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Panel
1. The Role of Placenta Biology in Risk for Psychiatric Illness

1.1 The Wonders of the Human Placenta: An Introduction

Susan Fisher

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Background: The human placenta is a singular organ. Its basic structure is established by the third week of pregnancy, maturing quickly so that it can support the development and growth of the embryo/fetus. Yet the placenta lives a mere nine months. At birth it shows the same signs of aging as an elderly individual.

Methods: We study human placental development, in normal pregnancy and in a variety of pregnancy complications, by using numerous approaches—transcriptomic, genomic and epigenomic technologies.

Results: The placenta's basic building blocks are termed chorionic villi. Their formation is linked to differentiation of specialized progenitors, termed cytotrophoblasts (CTBs). In one pathway, CTBs detach from the trophoblast basement membrane that surrounds the mesenchymal cores of chorionic villi and fuse to form a continuous layer of syncytiotrophoblasts (STBs) that covers the villus surface. These floating villi, so named because they are bathed in maternal blood, are the site of hormone production and the exchange of myriad substances between the mother and the embryo/fetus. In the other pathway, CTBs in anchoring chorionic villi, so named because they anchor the placenta to the uterus, form columns of cells that attach to and then penetrate the uterine wall where they give rise to invasive CTBs. During interstitial invasion, a subset of these cells commingles with resident decidual, myometrial and immune cells. During endovascular invasion, CTBs migrate into the vessels and plug the lumina, which eventually recanalize. Thus, CTB invasion anchors the placenta to the uterus and diverts maternal blood to the intervillous space. Defects in formation and/or function of the latter CTB population are associated with the major complications of pregnancy, including preeclampsia and some cases of preterm birth.

CTB transformation of the uterine vasculature is a singular process, without direct parallels in biology or pathology. In the columns, they begin the process of vascular mimicry in which they downregulate many epithelial-like molecules that are indicative of their ectodermal origin and upregulate numerous receptors/ligands that are typically expressed by endothelial or vascular smooth muscle cells. The result of this dramatic transformation is a placental cell with the molecular code of a vascular cell. Likely, it is this switch that enables their extensive interactions with uterine blood vessels.

In most cases, the placenta supports the normal growth of a baby. However, numerous pregnancy complications are associated with faulty placentation and poor fetal growth, which we now think has life-long consequences. The idea that conditions in the womb can affect later life is termed the fetal origins hypothesis. It is based on retrospective studies correlating the nutritional status of the fetus, as indicated by birthweight, with the risk of developing cardiovascular disease later in life. Originally, the hypothesis was put forth to explain the high incidence of cardiovascular disease in poor areas of England, where many women gave birth to underweight infants who died and survivors went on to develop these disorders. Some investigators think that poor nutrition and placental function may leave their mark at the level of the epigenome.

Conclusions: The placenta is an amazing but transient organ. Despite its short lifespan, the placenta's impact is life-long in terms of programming adult health.

Disclosure: Nothing to Disclose.

1.2 Placental H3K27me3 Promotes Female Resilience to Prenatal Stress

Tracy Bale

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Background: Prenatal stress is a risk factor for male-biased neurodevelopmental disorders, including early onset schizophrenia and autism. In our mouse model of early prenatal stress (EPS), stress exposure during the first week of

gestation imparts long-term HPA stress axis, metabolic and cognitive deficits to male offspring. The placenta, a fetally-derived tissue reflecting fetal sex chromosome complement, provides necessary factors for early brain development. Thus, sex differences in placental function might radically influence sex biases in neurodevelopmental vulnerability to prenatal insults. We previously identified the X-linked, stress sensitive, nutrient sensor O-linked-N-acetylglucosamine (OGT) as a critical mediator of the sex-specific effects of prenatal stress on offspring brain development. OGT modifies several epigenetic regulators including the H3K27me2/3 methyltransferase, EZH2.

Methods: To evaluate the novel roles of the X and Y -linked H3K27 demethylases in establishing sex differences in H3K27me3, and to confirm the role of H3K27me3 in male risk and female resilience to the neurodevelopmental deficits associated with EPS exposure, we generated trophoblast-specific mouse lines with reducible UTX and inducible UTY. We predict that reducing UTX in male trophoblasts will augment placental H3K27me3 and protect males from sensitivity to prenatal insults. In addition, we predict that reducing UTX while inducing UTY expression in female trophoblasts will masculinize genome-wide H3K27me3 patterns and masculinize placental responses to environmental perturbations.

Results: In mouse placentas with trophoblast-specific OGT reduction, we found that OGT determines higher protein levels of H3K27me3 in females and genome-wide sex differences in H3K27me3 patterns. We hypothesized that female-biased epigenetic repression (i.e. H3K27me3) protects females from prenatal insults such as EPS. To test this hypothesis, we reduced H3K27me3 in female placentas using trophoblast-specific manipulations of EZH2 and exposed these females to maternal stress. Decreasing placental EZH2, and H3K27me3, produced female vulnerability to the effects of stress, sensitizing their HPA axis reactivity and causing long-term changes in body weight. In addition, our production of XX female mice over-expressing placental Uty showed a significant reduction in H3K27me3, providing evidence for Uty role in demethylase activity in trophoblast cells of the placenta that may contribute to the overall low levels of this histone mark in male tissue.

Conclusions: These studies reveal profound sex-differences in mechanisms of transcriptional regulation in the placenta that directly contribute to increased risk for the developing male brain to maternal insults across gestation. We propose that placental H3K27me3 provides females with an overall resilience to the effects of prenatal disturbance, including maternal stress, by providing a broad transcriptional regulation that diminishes the effects of an altered intrauterine environment.

Disclosure: Nothing to Disclose.

1.3 Epigenetic Biomarkers at Birth Using Placenta Samples From a Prospective High-Risk Autism Study

Janine LaSalle

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Background: Placenta is a tissue normally discarded at birth but of potential use for identification of biomarkers of disease risk at birth. Designed to identify etiologies and early markers of autism spectrum disorders (ASD) and other neurodevelopmental disorders (NDD), the prospective MARBLES (Markers of Autism Risk in Babies: Learning Early Signs) study recruits mothers of at least one child diagnosed with ASD who are either planning or are pregnant with another child. Given the family history of ASD, along with shared environmental and genetic factors, the sibling is at higher risk for developing ASD and other NDD than children from the general population. Observed rates are 1 in 5 (22%) affected with ASD, with significant differences in risk between males (27%) versus females (10%), and another 23% with other aberrant NDD outcomes (OND) including attention problems or language delays or broader autism phenotype, and about 55% with typical development (TD).

Methods: To identify novel regions of differential methylation whole genome bisulfite sequencing (WGBS) was performed on DNA isolated from male placentas from MARBLES (20 ASD and 21 TD), and differentially methylated regions (DMR) identified using the DMR finder approach modified from the bsseq R package. To test potential effect modifiers, samples were stratified by genetic and environmental covariates.

