetectable viral load (i.e., $<$ 20 copies/mL). There was a significant difference between the HIV status groups on SQ, with the HIV+ group reporting poorer SQ compared to controls [t (95) = -2.08, p = 0.04].
COGNITION: There were significant HIV group differences in global cognition, with the HIV+ group demonstrating lower global cognitive scores than HIV- participants (p = .04). SQ was positively correlated with domains of executive functioning, learning and memory (all p's $<$ 0.05). There were statistically significant difference between the HIV/SQ groups on global neurocognitive performance [F (3, 93) = 3.08, p = 0.03], with the HIV+/SQ- group exhibiting lowest performance compared to all other groups. Domains of learning [F (3, 93) = 3.89, p = .01, $\eta 2$ = .11] and memory [F (3, 93) = 5.58, p = .001, $\eta 2$ = .15] contributed to this relationship.
BRAIN IMAGING: Significant HIV group differences were found in gray matter thickness in regions of the insula, cingulate, lateral orbital cortex, and left middle temporal gyrus (all p's $<$ .01), such that the HIV+ group demonstrated reduced thickness in these regions. SQ was negatively associated with medial orbitalfrontal volume (r = -.26, p = .03), brain stem volume (r = -.29, p = .01), thalamic volume (r = -.27, p = .02), and DTI mean diffusivity (MD) in the superior longitudinal fasciculus of the temporal lobe (r = -.27, p = .02). The HIV+/SQ- group demonstrated greater MD in the superior longitudinal fasciculus of the temporal lobe in comparison to the other HIV/SQ groups. The HIV+/SQ- group also demonstrated greater volume in regions of the brain stem, thalamus and nucleus accumbens.
Conclusions: While HIV status and sleep disturbance show independent associations with cognition and brain structure, results from this preliminary investigation show that HIV+ individuals who report poor sleep quality are at higher risk for cognitive impairment and neurological abnormalities than their uninfected counterparts.
Keywords: Sleep Disturbance, HIV, Brain Structure, Cognition
Disclosure: Nothing to Disclose.

T228. Subcortical Beta-Amyloid Burden is Independently Linked to Sleep History and Genetic Risk Factors of Alzheimer's Disease

Ehsan Shokri Kojori*, Gene-Jack Wang, Corinde Wiers, Sukru Demiral, Min Guo, Sunny Kim, Peter Manza, Elsa Lindgren, Gregg Miller, Veronica Ramirez, Clara Freeman, Amna Zehra, Tansha Srivastava, Susan De Santi, Dardo Tomasi, Helene Benveniste, Nora Volkow

National Institute on Alcohol Abuse and Alcoholism, Bethesda, Maryland, United States

Background: Sleep history and APOE-genotype are of the two factors associated with beta-amyloid burden (ABB) in the human brain (Spira et al., 2013 & Coon et al., 2007), leading to increased risk for Alzheimer's disease (AD). While there are indications that sleep history and APOE-genotype interact in their association with ABB (Lim et al., 2013 & Drogos et al., 2016), others have reported that their contribution is independent (Brown et al., 2016). These observations lead to the hypothesis that ABB in AD-prone brain regions (such as the hippocampus and precuneus) could be independently linked to sleep history and genetic risk for AD.
Methods: Healthy participants (n = 20, age: 22-72, 10 females) underwent PET imaging using 18F-florbetaben (FBB) radioligand after one night of rested sleep. T1w and T2w anatomical images were also collected both with 0.8-mm isotropic resolution. Images were processed in FreeSurfer and FSL for segmentation, co-registration, and normalization to the MNI space. FBB images were converted to relative standard uptake value (SUVr) using the whole cerebellum as reference region (Bullich et al., 2017). We also administered the Pittsburgh Sleep Quality Index questionnaire to evaluate subjective experience of sleep duration. Finally, APOE genotyping was performed for rs7412 and rs429358 SNPs to estimate an odds ratio for AD (ORAD) for each participant (Corneveaux et al., 2010).
Results: In our participants, ORAD was not correlated with self-report of sleep duration (p $>$ 0.6). Voxelwise correlation between sleep duration and FBB SUVr identified 2 bilateral subcortical clusters and one cluster within the precuneus, where more sleep duration was associated with less FBB SUVr (PFWE $<$ 0.05). The subcortical clusters included the lentiform nucleus, putamen, and parahippocampus in both

hemispheres. Higher ORAD was associated with higher FBB SUVr in the bilateral subcortical clusters that, similarly, included lentiform nucleus, putamen, and parahippocampus, but also the globus pallidus (PFWE < 0.05). In addition, higher ORAD was associated with increased FBB SUVr in the superior medial frontal gyrus (SMFG). Average FBB SUVr in the left and the right subcortical clusters (common between both analyses), each negatively correlated with sleep duration ($R2 > 0.36$, $p < 0.005$) and positively correlated with ORAD ($R2 > 0.25$, $p < 0.025$). However, average FBB SUVr in the precuneus cluster did not correlate with ORAD and average FBB SUVr in the SMFG did not correlate with sleep duration ($p > 0.5$).

Conclusions: Our data indicate that despite lack of significant association between ORAD and sleep duration in our participants, higher ABB in identical subcortical structures was associated with both higher ORAD and less sleep duration. However, ABB in the precuneus and SMFG were uniquely linked to ORAD and sleep duration, respectively. While emerging research suggest a bidirectional association between ABB and sleep history, poor sleep may also result from lifestyle choices. Our observations highlight that convergence of risk factors in affecting ABB in identical subcortical areas may contribute to higher susceptibility of these regions to AD-related pathology. In contrast, other AD-related structures, such as the precuneus and SMFG, were associated with distinct risk factors of AD. In summary, the findings provide new insights into how different AD risk factors could differently contribute to regional pathology of AD.

Keywords: Sleep, Beta-Amyloid Peptide 1-42, APOE, Hippocampus, [18-F] florbetaben

Disclosure: Nothing to Disclose.

T229. Viral-Mediated Rescue of Arc/Arg3.1 Knock-Out Demonstrates a Requirement for Function in the NAC in Regulating Mood and Drug-Related Behaviors

Rachel Penrod-Martin*, Laura Smith, Jaswinder Kumar, Brandon Hughes, Morgane Thomsen, Jakub Jedynak, Gabriella Barry, Daniel Wood, Makoto Taniguchi, Christopher Cowan

Medical University of South Carolina, Charleston, South Carolina, United States

Background: A key feature of drug addiction is the persistent vulnerability to relapse, despite long periods of abstinence. This vulnerability is conferred by long-lasting, drug experience-induced plasticity in key brain regions such as the mesocorticolimbic system. One key locus for these drug-induced changes is glutamatergic plasticity in the nucleus accumbens (NAc). Despite increasing evidence of functional consequences for drug-dependent changes in medium spiny neuron (MSN), the primary neuronal subtype of the NAc, the molecular mechanisms underlying these changes remain largely unknown. A clear understanding of the molecular regulation and behavioral consequences of altered glutamatergic signaling will be key for the development of new treatment strategies for drug addiction. The immediate early effector gene, activity-regulated cytoskeleton-associated protein (Arc/Arg3.1), is a known regulator of α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptor (AMPAR) surface expression, both through the promotion of endocytosis and the regulation of expression of the AMPAR subunit GluA1. Arc/Arg3.1 mRNA and protein are rapidly upregulated by acute cocaine exposure in the NAc, and Arc/Arg3.1 is critical for plasticity associated with long-term memory in other brain regions. Given that AMPAR regulation is a key site of cocaine-induced glutamatergic plasticity, Arc/Arg3.1 has potential to be a molecular contributor to this plasticity. We discovered that Arc/Arg3.1-deficient mice (KO) display several cocaine- and mood-related behavioral phenotypes, which implicate Arc/Arg3.1 as a negative regulator of these processes. Arc/Arg3.1-deficient mice show decreases in anxiety and increased locomotor responses to cocaine, but perhaps more interestingly, they show drug-experience dependent "reward sensitization". We find that prior cocaine experience appears to enhance cocaine reward in non-contingent (conditioned place preference; CPP) and contingent (intravenous drug self-administration) assays. While these behavioral phenotypes are likely linked to Arc/Arg3.1's regulation of AMPAR surface expression and/or GluA1 transcription, it is unclear what phenotypes are developmentally derived and which are dependent on Arc/Arg3.1's function in the adult brain. Additionally, it is not yet clear where Arc/Arg3.1 is functioning in the brain to regulate any of these behaviors.

Methods: Here, we examine the effects of viral-mediated, region-specific expression of Arc/Arg3.1 in KO and WT mice in anxiety and cocaine-related behaviors. Adult Arc/Arg3.1 KO mice received viral-mediated over-expression of Arc/Arg3.1 bilaterally in the NAc. Following surgical recovery and viral-mediated Arc/Arg3.1 expression, the mice were examined for performance on the elevated plus maze, locomotor sensitization, and CPP tasks.

Results: We find that Arc/Arg3.1 expression within the NAc of Arc/Arg3.1 KO mice rescues some, but not all, of these behavioral phenotypes. Preliminary findings indicate that Arc/Arg3.1 over-expression in the NAc of KO mice restores WT levels of open arm time on the elevated plus maze. Although still preliminary, it appears that long-term expression of Arc/Arg3.1 in the adult NAc of Arc/Arg3.1 KO mice also rescues drug-experience dependent "reward sensitization" in the cocaine CPP assay. However, Arc/Arg3.1 expression in the adult NAc of Arc/Arg3.1 KO mice fails to rescue enhanced cocaine locomotor sensitization. Current studies seek to complement these "rescue" studies using targeted loss of function approaches (i.e. shRNA and CRISPR/Cas9) in the adult NAc.

Conclusions: Overall, our results are consistent with a role for Arc/Arg3.1 in the NAc in regulating some of the phenotypes observed in the total Arc/Arg3.1 KO animals. Given the incomplete rescue of drug-related phenotypes, it is likely that some behaviors are mediated either by alternate brain regions or developmental effects. Interestingly, our preliminary findings dissociate Arc/Arg3.1 activity in the plasticity underlying locomotor sensitization and that contributing to reward sensitization observed in the CPP test, supporting the idea that sensitization and CPP behavior are not necessarily related to one another. To more clearly define the role of Arc/Arg3.1 in NAc-mediated drug-induced plasticity, future work will focus on spatially and temporally restricted knockdown of Arc/Arg3.1 and consequent behavioral and cell biological phenotypes.

Keywords: Cocaine, Molecular Mechanisms, Nucleus Accumbens, Drug Addiction
Disclosure: Nothing to Disclose.

T230. Mitochondrial Transcriptome and Epigenetic Changes in the Human Hippocampus Chronically Exposed to Cocaine

Yon Woo Jung*, Zhifeng Zhou, Qiaoping Yuan, Deborah C. Mash, David Goldman

National Institutes of Health/National Institute on Alcohol Abuse and Alcoholism/LNG, Bethesda, Maryland, United States

Background: Hippocampus is a brain region involved in addiction and drug-associated learning and memory. In hippocampus, long-term adaptation requires chromosome remodeling and gene expression changes. Several recent studies have shown that the functions of mitochondria are related to dendritic arborization and spine formation. Previously, we found that oxidative phosphorylation related genes were differentially expressed in the hippocampus of chronic cocaine addicts. Here, we have performed a deeper analysis of the changes in expression of mitochondria genes, both oxidative phosphorylation genes and genes encoding mitochondria structure regulating proteins as well as genes in other gene ontology and pathways, to better understand the nature and origin of altered mitochondrial function in chronically cocaine addicts.
Methods: Using the next generation sequencing technology, we re-examined gene expression changes and histone methylation changes in hippocampal region from chronically cocaine exposed individuals, cocaine addicts with excited delirium status, and carefully matched drug free control group. Eight chronic cocaine addicts, six cocaine addicts with excited delirium, and eight drug-free control subjects were selected for the study. Each hippocampal tissue sample was carefully matched for age, ethnicity, and postmortem interval and RNA quality was high as previously reported in detail. The quality of FastQ files for each sample were assessed using FastQC. Next, we aligned each reads from samples to the hg19 human reference transcriptome using Tophat2. We next generated individual sample count matrices using HTSeq-Count with Human GENCODE V19 annotation GTF file. HTSeq count files for each sample were imported into a created EdgeR and performed EdgeR analysis.
Results: Among the 23,336 hippocampal expressed genes, 1352 genes with an adjusted p value lower than 0.05 were differentially expressed in the chronic cocaine addicts and 486 genes were differentially expressed in the cocaine addicts with excited delirium. We subjected the differentially expressed genes with FDR lower than 0.05 to Gene Ontology (GO) and pathway analysis to detect molecular and cellular functional domains impacted by chronic cocaine exposure. We observed a significant effect of long-term cocaine exposure on genes involved in mitochondrial inner membrane functions and oxidative phosphorylation. Interestingly, the cocaine-mediated H3K4me3 histone methylation changes were also observed in mitochondrial genes responsible for ATP synthesis, electron transport, maintenance of mitochondrial membrane potential, and defense against oxidative stress.
Conclusions: Using RNA-Seq and ChIP-Seq for histone H3 lysine 4 trimethylation (H3K4me3), we identified hippocampal gene expression changes in both cocaine-addicted and cocaine-addicted with excited delirium individuals. We observed gene expression changes that were related to decrease mitochondrial inner membrane functions, oxidative phosphorylation, and energy metabolism. These mitochondrial changes are also observed in neurodegenerative diseases such as Alzheimer and Parkinson's disease. Histone H3K4me3 changes in mitochondria associated gene regions were also identified in chronic cocaine addicted and excited delirium samples. This implies possible correlations between cocaine related histone H3k4me3 and gene expression changes in mitochondria genes. These results indicate this organelle may plays an important role in long-lasting maladaptation occurring in the cocaine addiction.
Keywords: Mitochondria, Cocaine, Transcriptome, Epigenetics
Disclosure: Nothing to Disclose.

T231. NR4A2 is a Molecular Regulator of Medial Habenula-Mediated Cocaine Reinstatement Behaviors

Alberto Lopez*, Philip Hwang, Om Chitniss, Rianne Campbell, Janine Kwapis, Dina Matheos, Marcelo Wood

University of California, Irvine, Irvine, California, United States

Background: The habenula complex is an epithalamic region composed of lateral and medial (MHb) substructures. Mounting evidence has demonstrated that the MHb is able to code for the aversive properties of nicotine, serving as a substrate for nicotine withdrawal and aversion through the dense expression of nicotinic acetylcholine receptors along the MHb-interpeduncular nucleus pathway. Although typically characterized as an "anti-reward" pathway, the endogenous function of the MHb has yet to be fully characterized. The role of this pathway with regard to other drugs of abuse, however, remains particularly unclear. Work from our lab and others has demonstrated that the MHb is engaged by reinstatement of cocaine-associated behaviors, but is insensitive to acute doses of cocaine. Moreover, our initial studies demonstrated that chemogenetic activation of the cholinergic MHb population induces reinstatement of cocaine-induced CPP. Yet, the molecular mechanisms within the MHb that are altered leading up, and in response, to reinstatement of cocaine behaviors are unknown. NR4A2, a transcription factor critical in cocaine-associated behaviors and known regulator of dopaminergic signaling, is enriched in the cholinergic cell-population of the MHb. Furthermore, NR4A2 has been shown to be necessary for MHb development, but its role within the MHb in the adult brain remains elusive, specifically, given the lack of dopaminergic neurons within the MHb. We hypothesize that NR4A2 may mediate changes to the epigenome to changes in circuit function within the MHb to drive relapse-like behavior.
Methods: Here, we seek to characterize the role of the NR4A2 within the MHb in driving reinstatement of cocaine-

induced CPP. Animals underwent a previously described cocaine-primed CPP reinstatement paradigm. Briefly, animals were alternately conditioned with either 10mg/kg cocaine-HCl or saline (I.P., counter-balanced, unbiased). Following 4 conditioning sessions, animals were reintroduced to CPP chamber in a drug-free state to extinguish conditioned preference. Once extinction criteria was met (two consecutive days of preference score not significantly different from 0), animals were primed with either saline or 5mg/kg of cocaine-HCl prior to a reinstatement test session. To determine if NR4A2 within the MHb is necessary and sufficient for cocaine-reinstatement behaviors, we developed Cre-dependent constructs that will overexpress wild-type NR4A2 (DIO-NR4A2) or NURR2C (DIO-NURR2C), an endogenously-occurring, dominant-negative variant of NR4A2. After injecting either DIO-NR4A2, DIO-NURR2C, or DIO-GFP bilaterally into the MHb (0.5uL, A/P -1.5, M/L +/- 0.35, D/V -3.0) of male and female ChAT-IRES-Cre mice, animals underwent a cocaine-primed CPP reinstatement protocol. Following reinstatement, animals were sacrificed and tissue harvested to confirm viral expression via immunohistochemistry and measure changes in histone acetylation.

Results: Wild-type animals that underwent cocaine-primed reinstatement had a significant increase in H4K8 acetylation (H4K8Ac) in the MHb compared to saline-primed controls. H4K8Ac has been correlated with increases in NR4A2 expression, suggesting NR4A2 is a critical molecular mechanism regulating the reinstatement response. Furthermore, DIO-NR4A2 expressing animals showed an enhanced reinstatement response compared to DIO-GFP controls. Conversely, DIO-NURR2C expressing animals showed a blunted reinstatement response compared to DIO-GFP controls.

Conclusions: With regard to substance use disorder, the MHb has been characterized as a key regulator of nicotine-associated behaviors. However, evidence from our group and others has demonstrated it plays a key role in regulating the response to other drugs of abuse, such as cocaine. We have previously demonstrated that cholinergic activity within the MHb is critical in regulating cocaine-primed reinstatement. Here, we provide evidence that NR4A2 is a molecular mechanism within the cholinergic population of the MHb that modulates MHb-mediated reinstatement. Moving forward, it will be critical to identify the gene targets of NR4A2 in the MHb, as the MHb is a non-dopaminergic brain region, and how changes in NR4A2 lead to changes in MHb circuit function.

Keywords: Habenula, Reinstatement, Cocaine, Epigenetics
Disclosure: Nothing to Disclose.

T232. Measuring Alcohol Responses in Risky Drinkers in Their Natural Environment: A Proof of Concept Study

Andrea King*, Patrick McNamara, James Faria, Daniel Fridberg

University of Chicago, Chicago, Illinois, United States

Background: Laboratory alcohol challenges are the gold standard measure to ascertain participants' responses to alcohol administered in a controlled setting. Using this methodology, our group has shown that young adult binge drinkers show greater sensitivity to alcohol's stimulant and rewarding effects and these responses are more predictive of future development of alcohol use disorder (AUD) symptoms than lower alcohol sedative and impairing effects. However, laboratory-derived alcohol response protocols are labor-intensive and it is unclear if they measure effects that are indicative of real-world alcohol responses. These methodological issues have hampered extensions of alcohol challenge studies to intervention and epidemiological research. Thus, we conducted a proof-of-concept study in 28 young adult binge drinkers to assess their subjective responses to alcohol administered in the laboratory compared to alcohol consumed in their natural environment. We developed an accessible, user-friendly mobile smartphone technology and calculated estimated breath alcohol concentrations (eBrAC) based on recordings of number of drinkers consumed. Our goals were to determine the reliability and safety of mobile alcohol response assessment and the association between natural environment alcohol subjective responses to those measured in the laboratory.

Methods: The study was a within-subjects design with a laboratory session to measure responses for three hours after receiving a standard 0.8 g/kg dose of alcohol and smartphone assessments during the first three hours of a real-world binge drinking episode(s), with order (smartphone, laboratory sessions) randomly assigned. The main dependent measures were the Brief Biphasic Alcohol Effects Scale (BBAES), the Drug Effects Questionnaire, eBrAC/BrAC, and acceptability and feasibility survey responses. Participants were 28 non-alcoholic young adult weekly binge drinkers (mean age = 24.6 years; 46% female; 46% White, 18% Black, and 36% Other; AUDIT score = 12.0; average 7.3 heavy drinking days/month, 15.7 drinks/week).

Results: Results showed that participants consumed an average of 7.0 (range 3-11) drinks during a natural drinking episode with eBrAC steadily increasing to 0.12 ± 0.02 SD mg % three hours after initiating drinking compared with the full rising and declining limbs of the BrAC in the laboratory session, peaking at 0.09 ± 0.02 mg% one hour after alcohol consumption. Smartphone assessment response rates in the natural environment were excellent (228/252 assessments completed; 90%). At comparable eBrAC/BrAC (0.09 mg%, 1½ and 1 hour after initiating drinking in the real-world and laboratory settings, respectively), subjective alcohol stimulation and motivational reward (wanting) were of a higher magnitude in the natural environment than in the laboratory (ps $<$ .05), with a trend towards more hedonic reward (liking) in the natural environment ($p = .08$). Stimulation was highly correlated between natural environment and laboratory settings ($r = +.71$, $p < .001$), but sedation was relatively low and did not differ between settings. The smartphone assessment was rated favorably, based on moderate-to-high agreement on Likert-rated items that measured: ease of use of the mobile app (95% of participants), recommending the study to others (95%), and non-intrusiveness and length of time to complete surveys (67% and 76%, respectively). Overall satisfaction with the study was also very high (95% rated as moderate-to-high). The most common consequences experienced the day after natural environment drinking were less energy after drinking (80%) and mild hangover (76%), with no severe adverse effects reported.

Conclusions: In sum, real-world assessments of alcohol responses in young at-risk binge drinkers were acceptable and feasible with no serious adverse events observed. Smartphone methodology offers a practical solution for real-time, natural environment ascertainment of alcohol's pleasurable effects during a heavy drinking episode and may help facilitate more research on the role of alcohol responses in vulnerability to AUD and hazardous drinking over time in at-risk samples.

Keywords: Alcohol Sensitivity, Alcohol Consumption, Smartphone-Based App

Disclosure: Nothing to Disclose.

T233. Methamphetamine Use Does Not Modulate Interoceptive Processing of Soft Touch in HIV Infection

Amanda Bischoff-Grethe*, Stephan Jordan, Ronald Ellis, Susan Tapert, Martin Paulus, Igor Grant

University of California, San Diego, La Jolla, California, United States

Background: Methamphetamine use (METH) has been associated with altered frontostriatal reward processing circuitry that suggests impaired prediction error learning that guides decision-making. However, METH has also been associated altered interoception, defined as the sense of the physiologic condition of the body. Prior work has demonstrated attenuated insula and striatum response to the anticipation and receipt of pleasant touch in METH; this suggests an impaired ability to sense and integrate body homeostasis, which also guides motivated behavior. Given that METH use is also associated with an increased rate of HIV-infection, and that both METH and HIV can alter brain function, their comorbidity is likely to lead to significant behavioral dysregulation. Moreover, as neuropathy has been associated with HIV infection, an important question is whether this is further exacerbated in HIV individuals with METH use histories. Therefore, the purpose of this study is to examine interoceptive processing in individuals with a history of METH use (H-/M+), seropositive HIV individuals (H+/M-), HIV individuals with a comorbid history of methamphetamine use (H+/M+), and community controls (H-/M-). We predicted that while both H-/M+ and H+/M- would attenuate interoceptive processing as assessed by functional MRI, this would be further reduced in H+/M+.