Results: Two high-confidence DMR passed family-wise error rate (FWER) permutation testing and were also confirmed as significantly different between ASD and TD by the independent method of pyrosequencing. Percent methylation of both DMR significantly correlated with ASD severity and composite Mullen cognitive scores, in the directions predicted from ASD diagnosis. ASD DMR specifically correlated with ASD severity and Mullen scores, with parity and overweight/obese as the only significant covariates. A second cohort of 39 placenta samples are currently undergoing WGBS and analyses as a replication set.

Conclusions: Together, these results suggest the potential promise of placenta as a source of epigenetic biomarkers that may be multiplexed to predict ASD risk from birth, allowing for early behavioral intervention and improved outcomes.

Disclosure: Nothing to Disclose.

1.4 Placental Gene Expression, Obstetrical History and Polygenic Risk for Schizophrenia

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Background: Early life events influence later susceptibility to many adult diseases and may contribute to define the environmental context in which genes enhance risk for complex disorder like schizophrenia. Here we analyze the role of intrauterine and perinatal environment in modulating the association of schizophrenia with genomic risk.

Methods: We evaluated whether genomic risk for schizophrenia interacts with intrauterine and perinatal complications (Early Life Complications, ELCs) on case-control status, in three independent samples of healthy subjects and patients with schizophrenia from USA ($n = 501$), Italy ($n = 273$) and Germany ($n = 919$). We further analyzed the relationship between genomic risk and ELCs in two samples

of only patients with schizophrenia from Germany ($n = 1020$) and Japan ($n = 172$). Genomic risk was measured with polygenic risk profile scores based on GWAS-significant alleles (PRS), while ELCs history was assessed with the McNeil-Sjöström Scale. We tested whether genes overlapping the schizophrenia loci interacting with ELCs are enriched in placenta and differentially expressed in placental samples from complicated pregnancies, in 8 independent placental datasets. Finally, we evaluated whether GWAS SNPs marking loci containing genes highly expressed and dynamically modulated in placenta (PlacPRS genes) drive the interaction between PRS and ELCs, and performed pathway analyses on PlacPRS genes.

Results: PRS interacts with ELCs on case-control status, in the three independent samples from USA ($p = \ p = 0.004$), Italy ($p = 0.015$) and Germany ($p = 0.018$); in each sample the variance of schizophrenia explained by PRS is multiplicatively higher in the presence of a history of ELCs compared with the absence of such events. The relationship between genomic risk and ELCs is further replicated in the two independent samples of only cases from Germany ($p = 0.044$) and Japan ($p = 0.047$). The gene-set based on PRS loci interacting with ELCs is highly expressed in multiple placental tissues ($p < 0.001$) and dynamically regulated in placental samples from complicated, in comparison with normal, pregnancies ($p < 0.05$). These differences are significantly greater in placentae from male compared with female offspring ($p < 10e\text{-}8$). The interaction between PRS and ELCs is largely driven by PlacPRS genes ($p = 0.002$); PRS constructed from the remaining loci do not interact with ELCs (NonPlacPRS, $p = 0.60$). Pathways and biological functions associated with NonPlacPRS genes are reminiscent of previous analyses about schizophrenia risk-genes, while PlacPRS genes implicate an orthogonal biology, with roots in the fetal/placental response to stress.

Conclusions: Our data suggest that the most significant schizophrenia GWAS variants contribute to risk at least partly by converging on a developmental trajectory sensitive to ELCs and altered placental gene expression. The sex-associated effects on placental transcription suggest that the male preponderance of schizophrenia may arise from gene-environment interactions that influence placental biology. These results highlight placental health as a new public health frontier for primary prevention, particularly in high-risk males.

Disclosure: Nothing to Disclose.

Panel
2. Translating Advances in Fear Extinction Research From the Lab to the Clinic

2.1 Preventing the Return of Fear Using Reconsolidation Updating and Methylene Blue is Differentially Dependent on Extinction Learning

Marie Monfils

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Background: Many factors account for how well individuals extinguish conditioned fears, such as genetic variability, learning capacity and conditions under which extinction training is administered. We predicted that memory-based interventions would be more effective to reduce the return of fear (reinstatement) in subjects genetically predisposed to display more extinction learning.

Methods: We tested this hypothesis in rats genetically selected for differences in fear extinction using two strategies: (1) attenuation of fear memory using post-retrieval extinction training, and (2) pharmacological enhancement of the extinction memory after extinction training by low-dose USP methylene blue (MB). Subjects selectively bred for divergent extinction phenotypes were fear conditioned to a tone stimulus and administered either standard extinction training or retrieval + extinction. Following extinction, subjects received injections of saline or MB.

Results: Both reconsolidation updating and MB administration showed beneficial effects in preventing fear reinstatement, but differed in the groups they targeted. Welch's t-tests revealed that the differences between the MB and saline groups for individuals with poor extinction (Unsuccessful) was not significant, $t(41.5) = -0.34$, $p = 0.73$, but the difference for the Successful extinguishers ($n = 38$) was, $t(30.1) = 2.18$, $p = 0.03$. There was no evidence for an extinction X training interaction, $F(1,78) = 0.29$, $p = 0.59$. There was, however, a significant independent effect of retrieval+extinction, $F(1,79) = 5.6$, $p = 0.02$, which resulted in a reduction in fear reinstatement of 0.5 standard deviations, 95% Confidence Interval [0.08, 0.92]. In sum, reconsolidation updating showed an overall effect in reducing fear reinstatement, whereas pharmacological memory enhancement using MB was an effective strategy, but only for individuals who were responsive to extinction.

Conclusions: Reconsolidation updating (using retrieval-extinction) provides a potentially useful avenue for treating anxiety-related disorders, but more work needs to be conducted to establish the optimal parameters of administration and improve our understanding of boundary conditions that limit or prevent its efficacy. The present work is an important step in identifying which individual phenotypes might best benefit from available treatment avenues. We next propose a potential approach to determine, prior to treatment, characteristics of individuals that would respond best to one treatment avenue over another.

Disclosure: Nothing to Disclose.

2.2 Activity in the Ventromedial PFC, Striatum, and Hippocampus is Associated With Decreases in Long-Term Following Augmented Extinction

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Background: Research on overcoming unwanted behavior relies extensively on the principles of extinction—in which omission of expected events diminishes learned behavior. However, as a technique to permanently reduce fear behaviors, extinction is unsatisfactory as extinguished behaviors return under a variety of circumstances. Here, we used functional MRI to investigate a novel form of augmented extinction in which aversive outcomes are replaced by novel non-aversive outcomes. We have previously shown this technique reduces the return of fear

behaviors in humans and rats. This procedure may optimize the opportunity for new learning by maximizing surprise during extinction training, which, according to associative learning models, is critical to strengthening the effects of new learning.
Methods: The design was a between-groups (N = 46) functional MRI Pavlovian fear conditioning and extinction investigation. During conditioning, one conditioned stimulus (CS+) was paired with a shock to the wrist and another cue (CS-) was unpaired. Subjects either underwent standard extinction, or an augmented form of extinction in which an expected electrical shock outcome was replaced–rather than merely omitted–by a novel and surprising tone. Subjects were then scanned 24-hours later during a test of spontaneous recovery in the absence of the tone or the shock.
Results: Behaviorally, arousal (skin conductance responses) to the CS+ versus CS- were reduced in the 'novelty-facilitated extinction' compared to standard extinction group (P = .02). Skin conductance responses were fit to a computational model of associability, which provided new evidence that augmented extinction speeds the acquisition of new learning. Whole-brain functional MRI results from Day 2 recovery tests showed enhanced activity in the novelty-facilitated extinction group compared to the standard extinction group in the dorsal striatum, ventromedial prefrontal cortex, and parahippocampal complex (P < .001). These regions may constitute an inhibitory circuit for controlling the expression of threat-related behavior to cues that no longer signal threat.
Conclusions: These findings provide neurobehavioral evidence for a novel non-pharmacological behavioral strategies for enhancing fear extinction that can be straightforwardly adapted and implemented in clinical situations. We discuss ongoing work examining the crossover between novel approaches to optimize extinction and reducing the emotional enhancement for negative memories.
Disclosure: Nothing to Disclose.

2.3 Unpredictable Threat Modulates Amygdala-Prefrontal Functional Connections in Posttraumatic Stress Disorder

Rajendra Morey

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Background: Goal-directed processing in the context of unpredictable environmental threats may be impaired in people with posttraumatic stress disorder (PTSD). The impairment is hypothesized as dysregulated functional connectivity between the amygdala and ventromedial prefrontal cortex (vmPFC). Here, we tested this hypothesis in PTSD patients actively engaged in goal directed task goal directed tasks that involve balancing competing demands and priorities while exposed to unpredictable threats.
Methods: Veterans exposed to trauma ($n = 50$) were grouped based on evaluation with the Clinician Administered PTSD Scale (CAPS) into PTSD ($n = 25$) or trauma-exposed controls ($n = 25$). The participants underwent function MRI while performing a computer-gaming style task to optimize monetary rewards by capturing prey and avoiding capture by predators during exposure to unpredictable threat posed by randomly presented electrical shocks.
Results: The PTSD group versus controls showed smaller amygdala activation ($p < 0.05$, SVC; left peak at [-20, 0, -12]; right peak at [20,4,–16]) and weaker left amygdala-ventromedial PFC (vmPFC) functional connectivity to the threat vs. nonthreat contrast. The rate of participant capture by the predator (impaired goal directed performance under threat) was negatively correlated with vmPFC activation (R = -0.545, $p = 0.011$), and positively correlated with vmPFC grey matter volumes in PTSD patients (R = 0.571, $p = 0.007$). The rate of prey captures by the participant (successful goal directed performance under threat) was positively correlated with vmPFC activation (R = 0.449, $p = 0.041$) and negatively correlated with left amygdala-vmPFC functional connectivity (R = -0.041, $p < 0.05$ SVC) in trauma-exposed controls.
Conclusions: Exposure to unpredictable threat in PTSD is associated with diminished amygdala response and amygdala-vmPFC functional connectivity, which suggests dysregulated neural responses to threat and may explain impaired performance of PTSD patients during goal-direct activity. Furthermore, trauma survivors without PTSD perform better on goal-directed activity and are more reliant on vmPFC function than on amygdala-vmPFC connectivity, which may reflect post-trauma adaptation that confers resilience to PTSD.
Disclosure: Nothing to Disclose.

2.4 Facilitating Fear Extinction in Humans: Psychophysiology of Fear in the Clinic

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Background: Individuals who suffer from fear-related disorders, such as posttraumatic stress disorder (PTSD), have a deficient ability to extinguish psychophysiological measures of fear, such as fear-potentiated startle (Norrholm et al 2011) or skin conductance response (Milad et al 2008). Fear extinction involves a conditioning session in which a cue that was previously associated with danger is presented repeatedly without the aversive outcome, and can serve as a model for exposure-based therapy. Understanding the neurobiological bases of extinction is of great importance to optimizing treatment. Both pharmacological and non-pharmacological methods for facilitating fear extinction have been tested; for example, d-cycloserine, a partial NMDA receptor agonist, has been shown to decrease startle responses with exposure therapy (Rothbaum et al 2014). A potential non-pharmacological facilitator is reactivating the fear memory prior to extinction (Monfils et al 2009; Warren et al 2015). In the current study, participants with fear of flying were randomized to one of two conditions, where they either received a fear-eliciting cue prior to virtual reality exposure therapy (VRE) or a neutral cue prior to VRE. We hypothesized those patients who were presented with reactivating, fear-related cues prior to exposure therapy would demonstrate reduced fear post-treatment as compared to patients who were presented with a neutral cue prior to exposure therapy.
Methods: The study participants were 89 individuals who met DSM-IV criteria for specific phobia, situational type or

panic disorder with agoraphobia, in which flying was reported to be the primary feared stimulus. During VRE, patients wore an eMagin Z800 head-mounted display which provided audiovisual cues and a computer-generated view consistent with being inside an airport and passenger compartment of a plane that changed in a natural way with head and body motion. This treatment has been previously supported as an effective treatment for fear of flying (Rothbaum et al 2006). Ten minutes prior to exposure therapy, participants received either a reactivation cue (15 second VR clip of flying) or a neutral cue (15 second VR clip of library). Participants completed the Fear of Flying Inventory self-report measures (Scott, 1987). The psychophysiological data were collected using Biopac MP150. We recorded skin conductance (SC) and heart rate (HR) activity as in previous reports (Rothbaum et al 2014).
Results: Self-reported symptoms of fear decreased with treatment ($p<0.001$), but there was no interaction with condition. However, SC levels revealed a significant effect of condition at post-treatment, $F(1,46)=15.40$, $p=0.0003$, as well as at 3 months, $F(1,42)=5.25$, $p=0.03$. The group that received the reactivation cue prior to VRE showed smaller SC response to the VR stimuli compared to the group that received the neutral cue.
Conclusions: These results suggest that memory reactivation prior to exposure therapy did not have an impact on clinical measures but may enhance the effect of exposure therapy on a physiological level. Taken together with our other studies, these data suggest that physiological measures of fear can be reduced by facilitating extinction processes in exposure therapy.
Disclosure: Nothing to Disclose.