Methods: Eighty-three participants (28 H-/M-, 20 H+/M-, 19 H-/M+, 16 H+/M+) experienced a soft touch paradigm during blood-oxygen level-dependent functional MRI. Participants received slow brush strokes on the left forearm and palm. Visual cues indicated upcoming touch stimuli to permit examination of both anticipation and receipt. Participants completed visual analog scales to rate touch pleasantness and intensity. Participants also underwent a comprehensive neurological exam at a prior visit. Data were analyzed in R, using a Group (H-/M-, H+/M-, H-/M+, H+/M+) x Condition (anticipation, soft touch) + Location (palm, forearm) linear mixed effects approach. To assess differences in reward and interoception, voxelwise analyses were constrained to the bilateral insula and bilateral striatum. Intrinsic smoothness was estimated using the spatial autocorrelation function (acf) option in AFNI's 3dFWHMx. Minimum cluster sizes were calculated with AFNI's 3dClustSim to guard against false positives; a peak voxel of $p < 0.001$ with a cluster threshold of $p\alpha < 0.025$ was required for significance. Post-hoc pairwise comparisons were Bonferroni corrected for multiple comparisons. Exploratory Huber robust regression analyses were conducted within the insula and striatum to examine the relationship between neural activation and clinical variables related to METH use (age of first use, days since last use, METH use density), HIV infection (illness duration in months, current CD4, nadir CD4), and self-reported touch pleasantness and intensity (peak voxel threshold $p < 0.01$, cluster threshold $p < 0.025$).

Results: The H-/M+ and H+/M+ groups did not differ on age of first use, days since last use, or METH use density ($ps > 0.11$). Similarly, H+/M- and H+/M+ groups did not differ on illness duration or CD4 measures ($ps > 0.33$). Groups did not differ in self-report ratings of touch pleasantness or intensity ($ps > 0.11$). However, groups differed in terms of severity of neuropathic dysesthesias ($p = 0.03$, partial eta squared = 0.11), paresthesias ($p = 0.02$, partial eta squared = 0.12), and loss of sensation ($p = 0.01$, partial eta squared = 0.13), with H+/M+ reporting greater severity compared to CON ($ps < 0.05$) in all measures. A significant group x condition interaction was detected within the left posterior insula, bilateral anterior insula, and left caudate. Post hoc analyses suggested that only H-/M-, H+/M-, and H+/M+ differentiated anticipation and receipt of soft touch in the posterior insula. H-/M- also had greater BOLD response to pleasant touch relative to patient groups in the posterior insula. Exploratory regression analyses suggested that H+/M- with higher pleasantness ratings was associated with higher BOLD response in the bilateral anterior insula and putamen to soft touch forearm. In contrast, H+/M+ with higher intensity ratings were associated with lower left ventral caudate response to palm soft touch. No relationships with touch receipt were detected in H-/M- or H-/M+. Clinically, H+/M+ with higher current CD4 had lower BOLD responses in the left posterior insula to soft touch of the palm. In comparison, the H+/M- group had no significant relationship between BOLD responses and clinical measures. Finally, H-/M+ with longer abstinence had greater BOLD response to soft touch of the forearm and palm in both the insula and striatum.

Conclusions: In support of prior literature, H-/M+ showed an attenuated response to soft touch anticipation and receipt in the insula and caudate relative to the other groups. In contrast, both H+/M- and H+/M+ displayed an attenuated response to soft touch relative to H-/M- only in the posterior insula. However, H+/M+ also demonstrated higher posterior insula response to soft touch with higher current CD4, and lower striatal response to soft touch with greater self-reported intensity ratings. METH use in HIV did not appear to introduce additional interoceptive or reward processing impairment relative to HIV seropositive individuals with no METH use history, despite a higher prevalence of neuropathic symptoms. Taken together, these data suggest that an altered response to pleasant touch in HIV infection may be limited to interoceptive circuitry independent of METH use.

Keywords: Insula, Striatum, Interoception, HIV, Functional Neuroimaging

Disclosure: Nothing to Disclose.

T234. Repeated Delta-9-Tetrahydrocannabinol (THC) of Marijuana Upregulates DCC mRNA Expression in Prefrontal Cortex, but THC Combined With Cannabidiol (CBD) Does Not: Relevance to Psychiatric Symptoms Associated With Long Term Marijuana Use?

Bertha Madras*, Jack Bergman, Stephen Kohut, Susan George, Yasmin Hurd, Zhicheng Lin

Harvard Medical School, Belmont, Massachusetts, United States

Background: Regular heavy marijuana use in susceptible populations is associated with heightened risks for marijuana (cannabis) use disorder, memory impairment, and less frequently, psychiatric disorders, including schizophrenia, bipolar disorder, depression, social anxiety, and exacerbation of psychiatric symptoms. Of the 100 or more cannabinoids in the marijuana plant, THC is the most abundant cannabinoid followed by cannabidiol (CBD). Selective breeding has greatly increased the concentration of THC and reduced CBD levels in current marijuana products. THC is psychoactive, addictive, psychotomimetic and anxiogenic, whereas CBD does not engender these pharmacological effects and reportedly has anti-psychotic, anxiolytic, and anti-seizure therapeutic potential. With increasing evidence for a causal relationship between long-term potent marijuana use and psychiatric symptoms, we focused on an axonal guidance molecule, DCC, that conceivably mediates some of the adverse actions of marijuana based on research showing that: (a) heavy marijuana users exhibit reduced dopamine release, self-report blunted reward (less "high"), heightened negative responses (anxiety, restlessness) following a dopaminergic challenge, (b) aberrant prefrontal cortical dopamine signaling is implicated in psychosis; (c) the netrin-1 receptor DCC drives prefrontal cortex maturation by guiding dopamine circuit formation; (d) decreased DCC expression in mice increases dopamine innervation and improves specific behaviors, whereas exaggerated DCC expression is associated with major depression disorders in humans and behavioral anomalies in rodents; (f) GWAS association studies implicate the dcc gene in schizophrenia. We previously reported that dcc was upregulated in rats treated repeatedly with THC. Our present goals were to determine whether THC administered repeatedly to adult monkeys altered dcc and dopamine receptor mRNA expression in brain regions and whether CBD modulated the effects of THC.

Methods: Adult rhesus monkeys (Macaca mulatta) were treated for 24 days with escalating doses of THC, or THC combined with CBD, or vehicle control ($n = 3$/group). THC was administered during 24 total study days in a range of doses 0.1-3.2 mg/kg, i.m.) and brains were harvested on day 25. THC + CBD were administered for 24 total study days for THC (doses ranging from 0.1-3.2 mg/kg. i/m/) and CBD (days 6-24; doses ranging from 1-3 mg/kg i.m.), with brain harvest on day 25. Gene expression levels were measured following mRNA isolation from cerebellum, frontal cortex, hippocampus, and striatum, and cDNA synthesized. Expression of the selected genes were measured using real-time PCR using species-specific, intron-spanning when possible primers and probes. Data were analyzed and normalized to beta actin (ACTB) as a housekeeping gene. The average fold change values with standard error were analyzed for statistical significance, using a two-tailed, paired t-test in Excel.

Results: In nonhuman primates treated daily with THC for 24 days, dcc and the D1 dopamine receptor gene were upregulated in prefrontal cortex. CBD, if administered together with THC to nonhuman primates for 19 days, prevented THC up-regulation of dcc and D1 dopamine receptor genes in prefrontal cortex of nonhuman primates.

Conclusions: These exciting discoveries reveal that THC increased mRNA expression of the dcc gene, which encodes DCC, a receptor implicated in guiding formation of dopamine circuitry in prefrontal cortex. Conceivably THC modulates dopamine circuitry and function in the prefrontal cortex via DCC, with relevance to psychiatric and other adverse symptoms associated with chronic, frequent marijuana use. Cannabidiol effectively attenuated dcc up-regulation by THC, indicating that CBD can modulate THC-induced molecular adaptations. Although the mechanisms underlying THC and CBD effects on dcc remain unknown, we postulate that repeated exposure to marijuana containing a high THC:CBD ratio upregulates DCC in human prefrontal cortex, conceivably contributing to the emergence of adverse effects associated with heavy use marijuana. Others have reported overexpression of dcc in major depressive disorder in humans and behavioral anomalies in rodents. Current strains of marijuana have been bred to produce high THC:CBD ratios. Our results are relevant to candidate cannabinoid therapeutics and to regulatory oversight of the ratio of THC:CBD in strains of marketed marijuana.

Keywords: Marijuana, D1 Dopamine Receptors, Psychosis, Major Depression Disorder, THC

Disclosure: Part 1: RiverMend Health, Advisory Board, American Addiction Centers, Honoraria, Guidepoint, Consultant.

T235. Pilot Results in Recording Light and Dark Adapted Electroretinogram as a Measure of Dopamine Release

Sean Luo*, Diana Martinez, Stephen Tsang, Edward Nunes

Columbia University, New York, New York, United States

Background: Dopamine is a key neurotransmitter involved in multiple psychiatric disorders and mediating mechanisms of action for multiple psychotropic medications. However, methods for measuring dopamine release such as Positron Emission Tomography (PET) are cumbersome and expensive. A clinically accessible measure of dopamine release may allow for better treatment tailoring.

Methods: We designed a strategy to test whether electroretinogram signals (ERG) might be different before and after a dose of oral immediate release methylphenidate (60mg). We carried out two sets of ERGs using the standard International Society for Clinical Electrophysiology of Vision (ISCEV) protocol in two normal control subjects.

Results: The average light adapted ERG b-wave over 3 trials was increased from 102uV to 152uV in Subject One and from 150uV to 178uV in Subject Two. Dark adapted b-waves were robustly increased (>100uV) in both subjects (252uV to 372uV in Subject One and 415uV to 521uV in Subject Two). Other components of the ERG signal showed no change before and after the medication.
Conclusions: Our results suggest that ERG, especially dark adapted ERG, may be a useful peripheral marker of dopamine release. Current studies are ongoing in correlating the ERG signal to the PET signal of central release of dopamine.
Keywords: Dopamine, Neurotransmitters, Retina, Positron Emission Tomography
Disclosure: Nothing to Disclose.

T236. Using Functional Near Infrared Spectroscopy to Measure Effects of Delta 9-Tetrahydrocannabinol on Prefrontal Activity and Working Memory in Cannabis Users

Jodi Gilman*, H. Onur Keles, Milena Radoman, Nina Levar, Eve Manghis, Randi Schuster, Gladys Pachas, A. Eden Evins

Harvard Medical School, Boston, Massachusetts, United States

Background: Intoxication from cannabis impairs cognitive performance, in part due to the effects of Δ9-tetrahydrocannabinol (THC, the primary psychoactive compound in cannabis) on prefrontal cortex (PFC) function. Acute, detrimental effects of THC are well documented. In double-blind, placebo-controlled studies, oral administration of 40-300 μg/kg THC caused acute, dose-dependent impairment in performance on memory, divided and sustained attention, reaction time, visual tracking and motor function tasks. However, a relationship between impairment in cognitive functioning with THC administration and THC-induced change in hemodynamic response has not been demonstrated. We explored the feasibility of using functional near infrared spectroscopy (fNIRS) to examine the functional changes of the human PFC associated with cannabis intoxication and cognitive impairment.
Methods: Participants were given a single dose of up to 50mg of dronabinol, an FDA-approved synthetic THC ingredient in MARINOL® Capsules. Study physicians determined the dronabinol dose based on the degree of expected tolerance, given participant's average dose, frequency, and type of cannabis use, self-report of degree of intoxication (high) experienced with each use, history of any adverse effects experienced when using cannabis, and baseline characteristics such as participant's sex, height, weight, BMI and blood pressure. Participants completed the Drug Effects Questionnaire (DEQ), a 100mm visual analogue scale, pre-dose and every 20-25 minutes post-dose to assess the extent to which participants (1) felt any dronabinol effect(s), and (2) felt high. Heart rate and blood pressure measurements were collected at baseline and at 25-minute intervals after dronabinol administration. Participants underwent two fNIRS sessions; one before dronabinol administration ("pre-THC"), and the other at approximately two hours after dronabinol administration ("post-THC"), which is the median peak of pharmacokinetic effects of dronabinol. During each session, participants completed a letter N-back working memory (WM) task. For the 0-back condition (low WM load), participants were instructed to press a response button whenever a letter "X" appeared. For the 2-back condition (high WM load), they were instructed to press the button whenever the presented letter was identical to the letter presented two trials prior. Functional data were collected using a continuous-wave NIRS device, in which 8 Sources and 7 detectors were placed on the forehead, resulting in 20 channels covering PFC regions. Physiological changes and subjective intoxication measures were collected.
Results: Participant's DEQ intoxication ratings of (1) feeling a drug effect and (2) feeling high increased significantly from pre-THC to post-THC, with mean peak ratings of 66.23 ± 27.73mm and 63.31 ± 24.62mm, respectively. They also experienced an expected increase in heart rate; mean increase at peak "high" was 33.77 ± 6.75bpm. We found a significant increase in the oxygenated hemoglobin (HbO) concentration after THC administration in several channels on the PFC during both the high working memory load (2-back) and the low working memory load (0-back) condition. To assess for significant effect of THC on activation in regions related to working memory, we compared the mean changes in HbO concentration pre-and post THC drug treatment during the 2-back condition. The RM-ANOVA showed a trend-level effect of drug treatment ($F = 4.62$, $P = 0.052$), a significant effect of channel ($F = 4.56$, $P < 0.001$), and a significant interaction between drug treatment and channel ($F = 2.69$, $P < 0.001$), indicating that HbO concentration in some channels increased significantly after THC administration. After controlling for multiple comparisons, six channels showed significantly greater signal post-THC compared to pre-THC, three on the left and three on the right prefrontal cortex. No correlations were found between the mean HbO concentration changes and N-back behavioral performance during the 2-back condition. Though participants generally made more errors after THC (detected as a small to medium effect size), overall task performance was not significantly different between pre-and post-THC (accuracy: $F(1,22) = 0.98$, $P = 0.33$, Cohen's dz = .31; percent error: $F(1,22) = 1.06$, $p = 0.31$, Cohen's dz = .31).
Conclusions: The current study suggests that fNIRS may be used to investigate changes in hemodynamic blood flow to the PFC during cannabis intoxication. To our knowledge, this is the first report of fNIRS to examine the effect of THC administration on prefrontal hemodynamic changes during a WM task. In this preliminary study, we observed a significant increase in HbO concentration after THC administration in several channels on the left and the right PFC during both 0-back and 2-back WM conditions. Though behaviorally, we did not observe a performance decrement (likely due to the small sample size of 12 participants), increase in HbO may indicate that maintaining task performance was more difficult after THC administration.
Keywords: Cannabis, Working Memory, THC, Prefrontal Cortex
Disclosure: Nothing to Disclose.

T237. Metabotropic Glutamate Receptor Subtype 5 (mGluR5) Availability Associated With an Amphetamine Sensitization Regimen in Humans: A PET [11C]ABP688 Study

Kelly Smart*, Atsuko Nagano-Saito, Michele S. Milella, Gassan Massarweh, Pedro Rosa-Neto, Salah El Mestikawy, Marco Leyton, Chawki Benkelfat

McGill University, Montreal, Canada

Background: The metabotropic glutamate receptor subtype 5 (mGluR5) modulates neurotransmission and regulates some forms of synaptic plasticity. In animal models, reducing mGluR5 activity reduces cocaine self-administration, locomotor activation, and drug-related associative learning. In studies in humans, reduced availability of this receptor has been observed in people with cocaine use disorders, as measured by positron emission tomography (PET) and the selective mGluR5 ligand, [11C]ABP688. However, it is not yet known if this reflects a pre-existing vulnerability or an adaptation to repeated drug use. To address this question, the present study used PET with [11C]ABP688 to assess the relationship between striatal mGluR5 levels and psychomotor responses to a repeated d-amphetamine regimen that produces behavioral and neurochemical (dopaminergic) sensitization in healthy volunteers.
Methods: Eighteen healthy, stimulant-naïve volunteers (13 female, 5 male) were randomly assigned to receive 0.3mg/kg d-amphetamine (Dexedrine) or placebo. Treatment was administered in three consecutive doses given approximately 48 hours apart, followed by an identical challenge dose 16 days later. Behavioral responses to the drug were assessed by measuring speech rate in a standard talking task and subjective ratings of activation and alertness on visual analog scales (VAS, 0-10) and the Profile of Mood States (POMS). Behavioral sensitization was defined as a significantly larger drug-induced response to the fourth amphetamine dose as compared to the first.
All participants underwent two one-hour high-resolution HRRT PET scans with 370mBq [11C]ABP688, once prior to the first amphetamine dose and a second following the sensitization regimen and immediately prior to the final dose. Binding potential (BP(ND)) values were calculated relative to a cerebellar grey matter reference region and were extracted from three functionally defined divisions of the striatum: associative, sensorimotor, and ventral subregions.
Results: Behavioral sensitization was observed for speech rate and subjective ratings of racing thoughts (VAS - "Mind Racing") in the drug group ($n = 10$; mean change in number of words spoken per 5 minutes at challenge vs. dose 1: +49.1 SD 38.7, $p = 0.003$; mean change in VAS - Mind Racing score +2.7 SD 3.4, $p = 0.035$).
[11C]ABP688 BP(ND) values were significantly higher in men relative to women ($p = 0.037$). Controlling for this sex difference, BP(ND) did not differ between drug and placebo groups at baseline or follow-up, and no change in BP(ND) values was observed following treatment. In the drug group, BPND at follow-up was negatively correlated with subjective ratings of racing thoughts following the challenge dose in all three striatal subregions (associative $r = -0.84$, $p = 0.003$, sensorimotor $r = -0.81$ $p = 0.004$, ventral $r = -0.75$ $p = 0.013$). In female participants, a trend was observed towards a negative correlation between BP(ND) in the sensorimotor striatum and increase in speech rate at challenge dose ($r = -0.75$, $p = 0.054$).
Conclusions: This study assessed the relationship between mGluR5 binding and psychostimulant-induced psychomotor sensitization in humans. The results suggest that lower mGluR5 availability in the striatum following a repeated amphetamine administration regimen is associated with more pronounced psychomotor responses to a subsequent d-amphetamine challenge in healthy volunteers. Given that such responses are linked to increased drug-seeking behaviors in animal models, these results are consistent with the hypothesis that low striatal mGluR5 may be an early indicator of risk for developing stimulant dependence, as well as being a potential target for treatment. Sex differences in availability of this receptor may be important in understanding these relationships.
Keywords: Psychostimulants, Amphetamine, Metabotropic Glutamate Receptor, PET Imaging, Sensitization
Disclosure: Nothing to Disclose.

T238. Subcortical Functional Hyperconnectivity in Cannabis Dependence

Peter Manza*, Dardo Tomasi, Ehsan Shokri Kojori, Nora Volkow

National Institutes of Health, Bethesda, Maryland, United States

Background: Cannabis use has been associated with higher risk of psychosis (including schizophrenia), particularly for frequent use and early cannabis initiation. However, the mechanisms underlying this association are poorly understood. Because increased DA signaling in midbrain-striatal circuits is associated with psychoses, we hypothesized that regular cannabis abuse (CA) would be associated with increased resting functional connectivity in these circuits.
Methods: To test this hypothesis, we took advantage of the large dataset produced by the Human Connectome Project and examined resting brain activity of subcortical regions in 441 young adults, including 30 CA meeting DSM criteria for dependence, and 30 controls matched on age, sex, education, BMI, anxiety, depression, and alcohol/tobacco usage.
Results: Across all subjects, local functional connectivity density (lFCD) hubs were most prominent in ventral striatum, hippocampus, amygdala, dorsal midbrain, and the posterior-ventral brainstem. As hypothesized, CA showed markedly increased lFCD relative to controls in ventral striatum and substantia nigra/ventral tegmental area but also in brainstem and lateral thalamus. These effects were observed in the absence of significant differences in subcortical volume, and were most pronounced in the individuals who began cannabis use earliest in life. Behaviorally, the groups did not significantly differ on various metrics of cognitive performance. However, in line with previous research, CA reported higher levels of negative emotionality, which was associated with higher levels of subcortical lFCD in CA but not controls.
Conclusions: Together, these findings suggest that chronic cannabis abuse is associated with changes in resting brain function, particularly in dopaminergic nuclei implicated in

psychosis but that are also critical for habit formation and reward processing. These results shed light on neurobiological differences that may be relevant to the increased risk for psychoses associated with cannabis use.
Keywords: Resting State Functional Connectivity, Addiction, Basal Ganglia, Functional MRI (fMRI), Graph Theory
Disclosure: Nothing to Disclose.

T239. Gender Differences in Dopamine D3 Receptor Availability as Determined by [18F]Fluortriopride, a Dopamine D3 Receptor Selective Radiotracer

Jacob Dubroff*, Robert Doot, Chris Ward, Jenny Cai, Kyle Labban, Hsiaoju Lee, Chia-Ju Hsieh, Kuiying Xu, Erin Schubert, Shihong Li, Catherine Hou, Regan Sheffer, Alex Schmitz, Robert Mach

University of Pennsylvania, Philadelphia, Pennsylvania, United States

Background: The dopamine D3 receptor is thought to represent the principal neural substrate of the dopaminergic mesolimbic circuit, the primary reward pathway in the mammalian brain. There are recognized neurobiological differences in addiction between genders that remain incompletely explored (Becker, McClellan et al. 2017). The dopamine D2-like receptors (i.e., D2/3 receptors) have been the most extensively studied CNS receptor with PET. There are three different radiotracers which have been largely used in these studies: [11C]raclopride, [18F]fallypride, and[11C] PHNO. Pohjalainen used [11C]raclopride PET to examine gender difference and found a trend towards reduced D2 receptor availability in females(Pohjalainen, Rinne et al. 1998). More recently, Okita and colleagues used [11C] raclopride PET brain imaging and found elevated D2 receptor availability in the ventral tegmental area and substantia nigra in nicotine dependent females compared to males(Okita, Petersen et al. 2016). However, no gender differences were present between healthy control males and females. In response to cigarette smoking, Cosgrove et al. have also shown gender differences in the timing of endogenous dopamine release in different brain regions using [11C]raclopride PET(Cosgrove, Wang et al. 2014).
Raclopride and fallypride are nonselective dopamine D2/3 antagonists. Raclopride has an affinity for D2 and D3 receptors in the low nM range, whereas fallypride has a sub-nM affinity for both receptors. Consequently, PET studies using [11C]raclopride or [18F]fallypride provide a measurement of the D2/3 binding potential. The sub-nM affinity for D3 receptors indicates that imaging studies with [18F] fallypride result in a higher labeling of D3 receptors than what is observed with [11C]raclopride. Although [11C] PHNO is recognized to be preferential for D3 (vs D2) receptors, it is not selective for D3. [18F]Fluortriopride (FTP) is a novel PET radiotracer that demonstrates high specificity for D3 vs D2 receptors ($>$160-fold) (Mach, Tu et al. 2011; Tu, Li et al. 2011; Rangel-Barajas, Malik et al. 2014). In anesthetized non-human primates, [18F]FTP binding was sensitive to levels of endogenous dopamine but reflected the distribution of D3 receptors as determined by autoradiography (Mach, Tu et al. 2011; Sun, Xu et al. 2012). To further evaluate the applications of this radiotracer, we performed the described first-in-human studies and compared distribution between males and females.
Methods: 10 heathy adults, 6 males (31.8 years old+/-9.8) and 4 females (26.3 years old +/-6.2) underwent 120 minutes of dynamic PET brain imaging following the bolus intravenous administration of 6.87 mCi (+/-0.22) of [18F] FTP. Patients provided informed consent for the studies according to guidelines of University of Pennsylvania Institutional Review Board. Dynamic arterial sampling was performed with subsequent metabolite analysis. Each participant also underwent MRI T1 imaging with isotropic voxels for co-registration with and segmentation of the PET data. Kinetic image analyses (Pmod v3.7 software) of [18F]FTP uptake and metabolite-corrected, arterial blood input functions via a 2-compartment model yielded total distribution volumes (VT). Because one male participant experienced unanticipated stress during dynamic PET scanning (claustrophobia) but completed the scan, his data was excluded. The difference between VT values from males and females was examined using a 2-tailed, 2-sample Student t test with equal variances. P values less than 0.05 were considered significant.
Results: Whole Brain [18F]FTP regional brain distribution volumes of total ligand uptake in tissue relative to total concentration of parent ligand in plasma (VT), was significantly ($P = 0.048$) greater in males (3.60 +/- 0.96, $n=5$) than females (2.18+/-0.76, $n=4$). There was a trend toward this finding in individual brain ROIs including the putamen ($P=0.062$), a known dopamine D3 receptor rich region. There was no significant difference between males and females in free fraction, a measure of the non-metabolized parent compound.
Conclusions: Despite a limited number of studies, initial [18F]FTP PET brain imaging suggests greater dopamine D3 receptor availability in males than females. Although this supports a neuroreceptor basis for gender differences in reward and addiction, further studies are needed to understand this observation and verify this promising new radiotracer.
Keywords: Dopamine 3 Receptor Imaging, Gender Differences, PET, Radiotracer, Dopamine (D2, D3) Receptors
Disclosure: Nothing to Disclose.