Panel
3. Ketamine for Depression and Suicidal Thoughts: Exploring Dose and Potential Mechanisms

3.1 Neurobiological Markers of Suicide Ideation Response to Ketamine: Activity and Neuroimaging

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Background: Rapid-acting interventions, such as ketamine, suggests both exciting clinical promise as well as new avenues for research into the neurobiology of suicide. For this symposium, new findings related to potential biomarkers of suicide ideation response to ketamine will be presented across two modalities: actigraphy and neuroimaging. Previous results have demonstrated that nocturnal wakefulness, as defined by sleep EEG, is associated with suicidal thoughts and antisuicidal response to ketamine, but less is known about the relationship between suicidal thoughts and activity markers as assessed by wrist actigraphy. Additionally, objective biomarkers of implicit suicide risk such as the Suicide Implicit Association Task (S-IAT) may illuminate potential mechanisms of suicide risk as well as ketamine's antisuicidal response.
Methods: In the analysis of actigraphy data, 24-hour wrist actigraphy was correlated with next-day suicidal thoughts in a sample of 50 patients with treatment-resistant MDD or BP. Additionally, change in suicide ideation was compared with change in activity patterns after ketamine administration. As for the analysis of the S-IAT, an fMRI version of the S-IAT was piloted in 27 healthy volunteers (3T GE HDx scanner, resolution 3.5mm3). In particular, the contrast between the self-death block and self-life block was evaluated using a t-test.
Results: Contrary to expectations for the actigraphy results in patients, activity was lower in the suicidal patients as compared to the non-suicidal patients, particularly over the course of the night ($p<.05$). Additionally, in the patients with suicidal thoughts at baseline, reductions in nighttime activity was associated with antisuicidal response to ketamine. The S-IAT results in the healthy controls indicated increased activation in the insula in the self-death condition as compared to the self-life condition ($p<.05$, corrected for multiple comparisons). These task-related findings will be discussed as they relate to recent functional connectivity analyses of MDD patients, highlighting that ketamine may increase insula functional connectivity within the default mode network, potentially normalizing connectivity to that seen in healthy controls.
Conclusions: Implications of the findings both for understanding potential mechanisms of ketamine's impact on suicidal thoughts as well as elucidating the neurobiology of suicidal thoughts will be reviewed. Further research is needed to evaluate whether the wrist-activity markers or the fMRI S-IAT could serve as biomarkers of acute suicide risk or antisuicidal response to ketamine.
Disclosure: Nothing to Disclose.

3.2 A Randomized, Double Blind, Placebo Controlled Trial of Repeat-Dose Ketamine Augmentation for Chronic Suicidal Thinking

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Background: Suicide is the 10th leading cause of death in the Unites States, and unlike other top 10 causes of death (such as heart disease, diabetes, and cancer), suicide rates are on the rise. This highlights the urgent need for rapidly-acting, effective antisuicidal agents. Towards this end, several small single- and repeat-infusion studies indicate that subanesthetic doses of ketamine rapidly reduces thoughts of suicide in patients with suicidal thinking. However, the extent to which repeated doses of ketamine (versus placebo) reduces thoughts of suicide in the short- and long-term in depressed outpatients with current suicidal ideation remains unknown.
Methods: Twenty-six outpatients with severe treatment resistant major depressive disorder and current, chronic ($\geq$3 months) suicidal ideation were enrolled and randomized in a double-blind fashion to six infusions over three weeks of ketamine (0.5 mg/kg over 45 minutes) or saline placebo ("infusion phase"). Measures of suicidal ideation (Hamilton Depression Rating Scale (HDRS) Suicide Item (SI); Columbia Suicide Severity Rating Scale (C-SSRS); Concise Health Risk Tracking (CHRT); and the Death/Suicide Implicit Association Test (IAT D-score)) were assessed at the baseline

visits, 240 minutes post-infusion, and for three months thereafter in a naturalistic "follow-up" phase. Suicide remission was defined as a score of zero on the C-SSRS at endpoint. Measures of depression were assessed with the Hamilton Depression Rating Scale (HDRS); response was defined as $\geq 50\%$ decrease in score from baseline to endpoint, and remission was defined as ≤ 7 total score at endpoint.
Results: During the infusion phase, there was no significant difference between placebo and ketamine on measures of suicidal thinking (C-SSRS Ideation or Intensity scores, CHRT Propensity or Risk scores, or IAT D-scores) or depression severity (HDRS total; all $p > 0.05$). At the beginning of the naturalistic follow-up phase, four patients in the ketamine group met criteria for remission, but only one of them maintained remission at the end of the follow-up period. Three patients in the placebo group met remission criteria at the same time point; two remained in remission at the end of the follow-up phase.
Conclusions: Overall, repeated, non-escalating doses of subanesthetic ketamine did not outperform placebo in terms of antidepressant or antisuicidal effects in this double-blind, placebo controlled study of patients with severe treatment-resistant depression and suicidal thinking. This result could potentially support our previously published open-label data that in this severely-ill population, the commonly used subanesthetic ketamine dose of 0.5 mg/kg over 40 minutes is not efficacious in improving depression and suicidal ideation.
Disclosure: Part 1: Janssen Pharmaceuticals, Employee, **Part 2:** Janssen Pharmaceuticals, Employee, **Part 3:** Janssen Pharmaceuticals, Employee, **Part 5:** Janssen Pharmaceuticals, Employee.

3.3 A Randomized, Double-Blind, Dose Finding Study of Ketamine's Antidepressant Effect in Major Depressive Disorder

Matthew S. Milak

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Background: Major depressive disorder (MDD) is a highly prevalent illness, affecting over 15 million American adults annually. It was the fourth leading cause of disability globally in 2002, as assessed by disease-adjusted-life years according to the World Health Organization. MDD was projected to become the second leading cause of disability by 2030, however by 2015 it has already become the leading cause of disability worldwide. Compounding the disease burden and the related cost of illness in MDD is the several week delay between initiation of treatment and onset of therapeutic action of currently available treatments. Shortening this delay to clinically significant improvement in antidepressant treatment of MDD is a major unmet challenge. Although numerous clinical trials have shown that a single intravenous sub-anesthetic dose of ketamine (an NMDA receptor antagonist) can bring about full remission in hours, there has only been one dose-finding study ($N = 4$). We hypothesized that administration of ketamine in drug-free MDD will induce a dose-dependent reduction in the 24-item Hamilton Depression Rating Scale (HDRS-24) scores of these patients.
Methods: To evaluate the relationship between the plasma concentration of ketamine and its dehydro-metabolites and clinical improvement, we designed a randomized, placebo-controlled, double-blind study in MDD. Patients were randomized into six groups, receiving saline alone or one of five different doses of ketamine (0.1, 0.2, 0.3, 0.4, or 0.5 mg/kg).
Results: We find that percent improvement on the HDRS correlates with plasma concentration of norketamine measured at 120 minutes post-ketamine infusion (linear model: $F = 4.949$, $p = 0.040$; quadratic model: $F = 4.887$, $p = 0.022$).
Conclusions: Although we expected to find widespread strong correlations between clinical improvement and plasma concentrations of ketamine and its metabolites, these correlations were weak and did not survive correction for multiple comparisons. The only one we had a prior hypothesis about based on our previous study was norketamine, which was significant in the present trial as well. However, we only looked at the dehydro-metabolites of ketamine because at the time the prevailing theory was that ketamine's antidepressant mechanism of action is irrevocably linked to its antagonist activity at the NMDA receptor.
Disclosure: Nothing to Disclose.

3.4 Ketamine for Rapid Relief of Suicidal Thoughts in Depression: Cognition and Biomarkers in a Midazolam-Controlled Trial

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Background: Little is known about pharmacotherapy to relieve suicidal ideation in depression. Studies show reduction in suicidal thoughts after treatment with the NMDA receptor antagonist ketamine, but trial methods have limited conclusions. The mechanism of ketamine's effects on depression and suicidal thoughts is not well-understood, but is thought to involve neuroplasticity processes including upregulation of Brain Derived Neurotrophic Factor (BDNF) and rapid synaptogenesis in pre-frontal cortex and hippocampus.
Methods: This was a single-site, randomized, double-blind, midazolam-controlled trial of sub-anesthetic intravenous ketamine for relief of clinically significant suicidal ideation in major depressive disorder (MDD). Adults ($N = 80$) with current MDD and score ≥ 4 on the Beck Scale for Suicidal Ideation (SSI) were randomized to an infusion of adjunctive ketamine or midazolam. Primary outcome was SSI score 24 hours post-infusion. Participants completed neurocognitive and saliva cortisol awakening response (CAR) tests before and 24 hours post-infusion. A subgroup ($N = 53$) provided blood samples immediately pre- and post-infusion to measures levels of ketamine, its metabolites norketamine and dehydro-norketamine), and plasma and serum BDNF.
Results: Neurocognition: Neurocognition improved more after ketamine than midazolam (improvement of.32 (.45SD) across the battery vs..10 (.49SD) in midazolam; infusion day

by drug interaction, F[1,76] = 4.01, $p = .049$). Effect was most pronounced on specific tests, namely, reaction time (F [1,75] = 4.21, $p = .044$) delayed memory recall (F[1,79] = 5.91, $p = .017$) and Stroop interference (F[1,76] = 3.67, $p = .059$). Better baseline memory on a list learning task was associated with decline in HDRS after ketamine (r = -.39, $p = .016$) but not midazolam (r = +.12, $p = .465$). Patients showed greater improvement in response speed, retention of information, and interference/cognitive control after ketamine. In contrast to other ketamine studies, slower baseline processing speed was not associated with clinical response.

Cortisol Awakening Response: The midazolam group had a non-significant increase in CAR of 13% ($p = 0.359$). The ketamine group had a non-significant decrease in CAR of 12% ($p = 0.287$). These changes were not significantly different from each other ($p = 0.165$). Interaction terms between group and baseline cortisol were also tested for both awakening and 30-minute values, but neither were significant ($p = 0.298$).

Brain Derived Neurotrophic Factor: We compared the difference in Ln(BDNF) level (post- minus pre-infusion) between randomized ketamine and midazolam infusions. Results were non-significant for plasma ($p = 0.418$) and serum ($p = 0.225$).

Non-responders to midazolam received an open ketamine infusion with BDNF reassessed immediately after. Paired t-tests were used to compare Ln(BDNF) pre- and post-open ketamine infusion within subject (plasma N = 22; serum N = 26). There was a non-significant decrease of Ln(BDNF) in serum ($p = 0.12$) and a significant decrease in plasma ($p = 0.001$).

Ketamine, Norketamine, Dehydro-norketamine Levels: Post-infusion plasma levels of ketamine and metabolites were not correlated with post-infusion SSI, HDRS-17 or HDRS-24 scores nor with change in scores on these clinical rating scales from baseline to 24 hours post-infusion. There was a trend-level association of post-infusion dehydro-norketamine with change in HDRS-24 score (N = 26, r = -0.381, $p = 0.055$).

Conclusions: Baseline memory performance was associated with ketamine response. Reaction time, delayed memory recall, and cognitive control improved more after ketamine than midazolam. Memory and cognitive control deficits have been implicated in suicide attempt risk. Results of CAR, plasma and serum BDNF, and plasma ketamine and metabolite analyses were overall non-significant.

Disclosure: Nothing to Disclose.

Panel
4. New Concepts of how Thalamo-Cortical Interactions Regulate Complex Behaviors

4.1 The Mediodorsal Thalamus: An Essential Partner of the Prefrontal Cortex for Cognition

Christoph Kellendonk

Columbia University/New York State Psychiatric Institute, New York, New York, United States

Background: Cognitive deficits including deficits in working memory are observed across many psychiatric conditions. In schizophrenia they are thought to be core symptoms as they are linked to long-term functional outcome. Deficits in working memory have originally been associated with the prefrontal cortex but there is increasing evidence that the prefrontal cortex (PFC) acts in coordination with its main thalamic counterpart, the medio-dorsal thalamus (MD). Consistent with this, brain imaging studies have found thalamo-frontal hypo-connectivity associated with cognitive symptoms in schizophrenia.

Methods: To address causality we used optogenetic inhibition studies and determined whether thalamo-prefrontal projections directly support working memory in the mouse. Inhibition studies were combined with single unit recordings in the prefrontal cortex to identify underlying neuronal mechanisms of working memory.

Results: We found that reciprocal activity between MD and medial PFC supports the maintenance of working memory, whereas top-down inputs from the mPFC-to-MD guides successful choice selection. At the mechanistic level, we identified elevated activity that was critically dependent on MD inputs for its sustained maintenance across the memory delay. Last, increasing MD excitability selectively during the delay enhanced working memory performance.

Conclusions: The MD thalamus is critical for stabilizing cortical representations important for working memory. Boosting its function could be a potential therapeutic strategy for targeting cognitive deficits.

Disclosure: Nothing to Disclose.

4.2 Thalamic Control of Functional Cortical Connectivity

Michael Halassa

New York University, New York, New York, United States

Background: The thalamus is an evolutionarily conserved structure with extensive reciprocal connections to cortical regions. While its role in transmitting sensory signals is well-studied, its broader engagement in cognition is unclear.

Methods: Here, I will discuss our findings in mice combining quantitative behavior, multi-site multi-electrode recordings and causal manipulations targeting a frontal thalamo-cortical circuit (MD-PFC).

Results: I will present evidence that the thalamus regulates functional connectivity within and between cortical regions, determining how a cognitive process is implemented across distributed cortical microcircuits.

Conclusions: By combining our findings with those in the literature more broadly (including humans and monkeys), I will introduce a proto-theoretical framework, where thalamic circuits do not necessarily determine the categorical content of a cognitive process (e.g., sensory details in feature-based attention), but rather provide a route by which task-relevant cortical representations are sustained and coordinated. Additionally, I will discuss how thalamic control of cortical connectivity bridges general arousal to the specific processing of categorical content, providing an intermediate level of cognitive and circuit description that will facilitate mapping neural computations onto thought and behavior.