T240. Brain Signatures of Diminished Proactive Behavior Control in Cocaine Use Disorder

James Bjork*, Liangsuo Ma, Kathryn Cunningham, F. Gerard Moeller

Virginia Commonwealth University, Richmond, Virginia, United States

Background: Functional MRI research has indicated that the same neural circuitry recruited by demands to stop behavior is also recruited merely by signals that a stop might be necessary. However, it is not known whether individuals with cocaine addiction show this internal signature of proactive control. Also unknown are the effects of potential incentives on signatures of proactive control.
Methods: We scanned cocaine-dependent individuals and controls as they performed an incentivized stop signal task.

The task presented targets that indicated whether or not a stop signal might occur. Potential-stop trials were presented in potential gain, non-incentivized, and potential loss conditions.
Results: Under non-incentive conditions, controls showed activation in right inferior frontal gyrus by potential demands to stop (proactive control) but cocaine-dependent subjects did not. Cocaine dependent subjects showed exaggerated limbic responses to reward deliveries compared to controls. Potential reward and loss conditions diminished frontal cortical signatures of proactive control generally.
Conclusions: These data indicate that cocaine dependent individuals show blunted recruitment of behavior control neurocircuitry by potential need to restrain behavior, but show exaggerated limbic responses to rewards. These traits in combination would partly explain the difficulty in maintaining recovery.
Keywords: Impulsivity, Reward, Addiction, fMRI
Disclosure: Part 1: Boehringer-Ingelheim, Consultant.

T241. Genetic Risk Factor for Smoking Alters Reward Processing in Non-Smokers

Michael Tennekoon*, Betty Jo Salmeron, Bailey Mallon, Thomas Ross, Elliot Stein

National Institute on Drug Abuse, Baltimore, Maryland, United States

Background: The non-synonymous single nucleotide polymorphism (SNP) rs16969968 alters the α5 subunit of nicotinic acetylcholine receptors (nAChRs) and explains about 1-3%of nicotine addiction variance. Neurobiologically, preclinical work suggests this SNP may alter reward processing, particularly in the habenular complex. This altered reward processing is associated with lower nicotine aversion and higher nicotine consumption. In humans, this SNP is associated with lower resting connectivity in a circuit between the ventral striatum and dorsal anterior cingulate, components of the salience network (SN) in both smokers and non-smokers. This lower connectivity was correlated with nicotine dependence in smokers. However, despite it's relatively high penetrance (about 42% of the European ancestry), the mechanisms of action of this SNP remains unknown, especially in non-smoker carriers. Given that dopamine is known to play a role in addiction, that there are nAChR on dopaminergic neurons, and dopaminergic terminals are present in the SN, we hypothesized that this α5 SNP may mediate its affects through nAChRs on dopaminergic neurons during reward processing.
Methods: Sixty participants took part in this study. Participants were stratified by genotype; thirty-one participants in the major genotype group (G/G) and 29 participants in the minor (risk allele) genotype group (A/A or A/G). Participants were given methylphenidate, (to increase dopaminergic transmission), haloperidol (a D2 receptor antagonist), nicotine or a placebo on different study days in a randomized, counterbalanced design. To examine reward processing, participants performed a motion prediction task while BOLD data were acquired in a 3T scanner. During the task, participants briefly observed the motion of two balls travelling at different speeds, from different starting positions, towards a finish line. After a short period, the balls disappeared and the participants were asked to pick which ball they thought would have reached the finish line first. Difficulty levels of the task were dynamically adjusted so that error rates were approximately 33%. This constant error rate ensured participants remained uncertain about the correctness of their prediction. Feedback informed participants if their response was correct or incorrect. Additionally, on approximately 23% of trials, non-informative feedback (i.e. feedback that did not convey whether the participants response was correct or incorrect) was presented.
Results: Participants in both genotype groups were slower for error responses than correct responses – indicating a higher level of uncertainty for error responses. Neurobiologically, when examining responses to informative correct vs. error trials and averaged across all drug conditions, the minor group showed greater activation for error trials in the bilateral insula ($p < 0.05$ corrected at the whole brain level). As participants had slower reaction times for error trials than correct trials (irrespective of whether feedback was informative or non-informative), we investigated the neural underpinnings of this increased uncertainty. Specifically, the contrast of informative vs. non-informative feedback on error trials demonstrated significant gene x drug interactions across regions of the executive control network (ECN, e.g. bilateral middle frontal gyrus) and default mode (DMN, e.g. posterior cingulate cortex). This effect was driven by responses in the non-informative condition for the ECN in the minor allele group showing reduced activation on non-informative trials when nicotine was administered. The minor allele group also showed stronger suppression (increased deactivation) of the DMN for these non-informative trials when nicotine was administered. Given this finding we further examined neural processing of uncertainty, by looking at correct vs. error non-informative trials. Once again, there was a gene x drug interaction in both the ECN (middle frontal gyrus) and DMN (ventro-medial prefrontal cortex). Consistent with the above informative vs. non-informative contrast, nicotine reduced activation in the ECN for non-informative error trials in the minor allele group. Finally, haloperidol increased activation (reduced deactivation) in the DMN in the minor group for non-informative error trials.
Conclusions: Overall, the minor (vs. major) allele seems to promote increased salience attribution to negative feedback as demonstrated by increased activation of the SN to informative errors. In addition, our results suggest the involvement of dopamine in processing reward related uncertainty. Specifically, lowering dopaminergic transmission (with haloperidol) in the minor allele group reduced suppression of the DMN. Conversely, nicotine (which increases dopaminergic transmission, among other processes), increases suppression of the DMN and lowers activation of the ECN during reward related uncertainty. This may be a neural mechanism that helps individuals with the minor allele process uncertain situations, and may play a role in the increased nicotine dependence seen in smokers possessing the minor allele.
Keywords: Genetic Risk Factor, Nicotine Addiction, Reward Processing, Dopaminergic Modulation
Disclosure: Nothing to Disclose.

T242. Resting State Connectivity and Compulsivity: Association in Alcohol Use Disorder

Erica Grodin, Reza Momenan*

National Institute on Alcohol Abuse and Alcoholism, NIH, Bethesda, Maryland, United States

Background: Chronic alcohol dependence alters brain structure and the connectivity of resting state functional networks. One of the hallmarks of alcohol addiction is continued alcohol seeking despite adverse consequences (American Psychiatric Association, 2000), which can be viewed as compulsive alcohol use. Important hubs of some networks have been linked to addiction compulsivity. Among these regions are the anterior insula (AI) and anterior cingulate (ACC) purportedly known as hubs in salience network. In this study, we investigated the association between resting state connectivity of the anterior insula and anterior cingulate with compulsivity level seen in heavy alcohol users.
Methods: In the current study, we analyzed the association between self-report compulsivity and functional connectivity in a sample of alcohol dependent patients and non-treatment seeking heavy drinkers (N = 62, 21 females; average: age = 42.34 ± 11.96).
Participants underwent a battery of neuroimaging assessments including resting state functional magnetic resonance imaging (rs-fMRI). They also completed the Obsessive Compulsive Drinking Scale (OCDS), a self-report assessment of obsessive thoughts about alcohol and compulsive drinking behaviors (Anton et al., 1995). We used the compulsive drinking subscale score to assess compulsivity, which reflected the participants' current drinking (NTS-HD) or drinking pre-treatment (ALC).
Resting state fMRI datasets were collected using a single-shot gradient echo planar imaging pulse sequence with thirty-six axial slices acquired parallel to the anterior/posterior commissural line (TR = 2000 ms, TE = 30 ms, flip angle = 90 degrees, 3.75 mm x 3.75 mm x 3.8 mm voxels). For each subject, we acquired a total of 300 resting state frames (volumes) with eyes open.
To investigate the association between drinking compulsivity and functional network connectivity we utilized seed base connectivity, using the 3dGroupInCorr command in AFNI (Cox, 1996). We selected the anterior insula (L: 34, -14, 2; R: -36, -16, 2) as described by (Tyler et al., 2009) as seed locations (seed radius = 5 mm) and computed the whole brain functional connectivity. We then computed the correlation between the seed regions' whole-brain connectivity and the compulsivity score. In this preliminary analysis, we conducted the statistical correlation analysis at p = 0.005 (uncorrected).
Results: Connectivity between the right AI seed and the left Fusiform Gyrus (27 voxels), right Middle Frontal Gyrus (26 voxels), left Precentral Gyrus (23 voxels), right Superior Temporal Gyrus (22 voxels), and left Posterior Insula (21 voxels) were positively correlated with the compulsivity score. Connectivity between the left AI seed and the left Anterior Cingulate/medial frontal gyrus (36 voxels) and left Precentral gyrus (29 voxels) was positively correlated with compulsivity scores. We did not find any association with the anterior cingulate seed based connectivity and compulsivity.
Conclusions: Our preliminary results indicate association of self-reported compulsivity with the functional connectivity of anterior insula and regions related to motor activation and visual processing. To our knowledge there is no similar study of connectivity and compulsivity in alcohol addiction. However, one study has shown that functional connectivity of some networks including the salience network is associated compulsive use of cocaine (Hu et al., 2015). Another study has shown unmedicated patients with OCD showed positive correlation between global OCD symptom severity and the connectivity of the orbitofrontal cortex and the putamen (Beucke et al., 2013). We have also previously found associations between self-report compulsivity and salience network morphometry in alcohol dependent individuals (Grodin et al., in press). Together these results indicate that the salience, particularly the anterior insula, is implicated in alcohol associated compulsivity.
Keywords: Resting State Functional Connectivity, Compulsivity, Alcohol and Substance Use Disorders
Disclosure: Nothing to Disclose.

T243. A Molecular Mechanism for Choosing Alcohol Over a Natural Reward

Eric Augier*, Estelle Barbier, Russell Dulman, Gaëlle Augier, Esi Domi, Markus Heilig

Center for Social and Affective Neuroscience, Linköping, Sweden

Background: Once established, alcohol addiction is a chronic relapsing disorder, in which alcohol use becomes compulsive, i.e. continues despite negative consequences. Understanding the transition from controlled to compulsive alcohol use is therefore a critical challenge for addiction research. In humans, only a subset of users transition to compulsive drug use. In contrast, in commonly used animal models, nearly all rats learn to self-administer addictive drugs, including alcohol. This observation points to the possibility that focusing on self-administration alone may be insufficient to identify key mechanisms of addiction. This realization has prompted some important conceptual advances. For instance, it has been found that most rats will seize to self-administer cocaine when a high-value alternative becomes available, but that a subset of animals will continue their self-administration despite the presence of an alternative.
Methods: The neurobiological underpinnings of choosing alcohol over a natural reward are presently largely unknown. Here, we set out to identify molecular mechanisms underlying this choice behavior. We first established an exclusive choice-based method to identify rats that continue to self-administer alcohol at the expense of a high-value natural reward, a sweet solution.
Results: We observed that these animals show other characteristics reminiscent of clinical alcoholism.
Conclusions: We then carried out a discovery effort using gene expression profiling, and identified a novel molecular mechanism that appears to mediate compulsive-alcohol drinking at the expense of other high-value options.

Keywords: Alcohol Dependence, Choice, Compulsivity, Individual Variability, GABA Transmission
Disclosure: Nothing to Disclose.

T244. Circadian Rhythms and Opiates: Role of the Circadian Transcription Factor NPAS2 to Regulate Morphine Conditioned Reward

Ryan Logan*, Kelly Barko, Micah Shelton, Darius Becker-Krail, Puja Parekh, Lauren DePoy, Colleen McClung

University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania, United States

Background: Evidence from human clinical studies strongly suggest alterations to sleep and circadian rhythms as putative contributors to the pathophysiology of mood and addiction disorders. There are bidirectional relationships between drugs of abuse and circadian rhythms, such that rhythm disruption poses particular vulnerability to substance use, while acute and chronic administration of drugs of abuse impact rhythms, likely contributing to craving and relapse. However, the mechanisms underlying these relationships remain poorly understood.

Almost every cell in the body expresses genes comprising the molecular clock, a series of auto-regulatory transcriptional—translational feedback loops. A primary component of the molecular clock is the gas and metabolic responsive transcription factor, NPAS2, a member of the basic helix-loop-helix (bHLH)-PAS family of transcription factors. The bHLH domain is necessary for NPAS2 to form a heterodimer with BMAL1, another transcription factor critical for the regulation of circadian gene transcription. NPAS2 is particularly enriched in the mammalian forebrain, including the striatum, and particularly within the nucleus accumbens (NAc), a major substrate associated with mood and reward. A majority of the NAc is composed of medium spiny neurons (MSNs) expressing either dopamine receptor subtypes 1 (D1+) or 2 (D2+). Activation of D1+ MSNs typically promotes drug-related behaviors, such as conditioned place preference (CPP) to psychostimulants or opiates, whereas activation of D2+ MSNs inhibit the expression of these behaviors.

Our previous findings demonstrate psychostimulants increases Npas2 expression in the NAc, with an overwhelming enrichment of Npas2 specifically within D1+ MSNs of the NAc. Given these findings, we investigated whether opiates, in particular morphine, impacts the expression and activity of NPAS2 in the NAc, and the potential role of NPAS2 to regulate the behavioral responses to morphine using CPP. Moreover, we manipulated Npas2 specifically in D1+ or D2+ MSNs of the NAc to determine whether this circadian transcription factor has a cell-type specific role in the behavioral response to morphine.

Methods: Wild-type and NPAS2-bHLH-deficient (Npas2-bHLH -/-) male and female mice ($n = 8$-15 per group and condition) underwent an unbiased morphine CPP paradigm (i.p., morphine dose, 5 or 10mg/kg), whereby the amount of time spent on the conditioned vs. the unconditioned side was compared (CPP score). We also designed a Cre-inducible shRNA virus (AAV2) to knockdown Npas2 (or Scramble control) specifically in D1+ or D2+ MSNs by stereotaxic injection into the NAc of D1-Cre or D2-Cre mice ($n = 10$-15 per group and condition). Brains were extracted and NAc was punched for downstream molecular assays, including qPCR, Western blots, and protein IP. The effects of morphine on circadian gene expression in D1+ or D2+ MSNs of the NAc was achieved using either FACS analysis or RiboTag epitope IP followed by RNA-seq (50-60million, single end 75bp reads).

Results: Acute and chronic morphine administration altered the expression of NPAS2 in the NAc. Wild-type male and female mice showed an expected preference for the morphine-paired side at both doses. Npas2-bHLH -/- male mice displayed a significantly attenuated development of morphine CPP at 10mg/kg. Cell-type specific knockdown of Npas2 in D1+ MSNs of the NAc also significantly attenuated morphine CPP in male mice, with moderate effects in D2+ MSNs. Preliminary data suggests NPAS2 may bind several receptors in the NAc of morphine action, including mu, delta, and kappa opioid receptors.

Conclusions: Although there is continually accumulating evidence from human studies suggesting circadian rhythms are associated with mood and addiction disorders, the precise molecular mechanisms are largely unknown. We found NPAS2 regulates morphine conditioned behavioral reward, either by expression and/or activity modulation of transcriptional regulation of relevant downstream targets, such as opioid receptors. Furthermore, these actions may be specific to D1+ MSNs of the NAc. Currently, we are investigating whether NPAS2 may directly regulate these receptors as transcriptional targets, or via other mechanisms, and whether NPAS2 may modulate the activity of NAc MSNs during morphine exposure. These findings further demonstrate a link between circadian pathways in the brain and the behavioral response to drugs of abuse, possibly relevant for addiction.

Keywords: Circadian Rhythms, Addiction, Ventral Striatum, Opiates, Morphine
Disclosure: Nothing to Disclose.

T245. Occupancy of Dopamine D2 and D3 Receptors by a Novel D3 Partial Agonist BP1.4979:A [11C]-(+)-PHNO PET Study in Humans

Bernard Le Foll*, Patricia Di Ciano, Esmaeil Mansouri, Alan A. Wilson, Sylvain Houle, Isabelle Boileau, A. Leveille, M Capet, Thierry Duvauchelle, Jean-Charles Schwartz

Centre for Addiction and Mental Health, Toronto, Canada

Background: There has been a great deal of interest in the development of ligands selective for the dopamine D3 receptor (DRD3) over the DRD2 for the treatment of Psychiatric illnesses. Positron emission tomography (PET) is a non-invasive in vivo means to measure the binding of novel agents to receptors in the brain. [11C]-(+)-PHNO is an agonist radiotracer that can differentiate between binding of ligands to the DRD3 or DRD2. The purpose of the present study was therefore to investigate, in vivo in humans, for the first time, the occupancy of the DRD3 or DRD2 by a DRD3 partial agonist using PET imaging with [11C]-(+)-PHNO.

Methods: Participants were given BP1.4979 acutely at various time points prior to scanning with [11C]-(+)-PHNO. Based on these results, doses for sub-chronic once daily and B.I.D. administration were determined. This study was divided into 4 parts: 1) an acute dose-response study; 2) a time course study; 3) sub-chronic administration of BP1.4979 once a day; and 4) sub-chronic administration of BP1.4979 twice a day (B.I.D.). Blood samples were also drawn for determination of plasma levels of prolactin, BP1.4979 and two metabolites of BP1.4979. Participants were also given a visual analog scale to assess subjective aspects of BP1.4979. Adverse events were recorded throughout the study.
Results: It was found that BP1.4979 dose-dependently occupied the DRD3 and DRD2, and this occupancy was greater for the DRD3 at all doses. Occupancy was more long-lasting at the DRD3 than the DRD2. B.I.D. administration of BP1.4979 resulted in dose-dependent increases in occupancy of the DRD3. BP1.4979 was well-tolerated and there were no changes in subjective ratings after administration of BP1.4979. Prolactin was not appreciably increased at any dose or pre-treatment time, with the exception of the acute dose (30 mg) that also greatly occupied the DRD2. Plasma levels of BP1.4979 were dose-dependently increased but occupancy of the DRD3 was more long-lasting than elevations of plasma levels of BP1.4979, suggesting that the pharmacokinetics in the brain are different from those observed systemically.
Conclusions: This is the first study of the occupancy of the DRD3 in humans by a DRD3 partial agonist. These findings indicate the range of doses that can be used to occupy selectively the DRD3 over the DRD2 with BP1.4979 and speak to the use of in vivo imaging approaches in dose finding studies.
Keywords: Dopamine (D2, D3) receptors, PET Imaging, Partial Agonist
Disclosure: Part 1: Allergan, Consultant, Bioprojet, Grant, Pfizer, Grant, Mettrum, Honoraria, **Part 2:** Bioprojet, Grant, **Part 3:** Bioprojet, Grant, **Part 4:** Bioprojet, Grant.

T246. Effects of Smoked, Vaporized, and Oral Cannabis Administration on Appetitive Hormones and Relationships With Subjective Effects: A Clinical Study

Sofia Bouhlal*, Matthew Mewmeyer, Gray R. McDiarmid, Osama A. Abulseoud, Marilyn A. Huestis, Lorenzo Leggio

National Institute on Alcohol Abuse and Alcoholism, Bethesda, Maryland, United States

Background: Appetitive hormones may modulate reward and motivation not only for food but also for drugs of abuse. Therefore, appetitive neuroendocrine signaling may represent novel targets for addiction pharmacotherapy. Although cannabis impacts appetite, the acute changes in peripheral concentrations of appetitive hormones following Δ9-tetrahydrocannabinol (THC) administration, in otherwise healthy recreational cannabis users is less known.
Methods: Eligible participants took part in 4 randomized double-dummy sessions, during which equal amounts of placebo (0.001% THC), or active smoked, vaporized, or oral cannabis (6.9% THC) were administered (time 0 min). Circulating insulin, leptin, total and acyl-ghrelin, and glucagon-like peptide 1 (GLP-1) concentrations were assessed at times -25, +15, +30 and +90 min. Subjective responses were also measured at times -90, +15, +30 and +90 min.
Results: There was a main time ($p = .0005$) and time*route ($p = .001$) effect on insulin concentrations. For each route, Area Under the Curve (AUC) was assessed: circulating insulin significantly decreased under oral, smoked and vaporized cannabis administration compared to placebo ($p < .02$). No such effects were found for the other hormones. There was a main session effect ($p = .005$) on circulating leptin, as it increased from the first to the last session. No time ($p = .84$), route ($p = .43$) or time*route ($p = .05$) effects were observed for leptin. Similarly, the AUC model for leptin only showed a session ($p = .02$) but not route ($p = .16$) effects. No such effects were observed for the other hormones. There was a main time and time*route effect on positive subjective effects: "good drug effect" ($p < .0001$; $p < .0001$), "high" ($p = .0002$; $p < .0001$), "stoned" ($p = .009$; $p = .0002$), "stimulated" ($p = .002$; $p < .0001$), and "sedated" ($p = .01$; $p = .003$). In addition, a session effect was observed for negative effects such as "anxious" ($p = .02$), "irritable" ($p = .04$) and "restless" ($p = .004$).
Conclusions: These findings show the acute effect of THC in lowering insulin levels; these effects were more rapid after smoked or vaporized THC administration while delayed following oral administration. Through ongoing regression analyses, we are exploring the value of positive and negative subjective responses in prediction insulin and leptin levels respectively. This work could shed more light on the complex links between neuroendocrine regulation of appetite and the effects related to cannabis use.
Keywords: THC, Appetitive Hormones, Human Clinical Trial
Disclosure: Nothing to Disclose.