Disclosure: Nothing to Disclose.

4.3 Thalamo-Cortical Dynamics in Memory and Consciousness

Yuri Saalmann

University of Wisconsin - Madison, Madison, Wisconsin, United States

Background: Different cognitive operations require interactions between different ensembles of neurons across brain networks. However, it is not clear how information is flexibly routed between neuronal ensembles according to cognitive demands. There is growing evidence that the higher-order thalamus plays an important role in regulating information transmission between cortical neurons. Higher-order thalamic areas receive their major driving input from the cortex (rather than the sensory periphery like first-order thalamic areas), forming extensive cortico-thalamo-cortical pathways which strongly influence cortical excitability. I will discuss how the thalamus, particularly the anterior and intralaminar nuclei, contributes to memory processing as well as the neural correlates of consciousness.
Methods: For memory experiments, we simultaneously recorded (spikes and local field potentials, LFPs) from the anterior thalamus (ANT), hippocampus (HIP, specifically the subiculum) and retrosplenial cortex (RSC) of two macaques performing a visuospatial episodic-like memory task. For consciousness experiments, we simultaneously recorded from the frontal eye field (FEF), lateral intraparietal area (LIP), caudate nucleus (CN) and central lateral thalamic nucleus (CL) of two macaques, during general anesthesia (propofol or isoflurane) and wakefulness (resting state or fixation task). We used diffusion MRI to target linear electrode arrays to interconnected network sites (1mm isotropic, 60 diffusion directions, b = 1000 s/mm2 and NEX = 14, using a 3T GE MR750 scanner and 16-channel receive-only head coil), and structural MRI of electrodes in situ to confirm electrode positions (0.5mm isotropic).
Results: In memory experiments, our preliminary electrophysiological data show bidirectional conditional Granger causal influences in the 5-15 Hz range between the HIP and RSC during memory retrieval. During this time, neurons in the ANT (ventral division) responded preferentially to particular learnt visual scenes. We also measured increased coherence between thalamic spikes and the LFPs in both the HIP and RSC, in the 5-15 Hz range. Further, the ANT showed increased conditional Granger causal influences on the HIP and RSC in the same frequency range. In consciousness experiments, preliminary results suggest that CL stimulation counteracted the effects of propofol and isoflurane. Stimulating CL at 50 Hz during anesthesia increased behavioral measures of arousal (including eye opening and purposeful movements), reduced EEG power at low frequencies and increased EEG power at higher frequencies, as well as enhanced responsiveness to auditory oddball stimuli. These behavioral and neural changes started soon after, and were maintained during, CL stimulation. Stimulation was primarily effective in the CL, and not nearby thalamic nuclei. Further, current source density analyses suggest that anesthesia differentially influenced feedforward and feedback processing across fronto-parietal cortex.
Conclusions: These preliminary data suggest that the ANT regulates information transmission between the HIP and RSC based on episodic memory demands. In comparison, the CL regulates information processing in fronto-parietal cortex and the level of consciousness. Overall, the thalamus appears to regulate the gain of cortical neurons and their synchrony, thereby influencing information flow through brain networks.
Disclosure: Nothing to Disclose.

4.4 Spatially Organized Slow Thalamocortical Dynamics at the Transition Into Sleep Detected Through Accelerated Neuroimaging

Laura Lewis

Harvard Society of Fellows, Cambridge, Massachusetts, United States

Background: Many psychiatric disorders, including schizophrenia and bipolar disorder, are associated with deficits in specific electroencephalography (EEG) signatures of sleep. These EEG signatures are generated by oscillatory thalamic and cortical interactions, but investigating the underlying circuit mechanisms in the human brain has been challenging due to the limited methods available for noninvasive imaging. We have previously shown through optogenetic studies in mice that spatially organized inhibition in the thalamus can generate local cortical slow waves characteristic of sleep. Here we investigated whether recent technical advances in noninvasive neuroimaging could be used to localize slow waves in thalamus and cortex in the sleeping human brain.
Methods: We performed simultaneous EEG and functional magnetic resonance imaging (fMRI) in a 3 Tesla scanner (sleep study), and fMRI alone in a 7 Tesla scanner (visual study). We used simultaneous multislice techniques to acquire high temporal resolution fMRI data (TR < 400 ms). In the 7T study we tested the frequency response of the fMRI signal by presenting oscillating visual stimuli at different frequencies. In the 3T study we imaged subjects as they fell asleep inside the scanner, with scans beginning at midnight.
Results: We tested the fMRI response to different stimulus frequencies and found that our method enabled localization of oscillations of up to 0.5 Hz both in visual thalamus and visual cortex. We next tracked slow (0.1-0.5 Hz) oscillations in the resting state and found that thalamic and cortical slow oscillation power in the fMRI signal increased strongly at sleep onset. The phase of the fMRI oscillations corresponded to modulation of EEG power, suggesting they represented fluctuating neurophysiological dynamics.
Conclusions: We conclude that the transition into sleep is marked by increased coherence of thalamocortical slow oscillations, suggesting these dynamics may signal loss of behavioral responsiveness. These results parallel our findings in mice that local cortical slow waves can be induced by thalamic circuits even in the awake animal. In addition, we provide a new neuroimaging approach that can track oscillatory dynamics across thalamus and cortex, which may be broadly applicable for studies of neural dynamics in the human brain.
Disclosure: Nothing to Disclose.

Mini Panel
5. Bipolar Disorder Staging: Cross-Sectional Approaches and a Longitudinal Perspective