T247. Ketamine Prevents Reinstatement of Alcohol Seeking Induced by Cues+Yohimbine via an AMPA Receptor Independent Mechanism in Female, but not Male Rats

Megan Bertholomey, Bryan McElroy, Coreena Chan, Mary Torregrossa*

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: Women represent a vulnerable population in the development alcohol use disorders, as they transition to dependence faster, show greater craving in response to alcohol-associated cues and stress, and exhibit higher psychiatric comorbidity than men. While relapse rates are similar across gender, women are more likely to relapse when faced with stress or negative mood states during abstinence, suggesting that these factors are important when developing gender-based treatment strategies. In parallel with this hypothesis, we recently reported that female rats show increased alcohol craving-like behavior relative to males in response to cues, the pharmacological stressor yohimbine, and the combination of cues + yohimbine, indicating that females are more vulnerable and may benefit

from different treatment strategies than males. Thus, identifying sex-specific mechanisms mediating relapse-related behavior may improve treatment outcomes. We have begun to investigate these differences by determining sex differences in protein expression of glutamate receptor subunits in cortico-limbic circuits known to regulate reinstatement. In addition, we also tested the effect of the glutamatergic drug, ketamine, on reinstatement of alcohol seeking given that alcohol use disorders and depression are highly comorbid (especially in women), recent evidence that low-dose ketamine produces antidepressant responses in humans and rats, and that female rats are more sensitive to the antidepressant effects of ketamine.

Methods: Male and female rats underwent operant alcohol self-administration training wherein lever pressing resulted in access to a dipper containing 10% ethanol in a 0.1% saccharin solution. Control rats responded for saccharin. Each reinforcer delivery was accompanied by a 10s light +tone cue. After ~20 days of self-administration, rats underwent instrumental extinction training where lever presses produced no consequences. Rats were then tested for reinstatement of alcohol seeking induced by cues +yohimbine. Rats were given systemic injections of ketamine prior to self-administration (0, vs. 10 mg/kg) or reinstatement (0, 3, 10, 30 mg/kg) sessions. The role of AMPA receptors in mediating the anti-craving-like effects of ketamine was evaluated by injecting the AMPAR antagonist NBQX prior to ketamine, followed by the cue+yohimbine reinstatement test. A subset of female rats receiving 10 mg/kg ketamine or vehicle treatment during reinstatement testing were also tested for depression-like behavior in the forced swim test to determine both long-term prophylactic and acute antidepressant-like effects of ketamine. The amygdala and PFC were dissected from a separate group of drug-naive male and female rats for analysis of glutamate receptor subunit expression by Western blot. All animal procedures were approved by our Institutional Animal Care and Use Committee and were conducted in accordance with the NIH Guide for the Care and Use of Laboratory Animals.

Results: Analysis of sex differences in glutamatergic protein expression revealed that females had significantly greater expression of GluA1 and Glu2/3 in the amygdala relative to males, with no differences in the PFC. Ketamine pre-treatment significantly attenuated both alcohol and saccharin self-administration in females, while only affecting saccharin self-administration in males. Ketamine also completely blocked reinstatement of alcohol and saccharin seeking in females, while only reducing reinstatement to saccharin seeking in males. Alcohol seeking also tended to be blocked by a lower dose of ketamine (3 mg/kg) in females, that was not effective in males. Ketamine's anti-craving-like effects were not altered by prior NBQX treatment, and were transient in duration, lasting <48 hrs. In contrast, ketamine produced antidepressant-like responses in the forced swim test in females that lasted for at least 4 weeks.

Conclusions: Females are more sensitive to reinstate alcohol seeking in response to a combination of cues and a pharmacological stressor than males. The increased reinstatement may be related to enhanced AMPA-related signaling in the amygdala. Ketamine is extremely effective in reducing reinstatement of alcohol-seeking selectively in females. Unlike the antidepressant-like effects of ketamine, the anti-relapse-like effects have a much shorter duration of action and are not mediated through AMPAR activation, and the AMPAR antagonist disrupts reinstatement on its own, suggesting that ketamine's potential beneficial effects for the two disorders are mediated by independent mechanisms. Nonetheless, these data suggest that ketamine may be an effective treatment for women with comorbid alcohol use and mood disorders.

Keywords: Alcohol-Seeking Behavior, Ketamine, Sex Differences

Disclosure: Nothing to Disclose.

T248. Anterior Cingulate in Impulsivity and Cue Reactivity

F. Gerard Moeller*, Liangsuo Ma, James Bjork, Noelle Anastasio, Joel Steinberg, Andrew Snyder, Kathryn Cunningham

Virginia Commonwealth University, Richmond, Virginia, United States

Background: Previous clinical and preclinical research has shown a relationship between cocaine cue reactivity and impulsivity. The neurobiological basis for this relationship is not completely understood. This study integrates effective connectivity research in cocaine use disorder participants to examine what brain regions overlap between impulsivity and drug cue reactivity.

Methods: Fifteen cocaine dependent participants and ten controls underwent fMRI while performing a Go/NoGo task. Fifteen cocaine dependent participants underwent fMRI while performing a cocaine Stroop task. SPM Dynamic causal modeling was used to examine effective (directional) brain connectivity.

Results: Cocaine dependent participants showed a different pattern of brain connectivity compared to controls during successful inhibition on the Go/NoGo task. During successful inhibition on the Go/NoGo task, CocUD participants showed a significant reduction in left anterior cingulate cortex (ACC) to left caudate effective connectivity, while controls showed no change on this connectivity but an increase in the right dorsolateral prefrontal cortex to left caudate effective connectivity. During the cocaine Stroop task, the CocUD participants showed a significant increase in connectivity between the right ACC and right hippocampus, which was significantly correlated with greater attentional bias on the task.

Conclusions: The brain connectivity analysis found that both impulsivity as measured by the Go/NoGo task and cocaine cue reactivity as measured by the cocaine Stroop task were related to a network that included the ACC, whose controlling effects could reflect the pathology of cocaine use disorder. These findings, which will be discussed in light of preclinical research related to neuropharmacology of the ACC in these behaviors, suggest a key role for the ACC in both impulsivity and cocaine cue reactivity.

Keywords: Cocaine Addiction, Brain Connectivity, Anterior Cingulate Cortex

Disclosure: Part 1: Boehringer Ingelheim, Grant.

T249. A Human Laboratory Study of the mGluR5 Modulator Get 73 on Alcohol Craving, Pharmacokinetics and Pharmacodynamics in Individuals With Alcohol Use Disorder

Robert Swift*, Carolina Haass-Koffler, Lorenzo Leggio, Roberto Cacciaglia

VA / Brown University, Providence, Rhode Island, United States

Background: GET 73 is a novel, small molecule compound that reduces alcohol consumption and has anxiolytic and anti-stress activity in animal models. GET 73 appears to act as a negative allosteric modulator (NAM) at the mGluR5 receptor. mGluR5 NAMs could reduce the hyperglutamatergic / high stress state associated with alcohol use disorders.
Methods: We conducted a placebo-controlled, within-subjects crossover study with GET 73 in fifteen non-treatment seeking alcohol dependent participants. After screening for medical and psychiatric eligibility, participants were randomized to the 14-day study, receiving 3 days of medication treatment with GET 73 or placebo as inpatients, followed by a 7-day outpatient washout, followed by 3 days of the alternate medication as inpatients. Under each drug (GET 73 or placebo) condition, on the second treatment day (Day 2 and Day 12), participants received an oral dose of alcohol calculated to raise blood alcohol levels to 0.12 g/L. Alcohol pharmacokinetics and pharmacodynamics (stimulation, sedation, cognitive and motor impairment, mood, etc) and safety were monitored and compared between drug and placebo conditions. On Day 3 and Day 13, participants received a laboratory alcohol cue-reactivity (craving) session, followed by a laboratory alcohol self-administration session. Alcohol craving and laboratory alcohol consumption were compared between drug and placebo. Salivary cortisol levels were determined throughout the study.
Results: GET 73 enhanced the sedative effects of alcohol on the biphasic alcohol effects scale ($p = 0.044$), but had no effect on alcohol stimulation. There were no differences in alcohol-induced performance deficits between the GET 73 and placebo medication conditions. During the cue-reactivity session, GET 73, compared to placebo, reduced mean blood pressure ($p = 0.018$) and there was a trend to reduce craving, although this was not statistically significant. There were no differences in alcohol self-administration between the groups. There were no differences in number or pattern of adverse events between the drug conditions. There was a trend for GET 73 to reduce cortisol. GET 73 did not affect alcohol pharmacokinetics in these alcohol-dependent subjects (no difference in AUC), although there was a trend for a reduction in Cmax.
Conclusions: The results of this interim analysis demonstrate that the glutamate modulator GET 73 may reduce alcohol craving and enhance alcohol sedation in the human laboratory. The safety profile of GET 73 was excellent, as no differences in alcohol-induced performance deficits or in adverse events were observed between the groups. Enhanced alcohol sedation and reduced craving are seen in other medications used for the treatment of alcohol use disorders, including naltrexone and baclofen. The findings suggest that GET 73 may have promise as a treatment for alcohol use disorder.

Keywords: Alcohol Use Disorder, Metabotropic Glutamate Receptor, Alcohol Self-Administration, Pharmacotherapy
Disclosure: Part 1: Lundbeck, Honoraria, **Part 4:** Laboratorio Farmaceutico CT, Grant.

T250. Transient Innervation of the Dorsolateral Striatum by the Prelimbic Cortex and its Relation to Novelty Preferences During Development in Rats

Chloe Jordan, Susan Andersen*

McLean Hospital, Belmont, Massachusetts, United States

Background: Transient increases in dopamine activity during adolescence may lead to a perfect storm for vulnerability to drug use. Previous research has established the role of increased D1 on prelimbic cortical (PL) projections to the accumbens as important for increased sensitivity to cocaine, cocaine-associated cues, the motivation to take cocaine, and relapse with a reinstatement paradigm. However, none of these data explain how elevated novelty preferences predict heightened vulnerability to compulsive drug use. The process of addiction involves the progression between increased motivation to use drugs (mediated by the PL) that leads to increased action-oriented goal behavior. With repeated drug use, addiction becomes more compulsive and habitual in nature. The underlying neural process for this latter stage is associated with activity in the sensorimotor cortex (SMC) and its influence on the dorsolateral striatum (DLS). Here, we show that the PL transiently innervates the DLS during development, but is absent in adult rats. Activity within this pathway is associated with elevated novelty preferences.
Methods: Three different cohorts of female Sprague-Dawley rats were used to: 1) characterize the anatomical pathways between the PL and the SMC and the DLS with retrograde tract tracing (from the DLS) and analyzed with stereology at P25 and adulthood; $n = 4$-5/age; 2) identify relevant signaling molecules on the PL $\Rightarrow$ DLS pathway with combined retrograde tract tracing in the DLS, with labeled cells laser capture microdissected in the PL and analyzed with qPCR for dopamine D1-D5 receptor mRNA levels in animals characterized for novelty preferences at P25; $n = 10$/group; and 3) determine the behavioral relationship between the DLS and novelty preferences using Designer Receptor Exclusively Activated by Designer Drugs (DREADDs; $n = 6$-9/group). Briefly, subjects were habituated to the testing chamber for two days before receiving either a stereotaxic injection of the deactivating Gi/o DREADD (HSV-hM4Di-mCherry) or the control virus (m-Cherry) in the DLS or PL/DLS on P22. On P23, when the virus was fully effective, subjects received the inert drug clozapine-N-oxide (CNO; 1 mg/kg, i.p.) 30 min prior to novelty testing to assess any possible locomotor effects. They were then tested for novelty preferences at P25.
Results: In both identified neuronal pathways, cell counts were significantly higher at P25 when compared with the adults. Traced cells in the SMC $\Rightarrow$ DLS are abundant in both ages, but significantly elevated at P25 compared with the adults ($F1,9 = 7.95$, $P < .05$). Similarly, significant differences exist between P25 rats and adults ($F1,9 = 41.4$, $P < .001$) in PL $\Rightarrow$ DLS pathway, where the number of identified cells was very low (< 1 cell/mm2) in adulthood. Once the

PL ⇒ DLS pathway was identified with tracer, cells captured in the PL revealed that D1 and D4 mRNA significantly correlated with novelty preference scores (Pearson's product moment correlation r = 0.72 and r = 0.71, respectively; * P <0.05). Finally, the role of the DLS in novelty preferences was supported by the use of DREADDs. Here, CNO had no effect on activity on day 3 when comparing animals that did not receive CNO to those that did ($F1,53 = .04$, $P > 0.8$). Inactivation of the DLS or the DLS/PL regions with the DREADD Gi/o-hM4Di decreased novelty preferences significantly relative to the control mCherry virus ($F1,23 = 8.3$, $P < 0.01$), without a difference between regions.

Conclusions: These data demonstrate a transient, functional pathway between the PL and the DLS, thereby connecting brain regions that are associated with motivation and habit formation, respectively. Novelty preferences are mediated by DLS activity, and modulated by elevated D1 and D4 dopamine receptors in the PL. Together, these data may explain why a subset of animals show increased vulnerability to drug use at an early age and why novelty preferences are strongly associated with habitual drug use.

Keywords: Cocaine Addiction, Developmental Psychopathology, Habit

Disclosure: Nothing to Disclose.

T251. Desire to Drink Mediates the Effects of a GABRA2 Variant and Naltrexone on Drinking Level in Heavy Drinkers

Albert Arias*, Richard Feinn, Jonathan Covault, Howard Tennen, Henry Kranzler

Yale University School of Medicine, West Haven, Connecticut, United States

Background: A single nucleotide polymorphism (SNP) (rs279858, T/C) in GABRA2, which encodes the GABA α2 receptor subunit, has been associated with the risk of developing an alcohol use disorder (AUD), the subjective response to ethanol, and psychosocial treatment outcomes. We analyzed an existing sample of heavy drinkers to determine whether the SNP moderated the response to naltrexone treatment and whether the desire to drink mediated the effect.

Methods: The sample consisted of 153 European American heavy drinkers from a 12-week randomized controlled trial of daily or targeted naltrexone or matching placebo. We used daily interactive voice response technology to measure desire to drink and drinking. Using a series of multilevel models, we examined the main and interactive effects of rs279858 genotype and naltrexone treatment on daily desire to drink, and on lagged nighttime drinking, controlling for daytime drinking. To evaluate mediation, we used multilevel modeling in Mplus to test the effect of desire to drink on nighttime drinking and as a covariate in the nighttime drinking model.

Results: There was a significant interaction of genotype with medication on both desire to drink ($b = -0.60$, $p = 0.022$) and nighttime drinking ($b = -0.44$, $p = 0.045$). Subjects with the TT (low AUD risk) genotype showed less desire to drink and less nighttime drinking with naltrexone, while those with the C allele, previously associated with AUD, did not. Desire to drink significantly predicted nighttime drinking levels ($b = 0.31$, $p < 0.001$) and reduced the significance of the effect of the genotype by naltrexone interaction on nighttime drinking when included as a covariate, indicating that it partially mediated the interaction effect (indirect effect: $b = -0.24$, $p = 0.046$).

Conclusions: We found evidence for mediated moderation of the effects of naltrexone on nighttime drinking level in a sample of heavy drinkers. Naltrexone reduced nighttime drinking among rs279858*TT subjects but not C-allele carriers, an effect that was partially mediated by a reduction in the desire to drink. These findings add to the literature on the use of a "micro-longitudinal" approach to test the pharmacogenetics of AUD treatments and the clinical mechanisms by which they influence drinking. The findings may thus be useful for developing personalized AUD treatments.

Keywords: Alcohol Use Disorder, Naltrexone, Pharmacogenetics, GABRA2, Craving

Disclosure: Nothing to Disclose.

T252. A Long-Range GABA Projection From Ventral Tegmental Area to Ventral Accumbens Shell Promotes Positive Reinforcement

Ream Al-Hasani*, Gavin Schmitz, Julia Lemos, Veronica Alvarez, Michael Bruchas

St. Louis College of Pharmacy/ Washington University in St. Louis, St. Louis, Missouri, United States

Background: Projections from the ventral tegmental area (VTA) to the nucleus accumbens (NAc) have been implicated in a multitude of motivated behaviors. Interestingly, when investigating more discrete projections to the dorsal and ventral NAc shell, we identified a GABAergic projection from the VTA to the ventral NAc shell that modulates reward behavior. This long-range VTA GABA projection is has not been fully characterized. These long-range projections have been shown to selectively synapse onto cholinergic interneurons in the NAc and enhance associative learning, but the mechanism through which this same projection modulates positive valence and reward is unclear. Here we further characterize this projection in the context of motivated behavior and interrogate the cholinergic microcircuitry in the NAc to understand the mechanism.

Methods: We virally targeted AAV5-DIO-ChR2-eYFP to GABAergic cells in the VTA and implanted a fiber optic into the ventral and dorsal Nac shell and the NAc core. We then photostimulated the GABAergic terminals in the NAc from the VTA at multiple frequencies in a real-time place testing paradigm to determine the valence of this projection. This was followed by an operant self-simulation paradigm using fixed ratio 1, 5 and a progressive ratio to assess motivational value of the photostimulation. NorBNI (kappa opioid antagonist), Naloxone (mixed opioid receptor antagonist), SCH 23390 (Dopamine D1 receptor antagonist) and Sulpiride (Dopamine D2 receptor antagonist). To further investigate the valence of this projection we used in vivo fiber photometry combined with operant sucrose consumption in mice injected with GCaMP6s in the VTA with the photometry fiber in the ventral NAc shell in VGAT-Cre mice.

Results: We found that photostimulation of GABAergic terminals in the ventral NAc shell and not the dorsal NAc shell or the NAc core elicits a robust real-time place preference. Photostimulation of these GABAergic terminals was reinforcing enough to drive an operant self-stimulation under fixed ratio 1 and without prior training or food deprivation. Interestingly, when assessing the motivational value of these GABA cells in the ventral NAc shell the animals did not respond to either a fixed ratio 5 or a progressive ratio paradigm. We found that this robust reward behavior was not dependent on the opioid system or the dopaminergic system. Furthermore, using in vivo fiber photometry we show that this long-range GABA projection is involved in positive reinforcement following operant sucrose consumption.
Conclusions: Here we show that photostimulation of long range GABAergic projection specifically in the ventral NAc shell has a robust positive valence in both a real-time place testing paradigm and operant self-stimulation paradigm. Furthermore, using in vivo fiber photometry we were able to identify that these GABA cells are active during rewarding behavior. This behavior is not opioid or dopamine system dependent, but is likely mediated through cholinergic interneurons. Importantly, this projection does not drive motivation as shown by the lack of any response to FR5 or PR. The discrete mechanism through which the cholinergic system in the NAc may be modulating this reward behavior is currently under investigation.
Keywords: GABA Neuron, Cholinergic System, Nucleus Accumbens Shell, Reinforcement, Ventral Tegmental Area (VTA)
Disclosure: Nothing to Disclose.

T253. MRI Assessment of Brain Iron Content in Methamphetamine Users

Yosef Berlow*, William Hoffman, William Rooney

Alpert Medical School, Brown University, Providence, Rhode Island, United States

Background: Methamphetamine (MA) abuse produces long term changes to the dopamine system that likely contribute to the psychiatric and cognitive symptoms that are seen in MA users. Emerging evidence from preclinical studies suggests that brain iron accumulation plays a role in MA toxicity, both as a biomarker of damage and a potential source of oxidative stress; however, this relationship has not yet been characterized in human MA users. The goal of this study was to utilize in vivo magnetic resonance imaging (MRI) techniques to measure brain iron levels in human subjects with a history of MA dependence and determine if these measurements represent functionally significant biomarkers of MA toxicity. Iron is paramagnetic, and its presence increases the transverse relaxation rate constant (R2) of nearby water protons causing a loss of signal intensity on T2-weighted images. Measuring this effect using quantitative MRI techniques allows for in vivo investigation of the effects of MA on the distribution of brain iron.
Methods: MRI datasets were acquired from 27 currently abstinent MA users and 27 aged matched healthy control subjects. Series used in this study included a high-resolution, T1-weighted MPRAGE, T2-weighted Turbo Spin Echo sequences acquired with three different echo times and a 3D proton density (PD) sequence. R2 ($\equiv$1/T2) maps were calculated at each voxel using a monoexponential decay function. Maps of the fractional macromolecular (fM) content were created by normalizing PD images intensities to the peak intensity values of voxels within the cerebrospinal fluid. Parametric maps of the distribution of iron content were then calculated using a relaxometry model incorporating the combined effects of fM and iron content. These parametric iron maps were registered to a common brain space. The effect of MA on regional iron distribution was assessed using both region of interest analysis and voxelwise linear models. Additional analyses were also conducted using the R2 and fM maps, as well as T2-weighted signal intensity measurements.
Results: Using quantitative relaxometry measures that are specific for iron, this study was unable to detect any differences in regional iron content in former MA users when compared to aged-matched healthy control subjects. These measures yielded values for iron and fM that were in substantial agreement with literature values. These measures also detected strong age associated increases in iron content within basal ganglia regions consistent with previous reports providing a positive control for these methods. While no group differences in iron accumulation were found, the MA group had significantly reduced fM values in the thalamus, suggesting increased tissue water content in this region.
Conclusions: Contrary to the proposed hypothesis, this study found no evidence of altered iron accumulation in abstinent MA users, suggesting that iron accumulation is not a robust biomarker of MA toxicity in human users. This finding is in marked contrast to a study in nonhuman primates which demonstrated an MA induced increase in iron accumulation that was similar to the effects of advanced aging. The discrepancy between these findings is likely due to interspecies differences in brain iron accumulation.
Keywords: MRI, Methamphetamine, Biomarker, Iron
Disclosure: Nothing to Disclose.

T254. Neural Correlates of Alcohol Image Anticipation in Veterans With Concurrent PTSD and AUD: A Novel Cue Reactivity Paradigm

Andrea Spadoni*, Alan Simmons, Sonya Norman

VA San Diego Healthcare System, San Diego, California, United States

Background: Alcoholism in the context of posttraumatic stress disorder (PTSD) is common, especially among Veterans. Despite our best interventions, many continue to suffer following frontline treatments. Designing targeted and efficacious treatments would improve outcomes, but first, we must have objective markers of disease mechanism. Functional neuroimaging in individuals with PTSD and alcohol use disorders (AUD) may provide clinically relevant neuromarkers for the identification of treatment targets.
Methods: We used functional magnetic resonance imaging to assess brain activation during a novel visual alcohol cue reactivity paradigm in treatment seeking Veterans with a diagnosis of PTSD and AUD ($n = 32$). In addition to direct

cue exposures, alcohol and neutral associated stimuli were preceded by an anticipation period, during which time subjects either expected to see an alcohol beverage stimulus, a neutral beverage stimulus, or non-specific perceptual control stimulus.
Results: We found that Veterans with both PTSD and AUD demonstrated significant activation of medial prefrontal regions during anticipation of the alcohol cue relative to the non-specific perceptual control stimuli. We also found that during exposure to the alcohol associated stimuli, there was a relative increase in brain activation in the visual cortex relative to neutral beverage stimuli.
Conclusions: Treatment seeking Veterans with concurrent PTSD and AUD demonstrated not only enhanced brain activation during exposure to alcohol relative to neutral beverage cues, but also during anticipation of alcohol related visual stimuli. These data may indicate that anticipation of an alcohol cue, in addition to direct exposure, may be a sensitive probe for the motivational response that contributes to alcohol misuse. These results replicate those of previous cue reactivity studies in individuals with AUD alone, and extend them to demonstrate preferential activation for alcohol related stimuli in those with concurrent PTSD. Our findings represent the first steps in characterizing the motivational responses that may contribute to relapse in Veterans with PTSD who are trying to quit drinking.
Keywords: Posttraumatic Stress Disorder, Alcoholism, Functional Neuroimaging
Disclosure: Nothing to Disclose.

T255. Perineuronal Nets in the Insula Regulate Aversion-Resistant Ethanol Consumption

Hu Chen, Donghong He, Amy Lasek*

University of Illinois at Chicago, Chicago, Illinois, United States

Background: Perineuronal nets are specialized extracellular matrix structures that primarily surround parvalbumin-positive GABA neurons in the cortex and regulate synaptic plasticity and addiction-related behaviors in response to many drugs of abuse. Here, we examined the effect of repeated, intermittent binge-like drinking on perineuronal nets in the insular cortex, a brain region that is important for compulsive ethanol consumption, and the functional consequences of disrupting perineuronal nets on alcohol consumption.
Methods: To measure the effect of binge-like drinking on perineuronal nets, male C57BL/6J mice were subjected to the 4-day drinking in the dark (DID) procedure with 20% ethanol for 1 or 6 cycles and were sacrificed 1 day after the final drinking session. Fluorescence staining of Wisteria floribunda lectin (WFA) was performed to visualize perineuronal net intensity. In another set of experiments, the insular cortex was dissected to evaluate changes in gene and protein expression of the perineuronal net proteins aggrecan, brevican and phosphacan by quantitative real-time PCR and western blots. To determine the consequences of disrupting perineuronal nets in the insula on alcohol consumption, chondroitinase ABC (ChABC), an enzyme that degrades perineuronal nets, was infused directly into the insula. Three days after infusion, mice were tested in the two-bottle choice test for consumption of 15% ethanol vs. water or 15% ethanol and 100 μM quinine vs. water.
Results: DID for 6 cycles, but not 1 cycle, significantly increased perineuronal net intensity in the insular cortex. Furthermore, aggrecan, brevican and phosphacan protein expression, as well as aggrecan mRNA expression, were elevated in the insula after 6 cycles of DID. ChABC infusion into the insula resulted in decreased consumption and preference of the ethanol/quinine solution without affecting consumption of a quinine solution without ethanol.
Conclusions: We have demonstrated that repeated binge-like ethanol consumption using the drinking in the dark (DID) procedure increases the intensity of perineuronal nets in the insular cortex. In addition, digestion of perineuronal nets renders mice more sensitive to aversive ethanol solutions. Our results provide evidence that targeting insular perineuronal nets may be a mechanism to reduce aversion-resistant or compulsive alcohol drinking.
Funded by NIAAA (INIA-Neuroimmune Consortium and the Center for Alcohol Research in Epigenetics at UIC).
Keywords: Alcohol Drinking, Perineuronal Nets, Insula, Compulsive Models of Drug Use
Disclosure: Nothing to Disclose.

T256. Scavenging Endogenous BDNF Activity in the Dorsal Striatum Prevents Nicotine Withdrawal-Related Cognitive Flexibility Deficits in Mice

Vinay Parikh*, Robert Cole, Cassandra Wolsh, Evelynn Harrington

Temple University, Philadelphia, Pennsylvania, United States

Background: Persistent smoking of tobacco primarily occurs due to the highly addictive nature of its psychoactive ingredient nicotine. However, the behavioral and cellular mechanisms underlying nicotine addiction are not fully understood. Cognitive changes occurring during abstinence in tobacco smokers is hypothesized to predict relapse. We previously found disruptions in strategy set-shifting, a higher order cognitive flexibility process, in mice withdrawing from chronic nicotine. Moreover, these deficits were associated with an imbalance in the brain-derived neurotrophic factor (BDNF), a modulator of synaptic plasticity, in the frontostriatal networks. Here, we investigated whether nicotine withdrawal-related cognitive deficits are causally linked to overactive striatal BDNF signaling mediated by high-affinity tropomyosin-related kinase B (trkB) receptors.
Methods: Adult male C57BL/6J mice were trained in an operant task that required the animals to switch from using a spatial response-driven strategy to a visual cue-based strategy to achieve rewards. Mice were exposed to chronic nicotine (18mg/kg/d) or saline using subcutaneous mini-osmotic pumps for 14 days. Spontaneous nicotine withdrawal was induced by removing the pumps and the animals were tested on the strategy set-shifting phase. Animals received intracranial infusions of either recombinant trkB-fc fusion protein (a BDNF scavenging peptide that blocks trkB signaling) or IgG (control peptide) into the dorsomedial striatum (DS) or nucleus accumbens (NAc) on days 1, 4 and 7 of the testing phase.

Results: In general, mice undergoing nicotine withdrawal took substantially longer to acquire strategy set-shifting as compared to the saline-withdrawal mice ($F(1,63) = 11.49$, $p = 0.001$). Infusions of the trkB-fc peptide in the DS reduced the number of trials required to attain strategy-set shifting criterion in nicotine withdrawal mice ($p = .003$ vs. control peptide). Moreover, improved performance was attributed to reductions in strategy maintenance errors (control peptide: 46.13 ± 3.2 vs. trkB-fc: 27.9 ± 3.1; $p = .002$) in these animals indicating that restricting BDNF signaling in the DS improves the ability to execute a new learning strategy. Interestingly, NAc infusions of trkB-Fc were not able to reduce trials to criterion during the strategy-shifting phase in nicotine withdrawal mice ($p = 0.21$ vs control peptide). Moreover, trkB-fc infusions in the NAc neither reduced the total errors committed ($p = 0.19$) nor the number omissions during nicotine withdrawal (control peptide: 12.7 ± 6.4 vs. trkB-fc: 9.9 ± 6.1).
Conclusions: Here we show that deficits in cognitive flexibility that arise during nicotine withdrawal can be rescued by reducing BDNF activity in the DS. This brain region is crucial for the selection and maintenance of cognitive strategies to direct goal specific behavior. Thus, our findings indicate that cognitive rigidity observed during nicotine withdrawal is primarily mediated by augmented BDNF signaling and consequent plasticity changes in the DS. Although BDNF within the NAc appeared not to be critical for the manifestation of withdrawal-related cognitive deficits in our study, the involvement of BDNF signaling in this brain region in other aspects of withdrawal, such as affective symptoms and reward dysfunction, could not be ruled out. Taken together, our findings indicate that therapeutic strategies aimed at normalizing aberrant BDNF signaling in nicotine-abstinent smokers may help minimizing the cognitive deficits and eventually lower the instances of nicotine relapse.
Keywords: Cognition, Nicotine Addiction, Striatum, Mice
Disclosure: Nothing to Disclose.

T257. Development of Corticotropin Releasing Factor Binding Protein Allosteric Modulators

Carolina Haass-Koffler*, Chase Francis, Douglas Sheffler, Mohammad Naemmuddin, Carsen Nilsen, Selena Bartlett, Robert Swift, Lorenzo Leggio, Nicholas Cosford, Antonello Bonci

Brown University, Providence, Rhode Island, United States

Background: Stress response is believed to involve the corticotropin releasing factor (CRF) which interacts through its receptor and the CRF-binding protein (CRFBP). Recently, by utilizing novel a cell-based assay, we showed that the C-terminus of CRFBP(10 kD) fragment is able to potentiate CRF-intracellular Ca2+ release, demonstrating that CRFBP may possess excitatory roles in addition to the inhibitory role established by the N-terminus of CRFBP(27kD). This interaction is specific for the CRF receptor 2 (CRFR2). There are currently no small molecule ligands available that selectively interact with either CRFBP or CRFR2.
Methods: We miniaturized the developed cell-based assay, where we have expressed CRF-BP(10kD) on the plasma membrane fused as a chimera with CRF-R2 in order to develop a high throughput screening (HTS) assay.
Results: The identified two negative allosteric modulators (NAM) are able to blunt CRF-induced potentiation of NMDAR-mediated synaptic transmission on dopamine neurons in the VTA.
Conclusions: These results provide evidence of the first specific roles for CRFR2 and CRFBP in the modulation of neuronal activity and may be a target for both drugs of abuse and stress.
Keywords: Allosteric Modulator, Ventral Tegmental Area (VTA), Corticotropin-Releasing Factor (CRF), Brain Stress
Disclosure: Nothing to Disclose.

T258. Chronic Ethanol Exposure Alters GABAergic Transmission and GABA(A)-R Expression in Prelimbic MPFC via Epigenetic Mechanisms

A. Leslie Morrow*, Benjamin Hughes, J Peyton Bohnsack, Todd K O'Buckley

University of North Carolina School of Medicine, Chapel Hill, North Carolina, United States

Background: Alcohol Use Disorders are a leading cause of preventable death and account for $234 billion dollars in lost economic costs in the United States and effective therapeutic interventions for the treatment of alcohol use disorders are needed. Our laboratory and others have demonstrated that cortical GABAergic neurotransmission appears especially sensitive to alcohol exposure, with cortical neurons exhibiting substantial alterations in GABAA receptor (GABA(A)-R) function and subunit expression following chronic exposure. Further, chronic ethanol exposure alters GABA(A)-R α1 subunit expression in many brain regions. Previous work by our group demonstrated that histone deacetylase (HDAC) inhibitors can prevent changes in GABA(A)-R expression in cerebral cortex via elevations in HDAC2 and HDAC3 interactions with the Gabra1 promotor to reduce histone acetylation at the promoter. Furthermore, the HDAC inhibitor, Trichostatin A prevents ethanol-induced changes in zolpidem hypnosis and anxiety-like behavior in the open field. However, the electrophysiological significance of this change has not been studied to date in cerebral cortical slices. Therefore, we examined the role of histone deacetylation in ethanol effects on GABA(A)-R function and expression in prelimbic medial PFC, where ethanol is known to interact to influence executive function and ethanol dependence phenotypes.
Methods: We evaluated the effects of chronic ethanol exposure (5g/kg, i.g.,14 days) followed by 24h withdrawal on GABAergic transmission in prelimbic mPFC deep layer pyramidal cells by interrogation of spontaneous inhibitory postsynaptic currents (sIPSCs) in male and female rats. The HDAC inhibitor, Trichostatin A (TSA), was administered on the three days prior to sacrifice (2mg/kg, i.p.). We then performed electrophysiology as well as qPCR to determine if TSA prevents changes induced by chronic ethanol exposure. Whole-cell recordings of Layer V/VI pyramidal cells were performed using an Axon Instruments Multiclamp 700A amplifier sampling at 10 kHz and filtered at 2 kHz. GABAA receptor-mediated currents were isolated by supplementing

standard aCSF solution (in mM: 125 NaCl, 2.5 KCl, 1.5 NaH2PO4, 1.3 MgCl2, 2.6 CaCl2, 10 Glucose, 25 NaHCO3, 0.4 Ascorbate, osmolarity adjusted to ~310 mOsm with sucrose) with 10 μM CNQX, 50 μM dl-AP5, and 10 μM CGP-52432, and recordings obtained using 2-4 mOhm borosilicate glass filled with a cesium chloride-based internal solution (in mM: 140 CsCl, 2 MgCl2, 1 EGTA, 10 HEPES, 2 Na-ATP, 0.3 Na-GTP, 5 Phosphocreatine, 3 QX-314, pH 7.4, osmolarity adjusted to ~290 mOsm with sucrose). mPFC was microdissected from brain in ice-cold PBS then homogenized in Trizol (Ambion) and RNA was extracted and purified. RNA was quantified and quality controlled using a Nanodrop (all 260/280 and 230/260 values ≥ 1.8, Fisher Scientific). Purified RNA (2μg) was reverse transcribed to a cDNA library using High Capacity RNA to DNA kit (Applied Biosystems) following manufacturer's instructions. DNA (10 ng per reaction) was then subjected to qPCR analysis using TaqMan gene expression probes and Taq Gene Expression MasterMix (Applied Biosystems). Reactions were run in duplicate on a StepOnePlus RT-PCR system (Applied Biosystems) using glyceraldehdyde-3-phosphate dehydrogenase (Gapdh) as a loading control. Data was analyzed using the ΔΔCT method and expressed as fold control.

Results: Analysis of baseline sIPSCs revealed effects of chronic ethanol gavage associated with changes in post-synaptic function in prelimbic mPFC. We observed a 27.9 ± 3.7% decrease in 1 decay time ($p < 0.05$) that was prevented by the administration of TSA, but no change in baseline amplitude, rise time or 2 decay time. Furthermore, TSA had no direct effect on any sIPSC parameter in the absence of ethanol exposure. Chronic ethanol exposure further produced a 21.6 ± 4.6% decrease in baseline sIPSC frequency ($p<0.05$) that is likely indicative of presynaptic adaptations onto Layer V/VI pyramidal cells. Next, we conducted qPCR to examine changes in gene expression in PFC microdissections. Gabra1 gene expression was decreased (0.47 ± 0.05 fold control, $p < 0.001$) and this was prevented by the administration of TSA (0.87 ± 0.06 fold control, $p < 0.01$). We next evaluated if Hdac2 and Hdac3 gene expression were upregulated in mPFC. Hdac2 was increased after chronic ethanol gavage (1.8 ± 0.09 fold control, $p < 0.01$), but TSA treatment did not prevent this upregulation. Hdac3 was also increased after chronic ethanol gavage (1.84 ± 0.10 fold control, $p < 0.0001$) and this change was prevented by the administration of TSA (1.10 ± 0.05 fold control, $p < 0.0001$).

Conclusions: These studies demonstrate a loss of GABAergic inhibition in Layer V/VI prelimbic mPFC neurons following chronic ethanol exposure and withdrawal that can be prevented by HDAC inhibition using TSA. The changes in sIPSCs were accompanied by an increase in Hdac2 and Hdac3 gene expression along with a decrease in Gabra1 expression that may contribute to reduced inhibition resulting from the increase in tau1 decay time. Further, these studies demonstrate a novel mechanism for regulation of GABAergic sIPSCs, providing further evidence that HDAC inhibitors may regulate numerous brain circuits dependent upon GABA inhibition for proper function.

Keywords: Alcohol and Substance Use Disorders, GABA-A Receptors, Medial Prefrontal Cortex

Disclosure: Nothing to Disclose.

T259. Distinct Brain Activity in Response to Alcohol and Food Reward Anticipation During a Pharmacological Challenge With Intravenous Ghrelin in Alcohol-Dependent Individuals

Mehdi Farokhnia*, Erica Grodin, Mary Lee, Emily Oot, Ashley Blackburn, Bethany Stangl, Melanie Schwandt, Lisa Farinelli, Reza Momenan, Vijay Ramchandani, Lorenzo Leggio

National Institutes of Health, Bethesda, Maryland, United States

Background: There is a considerable overlap between the neurobiological circuitries that regulate alcohol and food seeking behaviors. Ghrelin, a peptide primarily synthesized by endocrine cells in the stomach, has been shown to modulate both central reward and stress pathways, in addition to its known role in increasing appetite and food intake. In rodent experiments, ghrelin administration increases accumbal dopamine release and enhances motivation for alcohol. Human studies also suggest a positive relationship between endogenous ghrelin concentrations and alcohol craving. However, whether and how ghrelin signaling may affect alcohol seeking behavior in humans remains unknown. For the first time, we investigated the effects of a pharmacological challenge with exogenous ghrelin on alcohol self-administration and brain function in regions associated with alcohol-related behaviors.

Methods: This was a proof-of-concept human laboratory study consisting of two experiments: intravenous alcohol self-administration (IV-ASA) and brain functional magnetic resonance imaging (fMRI). Each experiment included two visits with a crossover, randomized, double-blind, placebo-controlled design. During each visit, a 10-minute loading dose of IV ghrelin (3 mcg/kg) or placebo, followed by a continuous ghrelin (16.9 ng/kg/min) or placebo infusion was administered. Participants were heavy alcohol drinkers not seeking treatment for alcohol problems. During the IV-ASA experiment, participants had the opportunity to press a button to receive alcohol infusions using the Computerized Alcohol Infusion System with a progressive ratio schedule. Each infusion raised the breath alcohol concentration (BrAC) by 7.5 mg% over 2.5 min followed by a decrease of 0.5 mg% per min until the next infusion. During the fMRI experiment, task-based and resting-state scans were acquired under three pharmacological conditions: no drug, ghrelin/placebo, and ghrelin/placebo + alcohol. For the task-based scans, alcohol-food incentive delay (AFID) task was used. During this task, participants are repeatedly presented with neutral, food, and alcohol symbols; 2-6 seconds after seeing the cue symbol ("anticipation phase"), a target appears and participants can earn a point if they press the button while the target is on the screen. Each resting-state run included 5 minutes of scanning, during which participants were instructed to stay awake and not to move or close their eyes. Participants received IV alcohol during the last part of the fMRI experiment. This infusion was designed to raise the BrAC linearly to 80 mg% within 20 minutes and clamp the BrAC at this target value until the end of experiment. The blood oxygen level dependent (BOLD) signal change was assessed in the following brain regions of interest: amygdala,

anterior insula, medial orbitofrontal cortex (mOFC), and nucleus accumbens (NAc).

Results: A total number of eighteen individuals signed the informed consent; the final sample analyzed for the IV-ASA and fMRI experiments included eleven and eight participants, respectively. Participants self-administered higher number of alcohol infusions during the ghrelin than placebo session (percent change: 24.97 ± 10.65, $t = 2.34$, $p = 0.04$, Cohen's $d = 0.74$). Under ghrelin compared to placebo, participants started pressing the button sooner ($p = 0.01$) and received their first infusion earlier ($p = 0.03$). Analysis of the BOLD signal change during anticipation phase of the AFID task showed significant drug × AFID run × cue type interactions in left amygdala [$F (4, 26.8) = 3.54$, $p = 0.01$] and right mOFC [$F (4, 27.0) = 3.66$, $p = 0.01$] and a trend-level interaction in Left NAc [$F (4, 26.6) = 2.35$, $p = 0.08$]. Pairwise comparisons within these interactions indicated that IV ghrelin increased the alcohol-related signal in left amygdala ($p = 0.03$), decreased the food-related signal in right mOFC ($p = 0.04$), and increased the food-related signal in left NAc ($p = 0.03$). A trend-level drug × resting-state run interaction was also shown for correlation between right amygdala and right mOFC [$F (2, 28.4) = 3.00$, $p = 0.06$]. Analysis of the individual resting-state runs showed that, compared to placebo, the functional connectivity between these two regions was increased under ghrelin ($p = 0.02$), but decreased under ghrelin + alcohol ($p = 0.05$).

Conclusions: This study represents the first human evidence that exogenous ghrelin administration modulates both behavioral (self-administration) and neurobiological (brain function) elements of alcohol seeking behavior. Exogenous ghrelin administration increased alcohol intake in this study. The fMRI results showed significant effects of IV ghrelin on brain regions involved in alcohol and food seeking behaviors. These data suggest that specific and distinct brain regions are primarily engaged by IV ghrelin in anticipation for alcohol (amygdala) versus food (mOFC, NAc) reward. It can therefore be hypothesized that ghrelin's effects on motivation for alcohol and food are primarily mediated respectively through negative (stress) and positive (reward) reinforcement pathways. However, additional studies are required to obtain a definite conclusion in this regard. Together with previous human and rodent work, findings of the present study provide rationale for studying the ghrelin system as a potential pharmacological target for the treatment of alcohol use disorder.

Keywords: Ghrelin, Alcohol, Self-Administration, fMRI, Gut-Brain Axis

Disclosure: Nothing to Disclose.

T260. Use of Intracranial Self-Stimulation as a Screen for Novel Preclinical Therapeutics for Alcohol Use Disorders

Amanda Barkley-Levenson*, Andre Der-Avakian, Abraham Palmer

University of California, San Diego, La Jolla, California, United States

Background: Intracranial self-stimulation (ICSS) is a powerful tool for studying brain reward systems and provides insight into how drugs of abuse induce both hedonic and anhedonic states. ICSS has been extensively used to study rewarding effects and abuse liability of many illicit or therapeutic substances; however, ICSS has not been used as extensively to examine the motivational effects of alcohol. Alcohol is a compound stimulus that has both hedonic and anhedonic properties that depend on dose, time and individual differences in sensitivity. Pavlovian conditioning procedures are commonly used to assess sensitivity to alcohol reward and aversion, but these tests are limited because it is difficult to measure both reward and aversion in the same animal. Here, we show that ICSS can be reliably used to measure both reward enhancing and aversive effects of alcohol in the same mouse across a range of doses.

Methods: Adult C57BL/6J mice were implanted with electrodes targeting the medial forebrain bundle and were trained on a discrete-trial current-intensity ICSS procedure. After baseline reward thresholds were established, mice were tested to determine the effects of several doses of alcohol (0.5-1.75 g/kg, i.p.) on ICSS reward thresholds using a within-subjects Latin square design. In a separate experiment, we tested the effects of a potential anti-alcoholism compound (pBBG, 7.5 mg/kg, i.p.) that inhibits the enzyme glyoxalase 1 (GLO1). pBBG has been previously shown to reduce binge-like alcohol drinking in mice without altering the potency of other behavioral effects of alcohol.

Results: Reward thresholds were lowered by alcohol doses in the 0.5-1 g/kg range as compared to vehicle injection, indicating that these doses enhanced reward sensitivity. In contrast, the 1.75 g/kg dose of alcohol produced dramatic increases in reward thresholds, suggesting that it was aversive. pBBG alone had no effect on reward thresholds, indicating that this treatment produces neither undesirable anhedonic side-effects nor potential abuse liability due to hedonic effects. In ongoing experiments, we are testing whether pBBG can block the reward enhancing effects of alcohol and/or increase the aversive effects.

Conclusions: ICSS can be used in mice to measure both the reward enhancing and aversive effects of alcohol. Ultimately, we hope to use the ICSS procedure to screen potential AUD therapeutics for both their abuse liability and their effectiveness at modulating the rewarding and aversive effects of alcohol.

Keywords: Intracranial Self-Stimulation, Alcohol Use Disorders, Mouse Model, Novel Therapeutics, Reward and Aversion

Disclosure: Nothing to Disclose.