5.1 Prediction of Functional Impairment Among Patients With Bipolar Disorder - A Machine Learning Approach

Flavio Kapczinski

McMaster University, Oakville, Canada

Background: Early intervention has been associated to better functional outcomes in bipolar disorder. In this same vein staging models attempts to identifying groups of patients with more pernicious course of illness. Thus, calculators with sufficient sensitivity and specificity are needed to predict which patients will experience more pronounced functional impairment. The use of such calculators in the clinical setting will allow for preventative interventions that can potentially prevent worse outcomes. Machine learning techniques might be helpful in this scenario. The aim of the present study is to build a predictive signature with clinical variables to determine individual risk of functional impairment in patients with bipolar disorder.
Methods: Seventy-eight bipolar patent in euthymia were included. All subjects were over 18 years old. Diagnostic and clinical assessment were carried out using Structured Clinical Interview (SCID) for DSM-IV. Demographic and clinical characteristics were assessed with standardized questionnaire. We applied random forest with recursive feature elimination to predict which patients would have functional impairment as assessed using the Functioning Assessment Short Test (FAST). We used leave-one-out cross validation, up-sampling and down-sampling in order to circumvent cross imbalance. Demographic data, characteristics of the course and the treatments of bipolar disorder, lifetime co morbid psychiatric disorders, lifetime co morbid medical illness and family history of psychiatric disorder were used as variables.
Results: The algorithm differentiated patients with functional impairment from those without functional impairment with an area under the curve (AUC) of 85.8%, sensitivity of 90.9% and specificity of 74.0% in the up-sampling scenario. In the down-sampling scenario, the algorithm differentiated the groups with an AUC of 86.3%, sensitivity of 95.8% and specificity of 70.4%. For both models the most relevant predictor clinical variables in differentiate patients with bipolar disorder with functional impairment from those without functional impairment were age at onset, delay between age at first mood episode and first treatment, presence of rapid cycling and presence of lifetime specific phobias. Other relevant predictors for both models were lifetime hypothyroidism, lifetime generalized anxiety disorder, lifetime cannabis use disorder, a positive family history of bipolar disorder, a positive family history of suicide attempts and a positive family history of psychotic disorders.
Conclusions: We reported a clinical tool using machine learning algorithm with high accuracy to differentiate bipolar disorder patients with functional impairment from those without functional impairment. This is the first study to evaluate the feasibility of using a clinical variables tool developed with advanced machine learning algorithms to predict the risk of functional impairment in bipolar disorder. We believe that future studies with these advanced mathematical techniques with greater samples sizes, in longitudinal samples and in combinations with other variables such peripheral biomarkers could offer more accurate diagnostic and predictive tools.
Disclosure: Nothing to Disclose.

5.2 What are the Core Treatment Targets in Youth in the Earliest Stages of Bipolar Disorders

Jan Scott

Newcastle University, Newcastle, United Kingdom

Background: Classification systems used for mental disorders (e.g. ICD, DSM) have great utility for describing established illnesses in older adults. However, current systems are less applicable to recent onset disorders in young adults as they lack adequate predictive validity, undermining their research as well as clinical utility. General medicine uses a different approach for chronic disorders (e.g. IHD, cancer), employing 'staging models' which avoid dependence on cross-sectional assessment of symptoms and seek to identify where an individual lies on a 'continuum' from health to illness –identifying clinic-pathological boundaries between stages and prescribing stage specific treatments. We review therapeutic options and treatment targets and test prospectively whether these targets are important to transition and disease progression.
Methods: Systematic review of current treatment approaches for early stage bipolar disorders and an examination of more appropriate age- and stage-specific targets. One year, prospective study of >200 help-seeking youth with sub-threshold clinical symptoms who were assessed for sleep-wake cycle, cognitive emotional regulation and mood abnormalities at baseline. Outcome was transition to a threshold (stage 2) disorder.
Results: The systematic review suggested that although 28 studies were identified for early stage interventions for bipolar disorders, the quality of evidence is modest for efficacy or prevention. In contrast, significant evidence suggests that cognitive-emotional regulation (CER) processes (such as rumination) and sleep-wake cycle markers (such as delayed sleep phase) may be ideal targets as they appear to be putative markers of risk of transition to bipolar disorders in adolescents and can help explain the co-occurrence of mental and physical comorbidities. Prospective study demonstrated that an 'anergia' factor derived from the sleep-wake cycle measures, an atypical mood profile and ruminations about exhaustion represented a dimension that predicted transition and is amenable to change with interventions targeted at circadian and cognitive-emotional regulation.
Conclusions: Clinical staging models can offer a useful framework to better understand evolving phenotypes that can inform research on underlying pathophysiology and critically help to identify bipolar-specific and trans-diagnostic targets for future treatment interventions.
Disclosure: Nothing to Disclose.

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5.3 Stage of Illness in Bipolar Disorder: Relation to fMRI Findings

Thilo Deckersbach

Massachusetts General Hospital, Charlestown, Massachusetts, United States

Background: Bipolar disorder is characterized by recurrent manic and most often depressive episodes that interfere with functioning. Traditionally, the course of bipolar disorder has been viewed as episodic with symptomatic and functional recovery in between mood episodes. This view has been challenged by clinical and epidemiological studies that document a chronic, progressive, and often disabling course of bipolar disorder. The concept of staging assumes that bipolar illness advances in a temporal progression including periods with prodromal symptoms, first clinical manifestations to the development of recurrent, more severe and frequent mood episodes. Functional Magnetic Resonance imaging (fMRI) has been used to characterize functional abnormalities in the brain of individuals with bipolar disorder. However, by and large, studies have not related their findings to the stage of illness of the disorder. We recently completed series of fMRI studies investigating neural correlates of cognitive and affective impairments in individuals with bipolar disorder. In this talk we will review how the findings relate to the stage of illness.
Methods: Study participants in these fMRI studies were 78 individuals with DSM-IV bipolar disorder, 32 patients with DSM-IV major depression and 75 healthy control participants. They completed an affective Stroop interference task, a temporal discounting task or resting state scans during fMRI scanning on a 3.0-T whole-body scanner (Siemens Trio or Skyra-System). In the Affective Stroop paradigm, participants were shown three-digit numbers (e.g. 100, 020 or 311, 112) superimposed on neutral, negative or positive affectively valenced pictures taken from the International Affective Picture System (IAPS; Lang et al., 2005). The task was to decide which number was different than the two other numbers (non-interference trials: 100 – correct answer = 1; interference trials: 311 – correct answer = 3). In the temporal discounting task, participants decided whether they would like to receive a smaller reward sooner or a larger reward in the more distant future. The paradigm was completed in both a neutral state as well as after a sadness induction. In addition, participants completed resting state blood oxygenation level dependent signal (BOLD) scans. Task-based functional Data were processed using SPM8 software. Functional connectivity (resting state scans) was computed using combination of tools from FSL v5.0.4 (FMRIB, Oxford, UK) and SPM using scripts described in Van Dijk et al. 2010.
Results: For the Affective Stroop task during interference vs. non-interference trials participants with bipolar disorder (relative to controls), were characterized by increased activation in the dorsolateral prefrontal cortex (DLPFC, $p < .001$), inferior parietal lobule ($p < .001$), anterior insula ($p < .001$) as well increased activation in the dorsal anterior cingulate (dACC; $p = .001$). Negative affect was associated with abnormally increased right dACC activation ($p = .001$) and bilateral DLPFC activation ($p = .001$). In the temporal discounting paradigm, bipolar participants showed an increased present bias during the sad compared to the neutral state ($p < .05$); reflecting an increased likelihood to choose smaller rewards sooner rather than larger rewards later. This was associated with increased activation in the DLPFC, anterior insula, and posterior parietal cortex (all $p < .05$). More life-time suicidal ideation correlated positively with activation in the anterior insula and posterior parietal lobe. Finally, Bipolar patients showed significantly weaker right anterior insula functional connectivity with the inferior parietal lobule (IPL) of the executive control network relative to major depression participants and controls.
Conclusions: Results reveal functional abnormalities in brain regions that are largely consistent with previous findings in samples of individuals with bipolar disorder. These brain regions are involved in decision-making and various aspects of emotion regulation. For this panel, we will present how the stage of illness (number of previous depressive or manic mood episodes) relates to the above described findings.
Disclosure: Nothing to Disclose.