T261. Cell Type Specific Ventral Pallidum Regulation of Cocaine Seeking

Jasper Heinsbroek*, Ana-Clara Bobadilla, Daniela Neuhofer, Peter Kalivas

Medical University of South Carolina, Charleston, South Carolina, United States

Background: The ventral pallidum (VP) is a critical structure for regulating drug related motivation in addicts, yet precisely how distinct VP circuits mediate drug seeking and taking remains unknown. While the VP is thought to be an inhibitory relay and output for ventral basal ganglia circuits, recent data indicate complex information processing

in the structure. In addition to GABAergic neurons the VP contains a large number of glutamatergic projection neurons, and it receives inhibitory inputs from both nucleus accumbens dopamine D1- and D2- expressing medium spiny neurons as well as excitatory inputs from various limbic sources. We investigated how this information is integrated by VP GABAergic and glutamatergic projection neurons, and dissected the roles of these populations on the motivation to seek and take cocaine.
Methods: To investigate the connectivity and contributions of VP GABAergic and glutamatergic neurons to drug seeking and taking, we employed virus-based anterograde and retrograde tracing methods, as well as opto- and chemogenetics in Vglut2-IRES-Cre and Vgat-IRES-Cre mice, crossed with reporter lines. Mice received indwelling jugular vein catheters and were trained to self-administer cocaine, and after reaching a stable baseline of responding, contributions of VP GABAergic and glutamatergic neurons to drug taking were measured during progressive ratio tests. Afterwards, mice underwent extinction training and drug seeking was assessed during cue-induced reinstatement. Following extinction slice recordings were performed to assess the effects of cocaine self-administration on synaptic inputs and neuronal plasticity in glutamatergic and GABAergic neurons.
Results: Cell counts revealed that ~30% of VP neurons are glutamatergic, and that glutamatergic and GABAergic VP neurons receive differential but overlapping inputs from the nucleus accumbens and other limbic structures. After cocaine self-administration, chemogenetic stimulation of combined VP neurons greatly enhanced drug seeking during cue-induced reinstatement, while inhibiting the VP reduced reinstatement. Specific stimulation of VP glutamatergic neurons produced opposite effects by abolishing the motivation to take drugs during progressive ratio, and reducing cue-induced reinstatement of cocaine seeking. Furthermore, inhibiting glutamatergic VP neurons increased drug taking. By contrast, stimulation of VP GABAergic neurons produced mixed effects by increasing the motivation to take drugs, while decreasing the motivation to seek drugs during reinstatement.
Conclusions: These results reaffirm that the VP is a critical regulator of addictive behaviors, and expand our knowledge of the addiction circuitry by showing that motivated states in the VP are differentially regulated by GABAergic and glutamatergic neurons. In addition, these data reveal that GABAergic and glutamatergic VP neurons are integrated within overlapping but distinct basal ganglia circuits which differentially control the motivation to seek and take drugs.
Keywords: Cocaine, Drug Relapse, Ventral Pallidum
Disclosure: Nothing to Disclose.

T262. Pavlovian-To-Instrumental Transfer is Linked to Alcohol Consumption in Young Adults

Michael Smolka*, Stephan Nebe, Christian Sommer, Maria Garbusow, Daniel Schad, Quentin Huys, Florian Schlagenhauf, Andreas Heinz

Technische Universität Dresden, Dresden, Germany

Background: Pavlovian-to-instrumental transfer (PIT) tasks examine the influence of Pavlovian conditioned stimuli on instrumental behavior. The importance for (conditioned) context stimuli on (instrumental) alcohol consumption is widely acknowledged and rodent studies have shown that animals previously exposed to drugs show generally stronger PIT effects. However, only few studies have investigated PIT effects in human substance use disorders. So far, three studies investigated PIT effects in association to alcohol and reported increased PIT effects in alcohol-dependent patients compared to healthy control participants, but no association on a cross-sectional level in social drinkers. We now investigated whether PIT effects are linked to risky alcohol use in young adults.
Methods: We examined 191 18-year-old adults being neither abstinent from nor dependent on alcohol using a PIT paradigm and acquired information about their drinking behavior at the baseline of an ongoing longitudinal study.
Results: Baseline analyses revealed an association between stronger PIT effects and higher alcohol consumption. This was the case when examining the influence of Pavlovian stimuli directly on the instrumental behavior (i.e. number of button presses) as well as rather indirectly on the accuracy of these instrumental responses (i.e. percentage of correctly classifying stimuli and acting accordingly).
Conclusions: Our results indicate that individual strength of PIT effects is related to alcohol consumption even in a preclinical sample. Results of the longitudinal analyses will shed light on the role of these mechanisms for the individual drinking trajectories.
Keywords: Pavlovian, Instrumental Learning, Alcohol Consumption
Disclosure: Nothing to Disclose.

T263. RBP-6000 Buprenorphine Monthly Depot Demonstrates Sustained Clinical Efficacy and Safety in Phase III Opioid Use Disorder Trials

Barbara Haight*, Anne Andorn, Celine Laffont, Malcolm Young, Aksana Jones, Susan Learned, Christian Heidbreder

Indivior, Inc., Richmond, Virginia, United States

Background: RBP-6000 is a long-acting, sustained-release formulation of buprenorphine for monthly administration by subcutaneous injection. Two multicenter studies in the United States were conducted to assess the efficacy and safety of RBP-6000 in subjects seeking medication-assisted treatment (MAT) for opioid use disorder (OUD).
Methods: Both studies included MAT-seeking adults (not currently on medication) who met DSM-5 criteria for moderate or severe OUD for at least 3 months prior to enrollment. Study 1 (NCT02357901) included open-label (OL) induction with buprenorphine/naloxone (BUP/NLX) sublingual film (up to 2 weeks) followed by a double-blind treatment phase (24 weeks) in which patients were randomized to RBP-6000 300/300 mg (6 doses x 300 mg), RBP-6000 300/100 mg (2 doses x 300 mg, then 4 doses x 100 mg), or volume-matched placebo (6 doses x PBO). Study 2 (NCT02510014) included completers from Study 1 (roll-over subjects) and patients who did not participate in Study 1 (de novo subjects). All Study 2 subjects received up to 2 weeks of OL treatment with BUP/NLX sublingual film followed by an

initial 300-mg injection of RBP-6000 and subsequent 300-mg or 100-mg doses (based on tolerability). Roll-over subjects had 6 total injections in Study 2 (after 6 injections in Study 1), while de novo subjects had 12 total injections. All subjects in both studies received individual counseling in addition to RBP-6000 treatment. The primary efficacy endpoint in Study 1 was the cumulative distribution function (CDF) for percentage abstinence, defined as the percentage of urine samples negative for opioids combined with self-reports negative for illicit opioid use (collected from Week 5 through Week 24); missing urine samples and/or self-reports were imputed as non-negative. Percentage abstinence per week was also calculated without imputation. The main objective of Study 2 was to evaluate the long-term safety and tolerability of OL RBP-6000. Persistence of efficacy was also assessed in Study 2, based on percentage abstinence as defined in Study 1. For roll-over subjects (excluding placebo subjects), efficacy analyses were conducted using abstinence data from Study 1 linked to corresponding data from Study 2. A population pharmacokinetic (PK) model was used to characterize buprenorphine plasma concentration levels over 12 months of treatment using combined data from these 2 studies and data from a phase 2 study (NCT01738503). Using efficacy data from Study 1, exposure-response relationships were assessed for the following outcomes: percentage abstinence; Opioid Craving Visual Analog Scale (VAS) scores; Clinical Opiate Withdrawal Scale (COWS) scores; and Subjective Opiate Withdrawal Scale (SOWS) scores.

Results: Study 1 included 504 subjects in both the intent-to-treat (ITT) and safety populations (300/300 mg, $n = 201$; 300/100 mg, $n = 203$; placebo, $n = 100$). Study 2 included 669 subjects (roll-over, $n = 257$; de novo, $n = 412$). A majority of subjects completed Study 2 (roll-over, 78%; de novo, 50%). The most common reasons for discontinuation were loss to follow-up and withdrawal of consent by subject. Of the 201 subjects who had a dose reduction in Study 2, 25 subsequently had their dose increased to 300 mg. Forty-nine of the dose reductions were due to an adverse event (AE); of these, 39 were able to complete the trial and 8 subsequently had their dose increased to 300 mg. For the primary efficacy endpoint in Study 1, both RBP-6000 treatment groups were significantly superior to placebo ($P < 0.0001$), with mean percentage abstinence of 41% (300/300 mg), 43% (300/100 mg), and 5% (placebo). Among those on active treatment who completed Study 1 ($n = 245$), mean percentage abstinence at Week 24 was 69% (300/300 mg) and 60% (300/100 mg). Of those who completed another 6 months in Study 2 ($n = 174$), 61% were abstinent at Week 49. The population PK model (including $>19{,}000$ PK samples) indicated that target buprenorphine plasma concentrations of ≥ 2ng/mL were achieved from the first dose of RBP-6000, reached steady-state after 6 injections, and were further maintained at ≥ 2ng/ml up to 12 injections. Results of the exposure-response analyses at 6 months indicated that maximal responses for abstinence and Opioid Craving VAS were achieved at buprenorphine plasma concentrations of ~2 ng/mL and ~3 ng/mL, respectively, corresponding to ~70% and ~75% mu-opioid receptor occupancy. Maximal responses for both COWS and SOWS were achieved at a buprenorphine concentration of ~4 ng/mL, corresponding to ~78% mu-opioid receptor occupancy. Exposure-response relationships at 12 months would be expected to be similar given the consistency of buprenorphine plasma levels observed with long-term treatment. No unexpected safety signals were found in Study 2. The incidence of treatment-emergent AEs (TEAEs) were reported as follows (in roll-over and de novo subjects, respectively): any TEAE (56% and 73%), any serious TEAE (3% and 4%), any severe TEAE (3% and 9%), any TEAE leading to study discontinuation (2% and 3%).

Conclusions: There was no evidence of loss of efficacy with continued RBP-6000 dosing for up to 12 months of treatment, and no new safety signals were found. Therapeutic target plasma concentrations of buprenorphine were maintained throughout long-term treatment. This observed level of exposure was associated with abstinence and other measures of efficacy including control of craving and withdrawal symptoms.

Keywords: Opioid Dependence, Exposure-Response Model, Long-Term Trial

Disclosure: Part 2: Indivior, Inc., Employee, **Part 5:** Indivior, Inc., Employee.

T264. Uncertainty Breeds Want

Paola Mascia, Nichole Neugebauer, Jason Brown, Nancy Bubula, Qiang Wang, Kathryn Nesbitt, Robert Kennedy, Paul Vezina*

University of Chicago, Chicago, Illinois, United States

Background: As with drug abuse, pathological gambling (PG) is associated with symptoms of compulsion, loss of control, and continued pursuit despite negative consequences. Pathological gamblers are also at increased risk for developing substance use disorders suggesting that PG and drug abuse share common neuronal substrates. A defining feature of games of chance is uncertainty of outcome. Here we characterize the effects of prolonged exposure to conditions of uncertainty on a number of biochemical and behavioral effects normally observed in rats exposed to psychostimulant drugs. These rats show heightened levels of drug seeking consistent with excessive incentive wanting.

Methods: Rats in different groups were trained to nose poke for a saccharin solution under certain [fixed-ratio (FR)] or uncertain conditions (variable-ratio (VR)] for up to 55 1-hr sessions given twice/day 6-days/week. FR and VR ratios were escalated as follows following satisfaction of criteria: 1, 2, 3, 5, 7, 10, 13, 16, 19, 20. Rats were maintained on the last ratio (FR/VR 20) for 20-25 days. During the saccharin nose-poking sessions, number of nose-pokes, dipper entries, saccharin reinforcers, and sessions spent on each ratio were measured. Midbrain dopamine (DA) neuron reactivity was also assessed in some rats with microdialysis in the nucleus accumbens (NAcc) at FR/VR ratios 5, 13-16, and 19-20. Two weeks following the last saccharin nose-poking session, separate rats were tested for their locomotor or NAcc dopamine (DA) response to amphetamine or their self-administration of the drug using a lever press operant. Additional rats were also used to assess effects on NAcc protein levels of CaMKII, ΔFosB, and GLT1.

Results: Throughout the saccharin sessions, FR (certain) and VR (uncertain) rats did not differ in the number of nose-

pokes or dipper entries emitted, saccharin reinforcers obtained, or sessions spent on each ratio. On the other hand, the pattern of DA overflow observed in the NAcc during these sessions was remarkably similar to the flat certainty and exponentially increasing uncertainty variance curves associated with the escalating FR and VR ratios. When tested at ratios 5, 13-16, and 19-20, rats trained under the FR ratios (certain relationship between nose-pokes and reward; 0 variance) showed little to no increases in DA throughout the sessions. Conversely, rats tested under the VR ratios (uncertain relationship; increasing variance with increasing ratios) showed an exponential increase in DA overflow that was not associated with emission of the different behaviors but tracked the variance of the ratios (a measure of uncertainty). When tested two weeks following the last saccharin nose-poking session, VR rats exposed to conditions of uncertainty subsequently showed a higher locomotor and NAcc DA response to amphetamine (1.0 mg/kg, IP) relative to FR rats exposed to predictable reinforcement. In addition, when these rats were given the opportunity to self-administer amphetamine (100 μg/kg/infusion), they displayed enhanced work output and self-administered more drug infusions. Protein levels of CaMKII and ΔFosB were increased while those of GLT1 were decreased in these rats, effects paralleling those observed in psychostimulant exposed rats.

Conclusions: These results indicate that prolonged intermittent exposure to conditions of uncertainty can trigger neuroadaptations similar to those produced by repeated intermittent exposure to abused psychostimulant drugs. They also further support a relationship between uncertainty and DA activation. Because repeated intermittent drug-induced increases in DA have been implicated in the induction of sensitization, these findings provide a likely mechanism for repeated exposure to conditions of uncertainty to similarly promote its development. Together, these findings support a unified theory of addiction in which excessive incentive wanting drives the maintenance and progression of both drug and behavioral addictions such as PG.

Keywords: Addiction, Dopamine, Drug Abuse, Pathological Gambling, Sensitization

Disclosure: Nothing to Disclose.

T265. A Genome-Wide Association Study Reveals Association Between Alcohol Withdrawal Phenotypes and Sequence Variation in the GABRA2 and TDO2 Gene Regions

Victor Karpyak*, Duan Liu, Jennifer Geske, Antony Batzler, Mark Frye, Richard Weinshilboum, Doo-Sup Choi, Joanna Biernacka

Mayo Clinic, Rochester, Minnesota, United States

Background: Alcohol withdrawal syndrome (AWS) is a fundamental component of alcohol use disorder (AUD), reflecting a major step in its progression. Evidence supports a potential role for genetic variation in risk for AWS-related seizures and delirium tremens; however, single nucleotide polymorphisms (SNPs) associated with AWS have not been identified. Careful phenotype specification as well as proper selection of cases and controls are critical considerations in the design of genetic association studies and may improve the power to identify single nucleotide polymorphisms (SNPs) associated with AWS on a genome-wide scale. Here we present the results of a pilot project using genome-wide association data to compare which of the two commonly used definitions of the AWS phenotype is more suitable for the future large-scale studies.

Methods: We used the following definitions of the AWS phenotype: (a) two or more of the AWS symptoms, with one being hand tremor and (b) the total number of AWS symptoms in a single subject. An Illumina HumanCore genotyping array was used to genotype DNA samples collected from 400 European Americans who met DSM-IV criteria for alcohol dependence. Following quality control and imputation, over 7 million single nucleotide polymorphisms (SNPs) with a minor allele frequency (MAF) $\geq$ 0.01 were analyzed. SNP genotypes were tested for association with each of the two AWS phenotypes. Top association signals were queried in the GTEx database to identify potential expression quantitative trait loci (eQTLs).

Results: The presence of tremor together with at least one other sign of AWS was associated with a peak located on chromosome 4 ($p =$ 1.11E-07 for the top SNP). Most of the SNPs in this peak are common in European-Americans (MAFs vary from 0.27 to 0.33) and are located in the promoter and regulatory regions of the GABRA2 gene which has been shown to be associated with alcohol dependence. GABRA2 encodes the α2 subunit of the GABAA receptor, a subunit which is involved in receptor trafficking and endocytosis as well as benzodiazepine binding. Based on the GTEx dataset, the variant SNP allele is significantly associated with decreased GABRA2 expression in several brain areas, including cortex, hippocampus and cerebellar hemisphere, suggesting a possible role in AWS physiology. The association analysis that focused on the number of AWS symptoms revealed two SNPs, one located in "gene desert" area on chromosomes 14 and another on chromosome 2, with p-values close to genome wide significant levels ($p =$ 1.8E-08 and 2.5E-08, respectively). An additional strong signal ($p =$ 1.43E-07 for the top SNP) was located on chromosome 4. The SNPs in this peak had low MAFs (0.02) and were located 11 kb 5' of the TDO2 gene which encodes, tryptophan dioxygenase, an enzyme that plays a critical role in tryptophan metabolism. Of note, downstream metabolites in this pathway, namely kynurenic acid and quinolinic acid, are ligands for NMDA receptors, which may be implicated in AWS physiology.

Conclusions: Our pilot study revealed several important findings. First, two phenotypic definitions of AWS included in the analyses revealed strong, albeit different association signals. Therefore, both phenotypes should be included in future large-scale analyses in an effort to replicate and further explore these findings. Second, the strength of association signals discovered in this relatively small sample indicates that the selected phenotypes are robust and have the potential to reveal statistically significant associations in larger samples. Third, these association findings provide preliminary evidence in support of potentially important roles of the GABRA2 and TDO2 genes in AWS physiology, which warrants further investigation with functional studies.

Keywords: Alcohol Withdrawal, GWAS, GABRA2

Disclosure: Nothing to Disclose.

T266. A Major Depressive Disorder GWAS-Supported Variant Near TMEM161B / MEF2C Predicts Putamen Activation During Reward Processing in Alcohol Use Disorder

Christine Muench*, Melanie Schwandt, Jeesun Jung, Carlos Cortes, Reza Momenan, Falk Lohoff

National Institutes of Health/National Institute on Alcohol Abuse and Alcoholism, Bethesda, Maryland, United States

Background: Alcohol use disorder (AUD) frequently co-occurs with major depressive disorder (MDD). While this comorbidity is associated with an increase in disease burden, worse treatment outcomes, and greater economic costs, the underlying neurobiology remains poorly understood. A recent large-scale GWAS of MDD has identified a locus near the TMEM161B / MEF2C genes (rs10514299) as a risk variant; however, the biological relevance of this variant has not been studied so far. Given previous reports of disrupted reward processing in both AUD and MDD populations, we hypothesized that genetic variation in rs10514299 would be associated with differences in striatal BOLD responses during reward/loss anticipation in individuals with AUD.
Methods: DNA samples from 45 recently detoxified patients with AUD and 45 healthy controls (HCs) were genotyped for rs10514299. Participants were administered the Monetary Incentive Delay task in a 3 Tesla MRI scanner. Effects of genetic variation in rs10514299 on putamen BOLD responses during anticipation of high/low reward/loss were investigated. Furthermore, we examined the association between genetic variation in rs10514299 and depression severity, as assessed by the Montgomery-Asberg Depression Rating Scale, in a sample of individuals with a lifetime diagnosis of AUD ($n = 953$).
Results: Our results revealed significant diagnosis-genotype interaction effects on putamen activation. Specifically, AUD patients carrying the T allele showed significantly greater putamen activation during anticipation of high reward ($p = 0.014$), high loss ($p = 0.024$), and low loss ($p = 0.046$) compared to HCs. Additionally, carrying the T allele was significantly associated with greater depression severity in individuals with a lifetime diagnosis of AUD ($\beta = 2.35$, $p = 0.019$).
Conclusions: These findings confirm the GWAS-identified relevance of rs10514299 for depressive symptomatology and extend it to individuals with a lifetime diagnosis of AUD. More importantly, this is the first study to show functional relevance of rs10514299 in a neuroimaging phenotype of reward processing. Current DSM-5 nosology for AUD and MDD is merely descriptive and does not capture the underlying neurobiological pathophysiology. The fact that a MDD risk variant was also shown to have an effect in an AUD neuroimaging sample supports a potential role of this genetic variation in an endophenotype related to reward processing that might cross DSM-5 categories. Future imaging genetics studies are needed to confirm our findings.
Keywords: Alcohol Use Disorder, rs10514299, Depression, Reward Circuitry, TMEM161B / MEF2C
Disclosure: Nothing to Disclose.

T267. Nucleus Accumbens Subnuclei Regulate Motivated Behavior via Direct Inhibition and Disinhibition of VTA Dopamine Subpopulations

Hongbin Yang, Johannes de Jong, YeEun Tak, James Peck, Helen Bateup, Stephan Lammel*

University of California, Berkeley, Berkeley, California, United States

Background: Dopamine (DA) projections from the ventral tegmental area (VTA) to the nucleus accumbens (NAc), which comprise the mesolimbic DA system, play an imporant role in motivated behaviors, reinforcement learning, and reward processing. Dysfunction of this system has been implicated in neuropsychiatric disorders such as substance abuse disorder and depression. While this has led to intense study into DA neurotransmission and the influence of DAergic input to the NAc on motivated behaviors, much less is known about the architecture and function of inhibitory feedback projections from the NAc to the VTA.
Methods: To determine the circuit architecture and function of discrete NAc inputs to the VTA, we combined optogenetics and transgenic mice with neural tracing techniques, slice electrophysiology and animal behavior.
Results: We show that projections from separate NAc subdivisions to the VTA comprise direct and indirect connections that promote inhibition and excitation of distinct mesolimbic DA subpopulations, respectively. In addition, we demonstrate that neurons in the medial shell subregion of the NAc regulate different mesolimbic DA subpopulations via different GABA receptor subtypes. Lastly, using in vivo optogenetic manipulations we establish a critical role for NAc subregion-specific inputs to the VTA in regulating motivated behaviors.
Conclusions: Our results not only bridge the gap between previous anatomical and electrophysiological studies, which have controversially discussed the connectivity of NAc neurons with VTA neurons, but also suggest a remarkable distinction in the incorporation of inhibitory inputs between different subtypes of mesolimbic DA neurons, which have traditionally been considered a homogeneous population.
Keywords: Dopamine, Nucleus Accumbens, Ventral Tegmental Area (VTA), Reward, Motivation
Disclosure: Nothing to Disclose.

T268. Enhancing Effects of Cannabis Smoke Exposure on Working Memory Performance

Barry Setlow*, Shelby Blaes, Caitlin Orsini, Toeisha Stubbs, Shandera Ferguson, Sara Heshmati, Shannon Wall, Marcelo Febo, Adriaan Bruijnzeel, Jennifer Bizon

University of Florida College of Medicine, Gainesville, Florida, United States

Background: Cannabis is the most widely used illicit drug in the United States and worldwide, and cannabis use is reported to cause cognitive impairments. Studies in animal models show that acute administration of delta-9-

tetrahydrocannabinol (THC, the primary psychoactive component of cannabis) or synthetic cannabinoid receptor type 1 (CB1) agonists can impair performance on cognitive tasks that depend upon the hippocampus and prefrontal cortex. Given that the primary route of cannabis use in humans is through smoking, however, comparatively little research in animal models has investigated this route of administration. The primary goal of these experiments was to determine how acute exposure to cannabis smoke affects performance in a delayed response working memory task in rats, which depends upon the integrity of the prefrontal cortex. A secondary goal was to determine whether any such effects differ in males and females, as there are reported sex differences in sensitivity to cannabinoids.

Methods: Adult male ($n = 15$) and female ($n = 16$) Long-Evans rats were trained in a food motivated delayed response working memory task in operant test chambers. In each of the 40-minute sessions in this task, rats performed >100 trials on which they had to remember the location of a response lever over a variable delay period that ranged from 0-24 s. Smoke exposure began once rats reached stable performance in the task. One hour prior to sessions in the working memory task, freely moving rats were placed in a chamber where they were exposed to smoke generated by burning either cannabis (5.3% THC, ~0% CBD) or placebo (0% THC, 0% CBD) cigarettes in an automated cigarette smoking machine. A semi-randomized, within-subjects experimental design was used such that in separate sessions, each rat was exposed to smoke from 0, 1, 3, and 5 cigarettes of each type, with at least a 48 h washout period between successive exposures. Subsequent experiments evaluated the effects on working memory performance of acute administration of THC (0, 0.3, 1.0, 3.0 mg/kg) and the CB1 receptor antagonist rimonabant (SR141716A; 0, 0.2, 0.6, 2.0 mg/kg).

Results: Prior to smoke exposure sessions, male rats performed more accurately than females on the working memory task, particularly at longer delays. Analysis of performance in females revealed no effects of estrous cycle. Exposure to smoke from cannabis cigarettes had no effects on working memory accuracy in males, but significantly and dose-dependently enhanced accuracy in females. Acute exposure to placebo smoke had no effects on accuracy in either sex. Initial data suggest that acute administration of the highest doses of both THC and rimonabant tended to impair performance in both sexes.

Conclusions: The results of these experiments suggest that passive exposure to cannabis smoke can enhance performance on a prefrontal cortex-dependent working memory task. The fact that this effect was evident only in female rats is consistent with evidence that females are more sensitive than males to the behavioral effects of cannabinoids under some conditions. The absence of these enhancing effects following placebo smoke exposure suggests that smoke itself (or stress/arousal resulting from passive smoke exposure) was not the cause of the enhanced performance. Ongoing experiments will determine whether enhancing effects of cannabis smoke on working memory are mediated through CB1 receptors, and whether these effects are evident in other forms of cognition.

Keywords: Cannabis, Marijuana, Working Memory, Rats, Sex Differences

Disclosure: Nothing to Disclose.

T269. Novel Efficacy Endpoints Based on Shifts in the World Health Organization (WHO) Risk Levels of Drinking: Treatment Effects in Alcohol Pharmacotherapy Trials

Daniel Falk*, Raye Litten, Henry Kranzler, Karl Mann, Stephanie O'Malley, Katie Witkiewitz, Megan Slater, Raymond Anton

National Institutes of Health, Rockville, Maryland, United States

Background: Alcohol consumption endpoints are the main efficacy endpoints reported in pharmacotherapy trials to treat alcohol use disorder (AUD). At least a dozen such endpoints, both continuous and dichotomous, have been used in practice. The US Food and Drug Administration currently recommends two dichotomous endpoints for pivotal trials: total abstinence and no heavy drinking (FDA, 2015); however, there may be limitations in their sensitivity to detect the effects of medication for certain trial designs and populations. Thus, there is interest in alcohol treatment community to develop, assess, and validate new dichotomous endpoints that may be more sensitive (i.e., have larger treatment effects), while still being clinically meaningful. The current study, conducted by the Alcohol Clinical Trials Initiative (ACTIVE) (Anton et al., 2012), evaluated the sensitivity of two relatively novel endpoints that are based on shifts in the World Health Organization (WHO) risk levels of drinking (EMA, 2010) – the percentage of subjects who reduce their risk level by at least 1 and 2 levels.

Methods: For this secondary data analysis, raw data were obtained from five large, multisite alcohol pharmacotherapy trials that evaluated the efficacy of four medications: naltrexone, topiramate, varenicline, and nalmefene. In their primary publications, these trials showed statistically significant medication efficacy for at least one traditional endpoint of alcohol consumption (Anton et al., 2006; Gual et al., 2013; Mann et al., 2013; Litten et al., 2013; Johnson et al., 2007). Treatment effects (Cohen's h), capturing the medication-placebo differences for the new WHO 1- and 2-shift endpoints, were calculated and compared to treatment effects obtained from traditional continuous and dichotomous endpoints.

Results: In all four trials, statistically significant and comparable treatment effects could be obtained using both the WHO 1- and 2-shift endpoints (Cohen's h, small to medium effects: 0.19 – 0.56, p's $<.05$); although a grace period may be required to obtain maximal effects. Moreover, the magnitude of these treatment effects was often as large, or larger, than those obtained by using more traditional continuous and dichotomous endpoints.

Conclusions: These findings suggest that the WHO 1- and 2-shift may be worthy of further development as endpoints in alcohol pharmacotherapy trials. Future research should replicate these findings in other multisite trials, as well as validate the clinical utility of risk level shifts against clinically meaningful correlates using a variety of data sources.

Keywords: Alcohol Drinking, Clinical Trials, Novel Endpoints, Pharmacotherapy

Disclosure: Nothing to Disclose.

T270. Role of Oxytocin in a Mouse Model of PTSD-AUD Comorbidity

Howard Becker*, Courtney King, William Griffin, Marcelo Lopez

Medical University of South Carolina, Charleston, South Carolina, United States

Background: While there is a high prevalence for the co-occurrence of alcohol use disorders (AUD) and post-traumatic stress disorder (PTSD), the mechanisms underlying these disorders are not fully understood and few treatments are effective for those suffering with PTSD-AUD comorbidity. A growing body of literature suggests that the oxytocin (OT) system plays a role in a number of stress-related psychiatric disorders including PTSD and alcohol addiction. We have recently developed a mouse model of PTSD involving chronic exposure to a predator odor linked to alcohol self-administration and relapse procedures that demonstrates sensitization of male and female mice to later acute stress-induced reinstatement of alcohol relapse-like behavior. The present studies examined the ability of systemic administration of OT to attenuate this sensitized stress-related increase in alcohol seeking and drinking behavior.
Methods: Adult male and female C57BL/6J mice were trained using standard operant procedures to lever respond on a fixed ratio (FR4) schedule for 12% (v/v) alcohol (20 ul) during daily 20-min sessions. Once stable FR4 responding was established, half of the mice were exposed 15 min to a predator odor (2,3,5-Trimethyl-3-thiazoline; TMT) over 5 consecutive days while the remaining mice were similarly treated but just exposed to air. Baseline alcohol responding was re-established and then mice were tested under extinction conditions for ~14 days prior to testing relapse behavior. For reinstatement testing, all mice were exposed to TMT for 15 min and immediately placed into operant self-administration chambers to examine alcohol-seeking and drinking behavior. Separate groups of mice were injected (ip.) with OT (0.1-1 mg/kg) or vehicle 30 min prior to the reinstatement test session.
Results: Acute stress (TMT) exposure increased alcohol seeking behavior and this effect was significantly enhanced in male and female mice with a prior history of chronic TMT exposure. This sensitized stress effect in mice with a history of prior chronic stress experience was found to be long lasting (>60 days) and the sensitized effect also generalized to exposure to context cues associated with prior chronic TMT exposure. Systemic administration of OT reduced stress (TMT)-induced alcohol relapse-like behavior in a dose-related manner in males and females, with females showing greater sensitivity (shift to left in OT dose-effect function). Further, OT (1 mg/kg) blocked the sensitized stress response in male and female mice with prior chronic stress (TMT) experience.
Conclusions: Results indicate that chronic stress (TMT) exposure sensitizes mice to subsequent acute stress challenge. In our model of PTSD-AUD comorbidity, this sensitized stress response is demonstrated by enhanced stress-induced alcohol relapse like behavior. As such, the model appears to capture many of the key clinical features of PTSD as well as the ability of this stress disorder to aggravate alcohol relapse and problem drinking. Additionally, results suggest that systemic administration of OT effectively attenuates stress-induced relapse-like behavior in male and female mice. Collectively, this work suggests an apparent therapeutic effect of oxytocin treatment for PTSD-AUD comorbidity.
Keywords: PTSD, Alcohol Use Disorder, Oxytocin
Disclosure: Nothing to Disclose.

T271. Feature Selection and Classification for DNA Methylation-Based Subtypes of Substance Misuses

Xinyu Zhang, Ying Hu, Amy Justice, John Krystal, Ke Xu*

Yale University School of Medicine, New Haven, Connecticut, United States

Background: Opiate and cocaine misuses are common in the United States and have significant impacts on public health. Detection and differentiation of opiate and cocaine misuses are clinically challenging due to a short half-life in 2-5 days for each substance. However, chronic opiate and cocaine misuse alters epigenome architecture of DNA methylation. Thus, DNA methylation signatures may serve as a biomarker to differentiate chronic opiate and cocaine misuses. Here, we report the first study of applying a machine-learning approach to distinguish chronic opiate and cocaine misuses based on genome-wide DNA methylation data.
Methods: We selected 130 opiate users, 113 cocaine users, and 120 non-drug users from Veteran Aging Cohort Study. Among drug users, 43 participants used both opiate and cocaine. All participants were male African Americans and all were tested HIV-positive. DNA methylation in blood was profiled using Illumina Infinium Human Methylation 450 BeadChip. After probe quality control and normalization, we applied the following steps for feature selection and subtype classification of opiate, cocaine, and comorbid misuse: 1) CpG filter: We performed 3 mixed models separately for opiate misuse, cocaine misuse, and comorbid of opiate and cocaine misuses with batch as a random effect in each model, covariates of age, total white blood cells count, and 6 cell type compositions in blood (CD8+ T-lymphocytes, CD4+ T-lymphocytes, Natural Killer cell, B cells, Monocytes and Granulocytes). -log p value for each CpG resulted from each mixed model was summed up and top-ranked probes (sum -log p >=3) were selected for subsequent analyses; 2) We used two machine learning methods, Random Forest (RF) and Neutral Network (NN) separately to weight each selected CpG from step 1. RF is an ensemble learning method by constructing decision trees at the training time and predicting individual trees in the test for classification each group. NN is a non-linear statistical data modeling tool to capture the statistical structure in an unknown joint probability distribution between observed variables. Each method weighted an importance of each CpG. Then, a combined importance from RF and NN for each CpG was summed up. 3) Importance-based CpGs were re-ranked and were divided as subsets of CpGs to test prediction accuracy for each subset-CpG. 4) To identify best performance features for each subset of CpGs, we performed RF and NN separately in R caret package with 10-fold cross validations. 5) Based on prediction accuracy from

RF and NN, the final sets of CpG sites with minimal number and best performance were determined. 6) We used t-distributed stochastic neighbor embedding (t-SNE) algorithm to reduce dimensions and visualized the results in a scatter plot. Unlike principal component analysis, T-SNE is a non-linear and non-parametric method to reduce high dimensions to low dimensions based on probability distribution of methylation-phenotype similarity.
Results: After probe quality control, a total of 437,722 probes were used for analysis. With sum -log p $>=3$ as a cutoff, 6,261 CpG sites were used for the feature selection. Among 6,261 CpG sites, 30 CpG-sets were constructed based on the importance rank. To differentiate opiate and cocaine misuses, prediction accuracy of each CpG-set was estimated in a range of 45% to 65% for both RF and NN methods. Three sets of CpG sites including 235-CpGs, 1,311-CpGs, and 6,045-CpGs for dimension reduction analysis. Final, a set of 1,311 CpG sites showed the best performance to differentiate opiate and cocaine misuse. On a plot of t-SNE with perplexity 70, which is parameter of estimating the number of close neighbors each point, participants with opiate and cocaine misuse were separately clustered, and participants of misusing both opiate and cocaine spread out across two clusters. These 1,311 CpG sites were located on 863 genes in the genome.
Conclusions: The results from our preliminary analysis suggest that DNA methylation-based feature selection can identify subtypes of substance abuse. Despite a lack of the understanding underlying biology for each substance misuse, DNA methylation profile in blood may serve as a biomarker to dissect the complexity of substance misuse.
Keywords: DNA Methylation, Opiate, Cocaine, Machine Learning, Feature Selection
Disclosure: Nothing to Disclose.

T272. Developing Improved Preclinical Animal Models: Cross-Species Continuity in Cognition-Related Behavior

Brian Kangas*, Jack Bergman

Harvard Medical School, McLean Hospital, Belmont, Massachusetts, United States

Background: The importance of understanding how psychoactive drugs, including both commonly abused drugs and therapeutic medications, modify complex behavioral processes (e.g., learning, memory, motivation, attention) has been increasingly recognized. There has also been a corresponding awareness of the need for new translational animal models that are explicitly designed to capture features of behavioral phenotypes implicated in various neuropsychiatric disorders. In this regard, the stalled development of innovative psychotherapeutics has been attributed, at least in part, to the inadequacy of current animal models. Modern touch-sensitive technology provides an extremely flexible means to address the above-described need. First, this technology permits the exposure of laboratory animals and human subjects to a battery of similar complex behavioral tasks that are designed to assay multiple dimensions of cognitive function before, during, and after drug administration. Additionally, touchscreen-based apparatus can be designed to allow a broad-based, yet dynamic, interface capable of methodological possibilities limited only by the experimenter's imagination and programming abilities.
Methods: Novel touch-sensitive experimental chambers were developed for 4 common laboratory animals (rats, marmosets, squirrel monkeys, rhesus macaques). Modern variants of several gold standard cognition-related tasks (e.g., repeated acquisition, discrimination reversal, delayed matching-to-sample, psychomotor vigilance, progressive ratio) were assessed across animals to examine cross-species continuity on these complex behavioral endpoints and, as well, refine their ability to serve in a battery to assess psychoactive drugs. This battery was then employed in squirrel monkeys to assess the effects of synthetic, phyto-, and endo-cannabinoid agonists to determine relative potencies on various aspects of cognitive function.
Results: Tailored chamber specifications and software-based programmatic accommodations engendered orderly cognition-related behavior in all animal models and species tested. In the repeated acquisition task, the prototypical learning set phenomenon was observed with an acceleration rate that correlated across species with their evolutionary distance from humans (i.e., rat $>$ marmoset $>$ squirrel monkey $>$ rhesus macaque). Other tasks such as psychomotor vigilance and progressive ratio revealed negligible differences in baseline performance across rat and nonhuman primate subjects. Moreover, functional similarities across species were observed following temporal or reinforcer magnitude manipulations in, respectively, psychomotor vigilance and progressive ratio performance. The effects of a variety of cannabinoid agonists in squirrel monkeys confirmed that the battery of complex cognition-related behavioral tasks can effectively reveal important potency differences related to task complexity (discriminative capability $<$ learning $<$ cognitive flexibility $<$ short-term memory).
Conclusions: Although performances may differ based on dissimilar physiology, hardware specifications and behavioral programs can be tailored to accommodate such differences and maximize assay flexibility across species. The present touchscreen data provide an example of formally equivalent translational studies across species and furthermore suggest that, depending on experimental and pharmacological goals, this approach can be used as an indicator of appropriate model and species. More generally, the present studies illustrate available and relatively inexpensive means by which traditional cognitive assessment can be modernized for laboratory research. Perhaps the most important feature of modern touch-sensitive technology, however, lies in the potential for designing completely novel animal models to capture important dimensions of complex behavior and cognitive function that emerge through laboratory and clinical discovery.
Keywords: Animal Models, Touchscreen, Cognitive Function, Nonhuman Primates, Rats
Disclosure: Nothing to Disclose.

T273. Effects of Cocaine and Chronic Stress Exposure on Neuronal Activity in the Basolateral Amygdala

Soumyabrata Munshi, Aaron Caccamise, Marina Wolf, J. A. Rosenkranz, Jessica Loweth*

Rowan University School of Osteopathic Medicine, Stratford, New Jersey, United States

Background: A major challenge for treating cocaine addiction is the propensity for abstinent users to relapse. Two important triggers for relapse are cues associated with prior drug use and stressful life events. To study their interaction in promoting relapse during abstinence, we used the incubation model of craving and relapse in which cue-induced drug seeking progressively intensifies ("incubates") during withdrawal from extended-access cocaine self-administration. We have recently shown that exposure to repeated but not acute restraint stress during the first two weeks of withdrawal accelerates the initial rate of incubation of cue-induced cocaine craving, although craving plateaus at the same level observed in controls. These data indicate that chronic stress exposure during early withdrawal may result in increased vulnerability to cue-induced relapse during this period. Previous studies have shown that chronic stress exposure enhances excitatory drive to the basolateral amygdala (BLA), a region critical for behavioral responses to stress. Given that glutamate projections from the BLA to the NAc are critical for incubation of cocaine craving, we hypothesized that cocaine withdrawal and chronic stress exposure produce a synergistic increase in BLA neuronal activity which accelerates incubated craving.
Methods: In vivo single-unit extracellular recordings from anesthetized rats were conducted to compare neuronal activity in the BLA of rats that self-administer saline or cocaine (6h/day for 10 days) and then either undergo repeated restraint stress or control conditions during the first two weeks of withdrawal. Recordings were conducted between weeks 2 and 3 of withdrawal from extended-access cocaine or saline self-administration (0-5 days after the last restraint stress or control session).
Results: Our data indicate that, compared to saline-exposed controls, cocaine exposure alone enhances neuronal activity within the BLA (measured as an increase in spontaneous firing rate). We also found that, compared to cocaine-exposed controls, repeated stress exposure during withdrawal from cocaine self-administration further enhances BLA neuronal excitability.
Conclusions: These studies indicate that chronic stress exposure during withdrawal from cocaine self-administration may further enhance BLA neuronal excitability, which could contribute to the acceleration of incubation of cocaine craving observed in these animals. Future studies will continue to assess the synergistic effects of cocaine and chronic stress exposure during withdrawal on cellular and behavioral measures and, using a stress resilience model, identify neuroadaptations that can reverse such effects. Together, these studies will ultimately bring us closer to developing effective pharmacotherapies to prevent relapse.
Keywords: Cocaine Self-Administration, Basolateral Amygdala, Chronic Stress, Electrophysiology
Disclosure: Nothing to Disclose.

T274. The Effect of Childhood Trauma on Mood, Cannabis Use and Stress Response in Recreational Cannabis-Using Men and Women

Stephanie Reed*, Margaret Haney, Payal Pandya, Suzette Evans

Columbia University/New York State Psychiatric Institute, New York, New York, United States

Background: It is well established that early life stress, particularly during childhood, produces enduring neurobiological changes and increases stress reactivity in adulthood. Individuals exposed to childhood trauma are also at increased risk for substance use disorders (SUD), including cannabis. Clinical evidence shows that stress exposure is associated with increased drug craving, drug use and relapse, and that women are more likely to relapse, and relapse more quickly, than men. However, most studies have focused on individuals who meet criteria for a SUD and who are seeking treatment. Cannabis is one of the most widely used drugs in the United States and its use has been steadily increasing, in part related to the changing political and legal landscape. Little is known about the factors that contribute to the transition from recreational to problematic cannabis use. One rarely studied factor is a history of childhood trauma. The findings reported herein represent a secondary analysis of data collected as part of a study that examined the effects of oxytocin on stress reactivity in recreational cannabis using men and women. In that study, we assessed trauma history in all participants. Our objective was to (1) evaluate whether the Early Trauma Inventory (ETI), an extensive clinician-administered early trauma interview, was associated with current self-reported cannabis use, mood and distress, as well as the response to the Trier Social Stress Test (TSST), and (2) assess whether these associations differed between male and female recreational cannabis users, a group at risk for developing severe cannabis use disorders.
Methods: The ETI was conducted with 63 recreational cannabis users (32 women; 31 men). Trauma history was compared between men and women and correlations between trauma history and self-reported current cannabis use (amount and frequency), distress tolerance, anxiety, and depression was examined; note, all Axis I psychiatric disorders, including trauma-, depression-, and anxiety-related disorders, were exclusion criteria. Associations between trauma history and the subjective response to the TSST (negative affect, anxiety, and cannabis craving) were also explored.
Results: Of the 61 participants, all reported at least some exposure to trauma during childhood. There were no sex differences in cannabis use; men and women reported using cannabis approximately 3.7 days per week, smoking approximately 5.6 joints per week and smoking approximately 1.7 joints per smoking occasion. Trauma history was negatively correlated with number of days of smoking cannabis per week in women, but positively correlated with amount of cannabis smoked per week in men. Trauma history was also positively correlated with self-reported depression and anxiety and negatively correlated with distress tolerance in women, but not men. When examining the subscales of the ETI, the incidence of general, emotional and sexual childhood trauma was similar between men and women, but men reported more physical trauma than women. Despite this, after exposure to the TSST, a history of physical trauma was positively correlated with subjective ratings of negative affect, particularly in women. There were no associations between trauma history and TSST-induced cannabis craving or anxiety scores.

Conclusions: All of the recreational cannabis-using participants in the current study reported a history of childhood trauma. Incidence of early trauma was generally similar in men and women except men reported more physical trauma than women. However, women appeared to be more affected by their childhood trauma than men in that greater trauma exposure was associated with greater self-reported depression, anxiety and distress only in women. In addition, despite more reported physical trauma in men, physical trauma was associated with negative subjective response to acute stress exposure only in women. Lastly, childhood trauma had an opposite effect on current cannabis use in men and women; trauma exposure was negatively associated with cannabis use in women but positively associated with cannabis use in men. Together these data indicate that even among healthy recreational cannabis smokers, there is a high level of childhood trauma, suggesting that trauma history should be taken into account when examining the progression of cannabis use in future studies. Further, childhood trauma exposure may have a greater negative impact on mood in cannabis-using women than cannabis-using men, particularly during stressful situations, whereas men may use more cannabis as a result of childhood trauma exposure than women. These sex differences should be considered in future cannabis treatment development. Supported by NIDA grant R01A035850.

Keywords: Cannabis Use, Sex Differences, Childhood Trauma

Disclosure: Nothing to Disclose.

T275. Alcohol Effects on Emotion Processing: Activation, Performance & Functional Correlates

Lauren Hoffman, Sara Nixon*, Robert Prather, Julianne Price

University of Florida, Gainesville, Florida, United States

Background: Although alcohol-related deficits in emotion processing are receiving increased programmatic study, current research is inconsistent and infrequently integrates functional and neurobehavioral domains. Clarifying potential interactions among these domains informs current neurobehavioral models of substance use disorders as well as initiatives directed to enhancing neurocognitive recovery/compensation in early abstinence. Thus, based on extant literature and our working model, we examine behavior (accuracy, reaction time), neurophysiology (event related potentials: N170, P3), and interpersonal problems (IIP-64) in men and women seeking treatment for an alcohol use disorder (AUD) and community controls. We anticipated alcohol-related deficits in emotion processing, regardless of valence. However, we anticipated that deficits in negative emotions would be more robust. While expecting correlations between the IIP-64 and processing, we asked to what extent the IIP-64 were differentially related to emotion processes.

Methods: Detoxified, male and female treatment-seekers with an AUD ($n = 34$) were compared with age/education equated controls ($n = 39$) on a battery of neurocognitive tests including variations of the emotional face expression task (here, emotion judgement task (EJT)) and a control task requiring sex judgement (male vs female). Faces were selected from the Ekman stimulus set. Emotions (i.e., angry, happy, sad) were morphed with neutral expressions to depict 35%, 65%, and 95% of the target emotion using Fantamorph software (V.5.4.3). Using similar techniques, sex was morphed to these same percentages. Accuracy and reaction time for emotion and sex identification was recorded via button press. ERPs were collected using electro-caps with the I 10-20 system. For this analysis, the T5 and T6 sites were applied for the N170 and PZ was applied for the P3. Testing occurred in an electrically shielded/ sound attenuated booth. Treatment-seekers had a minimum of 21 days of sobriety and were not recently exposed to psychoactive medications. All participants were required to produce negative breath and urine tests for alcohol or other drugs (except nicotine). All participants completed the Inventory of Interpersonal Problems (IIP-64) to assess level of interpersonal difficulty.

Results: After correcting for multiple comparisons, there were no group differences on any outcomes for the sex judgement task or EJT. Hypothesis driven follow-up analyses omitted the 35% emotion morph due to exceptional difficulty. These analyses identified less accurate performance in the AUD group relative to controls ($p = .05$). This effect appeared to be driven by the anger condition, which produced the largest group mean difference at both remaining morph levels. There were no group differences in reaction time. Neurophysiologic data supported behavioral differences with the AUD group exhibiting lower P3 amplitude ($p = .04$) and the largest group mean difference occurring at the 65% morph level of anger. There were no group differences for N170 amplitude. AUD participants had significantly higher total scores on the IIP-64 ($p = .0005$) and all subscales (p's $< .05$). Total scores negatively correlated with accuracy on both the 65% ($r = -.36$, $p = .04$) and 95% ($r = -.37$, $p = .03$) morph levels of anger in the AUD group. When further investigated by subscale, performance on 65% and 95% anger was negatively associated with the intrusive/needy subscale ($r = -.34$, $p = .05$; $r = -.44$, $p = .01$, respectively); accuracy for the 65% morph was also associated with the nonassertive subscale ($r = -.37$, $p = .03$) whereas accuracy on the 95% morph was associated with the overly accommodating subscale ($r = -.36$, $p = .04$). There were no significant correlations for the control group.

Conclusions: Findings suggest that newly abstinent individuals with an AUD exhibit atypical emotional face processing as identified by subtle behavioral and ERP differences. Although the absence of group by emotion interactions suggests that this pattern is not valence-specific, group means suggest that differential response to anger is largely responsible for this outcome. In addition to the neurobehavioral alterations, AUD individuals endorsed greater interpersonal problems than controls. Of particular clinical relevance, interpersonal problems were correlated with aberration in anger processing. Future efforts should be directed to exploring potential sex differences (currently underway) and determining the degree to which deficits in emotion processing are related to post-treatment adaptation.

Keywords: Alcoholism, Emotion Processing, EEG/ERP Electrophysiology

Disclosure: Nothing to Disclose.

T276. Trial-By-Trial Analysis of the Behavioral and Neural Mechanisms of Simulated Drug-Related Choice in Currently-Using and Abstinent Individuals With Cocaine Use Disorder

Scott Moeller*, Anna Zilverstand, Anna Konova, Prantik Kundu, Muhammad Parvaz, Rebecca Preston-Campbell, Keren Bachi, Nelly Alia-Klein, Rita Goldstein

Stony Brook University School of Medicine, Stony Brook, New York, United States

Background: The choice for drugs over alternative reinforcers is core to drug addiction. This phenomenon is well-captured by the drug-choice procedure, where an individual selects between a drug reinforcer and a non-drug reinforcer (e.g., money). However, the neural basis of drug-choice is not well-understood, particularly in individuals with long-term abstinence who cannot ethically be offered drug-using opportunities; abstainers may differ from active users on drug-choice and underlying circuitry. Common tasks used in abstainers, such as attention bias, do not capture the choice aspects of drug administration studies and might engage different circuitry. Therefore, here we investigated value-based decision-making for viewing drug images, rather than receiving actual drugs, using a new fMRI drug-choice task that was directly inspired by the neuroeconomics of simple choice. We tested for group differences in the overall value of cocaine-related images, and (separately) in the trial-by-trial process of computing such value, in currently-using and abstaining individuals with cocaine use disorder (CUD) compared with matched healthy controls. We expected group activation differences in brain regions that compute and compare reward values.
Methods: Thirty-seven individuals with a DSM-IV history of CUD (18 current: median 3 days abstinent; 19 remitted: median 365 days abstinent) and 26 controls viewed, in an event-related manner, images depicting food (e.g., hamburger), threat (e.g., pointed gun), or cocaine (e.g., razor) (75 trials per category). On each trial, participants indicated their choice (Strong No, No, Yes, or Strong Yes) to view the currently available image or a neutral reference image (wicker basket). Scanning occurred shortly before lunch on a Siemens 3T Skyra, using Human Connectome Project acquisition parameters. Behavioral analyses were conducted trial-by-trial on choice preference (i.e., Strong No to Strong Yes, linearly coded 1-4) and separately on choice certainty (i.e., the parabolic ordering of these preferences); predictors were diagnosis, image type, reaction time, and baseline craving. Imaging data were modeled from stimulus onset until decision, and were analyzed using general linear models (GLMs), where the trial-by-trial fMRI response at the decision phase was correlated with the trial-by-trial choice preference value and (separately) the trial-by-trial choice certainty value. Finally, to test the direct "preference difference" between the image categories, additional ANOVAs analyzed the averaged responses for cocaine minus food (i.e., cocaine > food), for both behavior (choice preference and certainty) and brain (averaged activations on cocaine trials minus those of food trials). Significance was set at $p < 0.05$ for behavior, and conservatively at $p < 0.01$ corrected for imaging.
Results: Trial-by-trial behavioral analyses revealed Diagnosis × Image Type interactions: active CUD had the highest preference ratings and lowest certainty ratings, especially during cocaine trials. Similarly, at the participant level, active CUD provided the highest relative choice preferences, and lowest relative choice certainty, for cocaine versus food images. The fMRI data showed that, across image categories and study groups, activations in the midcingulate cortex were negatively correlated with trial-by-trial preference ratings, and activations in visual regions (e.g., occipital cortex) were positively correlated with trial-by-trial choice certainty ratings. When examining group differences, all CUD participants had more positive associations (i.e., higher beta values) than controls between the trial-by-trial activations in the DLPFC and choice certainty. Finally, at the participant level, both CUD groups had higher activations than controls when making decisions about cocaine (versus food) images in the dorsal anterior cingulate cortex (dACC)/supplementary motor area, and active CUD had higher activations than other groups in the midbrain. In all participants, higher cocaine > food dACC activations correlated with lower cocaine > food choice certainty, and higher cocaine > food midbrain activations correlated with higher cocaine choice preference.
Conclusions: Active CUD had the greatest decision difficulty during drug-related choice: their cocaine choice preference ratings neared neutral (while other groups showed aversion), and their certainty ratings indicated ambivalence (while the other groups showed conviction). This heightened cocaine-choice in active CUD is consistent with prior work (including our own) linking real or simulated drug-choice to drug use recency. For fMRI, CUD participants had higher cocaine > food activations in the dACC, a region previously linked to choice difficulty in neuroeconomic contexts. In contrast, activations in the dopaminergic midbrain may have contributed to the enhanced drug-related choice in active CUD. Finally, as no group differences emerged on trial-by-trial modulation of choice preference, our results suggest that although CUD and controls differ on what stimuli they value, they engage in similar processes when computing such value. Current findings could inform intervention efforts that aim to decrease the value of drug stimuli and increase the value of alternative reinforcers. More broadly, these results suggest that the neural mechanisms underlying drug-biased choice in human addiction may involve brain regions that contribute to resolving difficult decisions (i.e., in addition to those that compute value or reward).
Keywords: Drug Addiction, Decision-Making, Functional MRI (fMRI), Drug Cues, Choice
Disclosure: Nothing to Disclose.

T277. Epigenetic Regulation of the Dynorphin and Neuropeptide Y Systems: A Role in Rapid Tolerance to Anxiolytic Effects of Ethanol

Subhash Pandey*, Martina Palmisano, Harish Krishnan, Huaibo Zhang

Center for Alcohol Research in Epigenetics, University of Illinois at Chicago & Jesse Brown VA Medical Center, Chicago, Illinois, United States

Background: Development of alcohol tolerance has been associated with promoting dependence as well as higher

alcohol intake. It has been shown that the dynorphin (DYN) / kappa opioid (KOP) receptor and neuropeptide Y (NPY) systems contribute to the negative reinforcing effects of alcohol. We investigated here the role of DYN/KOP system and its interaction with NPY in an animal model of rapid ethanol tolerance (RET) focusing on the amygdala (AMY), a brain region known to be associated with fear, anxiety and alcoholism.
Methods: Adult male Sprague-Dawley rats were acutely treated with ethanol (1g/kg intra-peritoneal) or n-saline once or twice (24hr apart). Anxiety-like behaviors were measured using the elevated plus maze (EPM). Immediately after behavioral measurement, AMY brain regions were collected for biochemical studies. mRNA levels were measured using qPCR and histone methylation/acetylation of genes were measured by the chromatin immunoprecipitation (ChIP) assay.
Results: It was found that one injection of ethanol exerts anxiolytic effects while two injections do not produce anxiolysis, indicating development of rapid tolerance. Gene expression analysis revealed that ethanol induces a significant up-regulation of PDYN mRNA levels that remain increased after the second injection. On the other hand, NPY but not KOP mRNA levels were increased in the AMY by acute ethanol exposure. However, during ethanol tolerance an increase of KOP receptor gene expression, but no change in NPY mRNA levels, was observed in the AMY. The changes in PDYN and KOP receptor gene transcription do not appear to be mediated by H3K9/14 acetylation or H3K9 dimethylation since no correlation between these histone modifications at the gene promoters and gene expression was observed. However, high levels of H3K27 trimethylation and H3K4 trimethylation were observed in these promoter regions suggesting that the PDYN and KOP gene promoters could be considered bivalent promoters and these epigenetic modifications may be involved in their gene expression. Acute ethanol increased H3K9/14 acetylation of the NPY gene in the AMY that was normalized during RET. Interestingly, nor-BNI (KOP antagonist) pretreatment prevented RET by increasing NPY mRNA levels in the AMY of tolerant animals via increased H3K9/14 acetylation of NPY gene.
Conclusions: These are first data to suggest DYN/KOP system interacts with NPY through epigenetic mechanisms that could be involved in the regulation of rapid tolerance to the anxiolytic effects of ethanol (Supported by NIH-NIAAA P50AA022538, UO1AA-019971, RO1AA-010005 and by the VA Senior Research Career Scientist award to SCP and a fellowship from University of Bologna to MP).
Keywords: Dynorphin, Kappa Opioid Receptor, Alcohol, Epigenetic Regulation, Amygdala
Disclosure: Nothing to Disclose.

T278. Methylomic Profiling and Replication Implicates Deregulation of PCSK9 in Alcohol Use Disorder

Falk Lohoff*, Jill Sorcher, Allison Rosen, Kelsey Mauro, Rebecca Fanelli, Reza Momenan, Colin A. Hodgkinson, Leandro Vendruscolo, George Koob, Melanie Schwandt, David George, Andrew Holmes, Bin Gao, Hui Sun, Christine Muench, Zachary Kaminsky

National Institutes of Health/National Institute on Alcohol Abuse and Alcoholism, Bethesda, Maryland, United States

Background: Alcohol Use Disorder (AUD) is a common and chronic disorder with substantial effects on personal and public health. The underlying pathophysiology is poorly understood but strong evidence suggests significant roles of both genetic and epigenetic components.
Methods: Given that alcohol affects many organ systems, we performed a cross-tissue and cross-phenotypic analysis of genome-wide methylomic variation using Illumina HM450 and EPIC chip arrays in AUD samples from 3 discovery, 4 replication, and 2 translational cohorts. The discovery samples consisted of postmortem brain tissues ($n=46$), bloods form a resting-state functional connectivity imaging endophenotypes ($n=68$) and postmortem brain tissues sorted into neuronal and non-neuronal cells ($n=58$).
Results: Overrepresentation analyses identified 68 significant CpG probes of which the most significantly associated probe cg01444643 was in in the promoter of the proprotein convertase subtilisin/kexin 9 (PCSK9) gene ($p=0.002$). Biological validation showed that PCSK9 promoter methylation is conserved across tissues (brain-blood-liver) and positively correlated with expression. Replication in AUD datasets confirmed PCSK9 hypomethylation ($n=392$, $p<0.05$) and a translational mouse model of AUD showed that alcohol exposure leads to PCSK9 mRNA and protein downregulation ($p<0.0001$). Postmortem human liver tissue analyses in control ($n=47$) and liver transplant cases due to alcohol cirrhosis ($n=50$) showed marked increase of methylation at cg01444643 ($P<0.0001$) and significantly decreased PCSK9 expression ($p<0.01$). PCSK9 is primarily expressed in the liver and regulates low density lipoprotein cholesterol (LDL-C).
Conclusions: Our finding of alcohol-induced epigenetic regulation of PCSK9 represents one of the underlying mechanisms between the well-known effects of alcohol on lipid metabolism and cardiovascular risk, with light alcohol use generally being protective while chronic heavy use has detrimental health outcomes.
Keywords: Epigenetics, Alcohol, Lipids, Cholesterol Biosynthesis
Disclosure: Nothing to Disclose.

T279. History of Cocaine Self-Administration Alters Transcriptome-Wide Responses to Cocaine Re-Exposure Throughout the Brain's Reward Circuitry

Deena Walker*, Erin Calipari, Hannah Cates, Eddie Loh, Immanuel Purushothaman, Arthur Godino, Philipp Mews, Li Shen, Eric Nestler

Icahn School of Medicine at Mount Sinai, New York, New York, United States

Background: Cocaine addiction is a chronic, relapsing disorder involving maladaptive plasticity in brain reward circuits associated with changes in gene expression. The behavioral responses to self-administration withdrawal and re-exposure have been well-defined in rodent models. However, the gene expression changes underlying the observed circuit wide and behavioral dysregulation remains elusive. We hypothesized that chronic cocaine "re-programs" the transcriptome, resulting in sensitization and desensitization of molecular targets upon re-exposure to cocaine.

Methods: To test this hypothesis, we assigned male mice to one of six groups: saline or cocaine self-administration (SA) + 24 hr withdrawal (WD) (S24/C24); saline/cocaine SA + 30 d WD + acute saline/cocaine + 1 hr (SS/SC/CS/CC). Then we conducted RNA-sequencing on 6 interconnected reward-associated brain regions (PFC, NAc, DS, BLA, vHIP, VTA) to determine how a history of cocaine SA influences transcriptome-wide responses at baseline and after cocaine re-exposure. We focused on patterns of gene expression that were altered in response to specific stimuli when compared to the same baseline (S24).
Results: Genes that were uniquely altered by acute cocaine after cocaine SA+WD (CC) displayed region-specific regulation, with the greatest numbers seen in NAc, DS, and BLA. Further analysis revealed that the transcription factor, HNF4A, is a predicted upstream regulator in all three regions and associated with genes involved with dopamine signaling and long-term potentiation. In contrast, genes uniquely altered by cocaine SA + acute saline (CS), were most affected in NAc and PFC, associated with immune signaling and predicted to be regulated by CREB1. We also utilized factor analysis to identify genes whose regulation was associated with specific behaviors. We found that genes associated with cocaine intake and infusions are sensitive to cocaine re-exposure in the PFC and NAc. Interestingly, genes negatively associated with cocaine intake in the NAc are further downregulated by cocaine re-exposure when compared to acute cocaine (SC), an effect not seen in the PFC. Rather genes positively and negatively associated with intake in the PFC are up- and downregulated after 24 hr WD, but not after cocaine re-exposure. Fisher's Exact testing revealed significant overlap ($p < 0.001$) of genes positively, but not negatively, associated with intake in the NAc and PFC and significant overlap of genes associated with intake and genes upregulated by cocaine re-exposure.
Conclusions: These data provide further insight into the relationship between these correlated brain regions. Further, this is the most comprehensive picture to date of transcriptome-wide regulation by cocaine SA and WD throughout the brain's reward circuitry, and it will guide future studies of the molecular basis of cocaine addiction.
Keywords: Cocaine, Addiction, Transcriptome, Gene Expression, Mesolimbic Reward Circuitry
Disclosure: Nothing to Disclose.

T280. Using Genome-Wide Association and RNA Sequencing in Diversity Outbred Mice to Identify Genes Associated With Ethanol-Related Traits

Clarissa Parker*, Vivek Philip, Lauren Kuffler, Daniel M Gatti, Troy Wilcox, Erica Busch, Sophie Masneuf, Erica Sagalyn, Elissa J Chesler, Andrew Holmes

Middlebury College, Middlebury, Vermont, United States

Background: 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.
Methods: We have taken advantage of the newly developed 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 (LORR). We used the high density MEGAMuga and GIGAMuga arrays to obtain genotypes ranging from 77,808 – 143,259 SNPs. We also measured gene expression using RNA sequencing in two brain regions.
Results: We identified 10 suggestive and significant QTLs associated with ethanol sensitivity related traits on chromosomes 1, 2, 4, 9, 10, 12, and 16. The implicated regions were narrow (500 kb – 4 Mb in size) and contained a number of promising quantitative trait genes based on their reported functions. We are currently integrating the behavioral QTL and eQTL results to prioritize among the candidate genes associated with ethanol sensitivity.
Conclusions: The high genetic precision and phenotypic diversity in the DO have facilitated the discovery of previously undetectable mechanisms underlying predisposition to develop AUDs. With the inclusion of RNA-Seq and other molecular profiling we will be 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.
Keywords: Genome-Wide Association Studies, Expression Quantitative Trait Loci, Alcohol Sensitivity
Disclosure: Nothing to Disclose.

T281. Low Translocator Protein Expression in Alcoholism: a [11C]PBR-28 PET Study in Humans and Rats

Corinde Wiers*, Sunny Kim, Ehsan Shokri Kojori, Ryan Tyler, Luke Park, Amna Zehra, Clara Freeman, Veronica Ramirez, Victor Pike, Robert Innis, Marisela Morales, Leandro Vendruscolo, George Koob, Dardo Tomasi, Gene-Jack Wang, Nora Volkow

National Institute on Alcohol Abuse and Alcoholism, Bethesda, Maryland, United States

Background: The biological mechanisms of alcohol-induced neurotoxicity have been poorly understood and may involve neuroinflammation.
Methods: Here we investigated 18-kDa translocator protein (TSPO) expression, a marker of microglial activation and inflammation, using [11C]PBR28 and Positron Emission Tomography (PET) in vivo in 36 human participants: $n = 19$ patients with alcohol use disorder (AUD) within a week after their last drink, and $n = 17$ healthy controls (HC). Complementary, [11C]PBR28 PET scans were performed in $n = 20$ rats: $n = 10$ chronically alcohol-vaping rats and $n = 10$

alcohol-naïve control rats. Rat brains were harvested after each scan for in vitro autoradiography using [3H]PK-11195. Since TSPO gene insertion has recently been shown to play a pivotal role in cell metabolism and energy production, we further associated TSPO genotype rs6971 and [11C]PBR28 binding with brain glucose metabolism in all human participants using PET and [18F]-FDG.
Results: AUD patients showed lower [11C]PBR28 binding compared to controls; in medium binders only. In line with these findings, rats chronically exposed to alcohol also showed lower [11C]PBR28 uptake in vivo, and lower [3H] PK-11195 binding in vitro. AUD patients also showed lower brain glucose metabolism compared to controls; which is consistent with previous reports. There were no associations between rs6971 and brain glucose metabolism in AUD patients or healthy controls. However, a pixel-wise Biological Parametric Mapping model showed significant positive correlations between brain glucose metabolism and [11C] PBR28 in the bilateral calcarine in AUD patients and healthy controls pooled together.
Conclusions: These and previous human PET findings suggest lower activation of microglia in the brain in alcoholics. Preliminary immunohistochemistry findings in post-mortem brains of the alcohol-dependent rats indicate astrocytosis but not increased TSPO expression in alcohol-dependent versus control rats; suggesting that future studies may consider using tracers binding to neuroinflammation targets other than TSPO. Our finding that brain glucose metabolism and [11C]PBR28 are positively correlated in the calcarine confirms a role of TSPO in energy metabolism.
Keywords: Neuroinflammation, Alcoholism, Brain Glucose Metabolism
Disclosure: Nothing to Disclose.

T282. Stimulation of NMDA Receptors via D-Serine Augmentation Impairs Cocaine Reinstatement

Kati L. Healey, Marian T. Sepulveda-Orengo, Ben Wu, Kathryn Reissner*

University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, United States

Background: Our lab has previously shown that cocaine self-administration and extinction leads to adaptations in astrocytes from the nucleus accumbens (NAc) core, including decreased surface area, and synaptic colocalization (Scofield et al, 2016). Because astrocytes are an important source of the NMDA co-agonist D-serine, we hypothesized that the reduction in synaptic colocalization would lead to impaired astrocyte modulation of NMDA receptor function, and that restored function might normalize synaptic processing and consequently reduce cocaine seeking. Separate reports in the literature have indicated that pharmacological antagonism of NAc NMDA receptors can promote cocaine reinstatement. Accordingly, we hypothesized that stimulation of NMDA receptors with D-serine would oppose reinstatement, independently of any effect of D-serine on extinction learning.
Methods: We employed the rat self-administration and reinstatement model of cocaine relapse. D-serine levels were stimulated by administration (both systemic and intra-NAc) of D-serine as well as the D-amino acid oxidase inhibitor sodium benzoate, together. In addition to behavioral reinstatement experiments, we utilized whole cell patch-clamp recordings to measure the stimulus-response relationship of NMDA receptor-mediated currents, in medium spiny neurons (MSNs) from cocaine versus saline extinguished rats treated with either D-serine or saline vehicle. We also assessed the contribution of specific NMDA receptor subunits to the effect of D-serine stimulation NMDA receptor currents using subunit specific antagonists.
Results: Three days of administration of D-serine and sodium benzoate dose-dependently impaired cocaine plus cue-induced reinstatement. This result was observed in cocaine-extinguished rats, as well as cocaine-abstinent rats, indicating that the effect was not dependent on extinction learning. No effect was observed on food reinstatement or locomotor behavior. Moreover, intra-accumbens administration of D-serine paired with subthreshold, low-dose administration of NMDA also significantly impaired cocaine reinstatement. The systemic regiment of D-serine and sodium benzoate significantly elevated levels of NAc D-serine, and significantly increased the amplitude of NMDA receptor currents in NAc core MSNs. In addition, ifenprodil application reduced NMDA receptor currents most effectively in MSNs from cocaine-experienced rats treated with D-serine augmentation, suggesting that enhancement of NMDA receptor function by D-serine augmentation occurs at least in part through increased contributions of NR2B receptor subunits. Ongoing experiments employ cell surface biotinylation of NAc tissue to assess the effects of D-serine augmentation on surface levels of NMDA as well as AMPA receptors subunits.
Conclusions: These findings collectively indicate that D-serine augmentation during cocaine withdrawal impairs subsequent reinstatement behavior, with no effect on locomotor behavior or food seeking. These results are observed following either systemic or direct intra-accumbal stimulation of NMDA receptors with D-serine. These finding support previous observations in the literature that antagonism of NAc core NMDA receptors may induce or enhance cocaine reinstatement, and suggest the hypothesis that NAc NMDA receptors may exert an inhibitory influence over cocaine seeking. Future studies will be required to determine the precise mechanism by which this may occur.
Keywords: Cocaine, NMDA Glutamate Receptors, Nucleus Accumbens
Disclosure: Nothing to Disclose.