Panel
6. Tracking Mediators of Susceptibility and Resilience to Psychopathology During Adolescent Brain Development

6.1 Molecular Mechanisms Driving Prefrontal Cortex Development in Adolescence

Cecilia Flores

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Background: Adolescence is an age of heightened vulnerability to develop psychiatric disorders that involve alterations in prefrontal cortex circuitry and cognitive dysfunction. The maturation of prefrontal cortex function is linked to the

establishment of dopamine connectivity in this region. Development of mesocortical dopamine is a protracted process that peaks in adolescence and finishes only in early adulthood. Because of its extended maturational course, this system is particularly susceptible to environmental influences. Yet there is a significant gap in our knowledge about the cellular and molecular mechanisms underlying adolescent prefrontal cortex dopamine development and how they are influenced by experience.
Methods: We examined the role of the Netrin-1 guidance cue receptor, DCC, and its microRNA repressor, miR-218, on adolescent mouse prefrontal cortex development. We used axon-initiated recombination and cell-specific knock-down techniques to characterize the spatiotemporal growth of mesocortical dopamine axons and the role that DCC and miR-218 play in this process. Next, we assessed whether stimulant drugs in adolescence alter miR-218/DCC signaling, thereby disrupting mesocortical dopamine axon growth. Finally, we determined whether altered dopamine axon growth influences prefrontal cortex organization and function by quantifying pyramidal neuron morphology and cognitive performance in adulthood.
Results: Here we show, for the first time, that dopamine axons continue to grow from the nucleus accumbens to the prefrontal cortex during adolescence. We discovered that DCC receptors control the extent of this protracted growth by determining where and when dopamine axons recognize their innervation target. Exposure to stimulant drugs leads to disruption of DCC-dependent adolescent targeting events, causing dopamine axons that should innervate the nucleus accumbens, to grow ectopically to the prefrontal cortex. This effect profoundly changes prefrontal cortex structural and functional development, producing alterations in cognitive processes known to be impaired across psychiatric conditions. Importantly, miR-218 controls DCC receptor expression in dopamine neurons across postnatal development and acts as a molecular mediator of the effects of stimulant drugs on prefrontal cortex development.
Conclusions: The prolonged growth of dopamine axons during adolescence represents an extraordinary period for experience to influence their growth and predispose to or protect against psychopathology. MicroRNA control of DCC receptor in dopamine neurons is a molecular link where genetic and environmental factors seem to interact in adolescence to influence the development and function of the prefrontal cortex.
Disclosure: Nothing to Disclose.

6.2 Impact of the 22q11.2 Microdeletion on Adolescent Brain Development

Francesco Papaleo

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Background: The hemizygous genetic deletion in the 22q11.2 locus causes a syndrome (22q11DS) characterized by developmental social and intellectual disabilities, high prevalence of attention deficit hyperactivity disorder (ADHD; ≈37%) during childhood and schizophrenia (≈41%) in adulthood. Although this peculiar behavioral alterations, the specific brain and molecular factors influencing these developmental trajectories are still unknown. Preclinical animal studies could help to disentangle these mechanisms. However, no studies in animal models had so far checked the impact of the 22q11.2 microdeletion in behavioral phenotypes from birth to adolescence, to adulthood.
Methods: Use of LgDel mutant mice that carry the same 1.5 Mb deletion of the human 22q11.2DS and their wt littermates tested in a series of behavioral paradigms and anatomical assessments with a special focus on the adolescent transitional period from prepubertal to adulthood.
Results: We first unraveled in LgDel mice altered startle responses at pre-pubertal ages (postnatal day PND 14) that ameliorated in early development from PND 19. Moreover, sensorimotor gating deficits started to appear as early as PND 19 lasting throughout adulthood. Motor coordination assessed with the Rotarod Test, instead revealed in LgDel mice motor deficits in pre-pubertal period (PND 15-16), that disappeared from adolescence (PND 35). Next, in an implemented 5-Choice Serial Reaction Time Task, we found that LgDel adolescent mice showed selective higher distractibility than controls as it has been shown in patients with schizophrenia. All these developmental behavioral alterations were accompanied by selective altered maturation of the prefrontal cortex, as demonstrated by parallel studies in mice and humans.
Conclusions: Overall, our experiments are starting to elucidate how clinically-relevant genetic alterations can influence the developmental trajectories of behavioral phenotypes through an altered maturation of the prefrontal cortex. This will be important in the context of the development of early diagnosis and therapeutic intervention.
Disclosure: Nothing to Disclose.

6.3 Neuroimaging Evidence of Developmental Changes in Mesolimbic and Cortical Processing Through Normative Adolescence

Beatriz Luna

University of Pittsburgh, School of Medicine, Western Psychiatric Institute & Clinic, Pittsburgh, Pennsylvania, United States

Background: Critical maturational changes in the dopamine (DA) system in adolescence may play a significant role in plasticity of systems underlying cognitive control that can inform susceptibility for impaired development and mechanisms of resilience. In our Experience-Driven Adaptive Cognition (EDAC) Model (Murty et al., 2016) we propose that underlying unique refinements in cognition through adolescence is the increased integration of hippocampus (HPC) and prefrontal (PFC) systems underlied by enhanced modulation of DA processing. To test this model we present results from two studies that probe developmental changes through adolescence in DA related to striatal, HPC, and PFC systems. We characterize developmental changes through adolescence in task and resting state fMRI (rsfMRI) connectivity amongst mesolimbic DA related regions, including VTA, striatum, HPC, and PFC and its relationship with executive function. Additionally, we present relationships to Positron Emission Tomography (PET) data on DA processing.

Methods: Multimodal imaging including resting state and task functional Magnetic Resonance Imaging (fMRI) and PET were used to characterize developmental changes in the integration of DA related regions including striatum and ventral tegmental area (VTA) with HPC and PFC. The first study characterized background fMRI connectivity of VTA and Nucleus Accumbens (NAcc) during a rewarded cognitive antisaccade fMRI task in 170 10-30-year olds in a 3 year longitudinal project. The second study characterized rsfMRI connectivity between regions of HPC and PFC in 143 subjects in a longitudinal study (1-10 visits) and in a PET/MR study of 80 18-30-year olds. Associations and mediation effects with DA related connectivity as well as executive function were also probed.
Results: The first study showed a significant developmental decrease in VTA/NAcc background connectivity that was partially mediated by vmPFC activity and was exclusive to rewarding and not neutral contexts. The second study revealed that bilateral anterior and posterior HPC connectivity to both left and right vmPFC increased with age and was partially mediated by VTA-vmPFC connectivity. There were no age associations for connectivity to either dlPFC or vlPFC. Importantly, age-related changes in HPC-vmPFC coupling predicted individual differences in performance on the Stockings of Cambridge task after controlling for age. Relationships with PET indicators of pre- and post-synaptic DA will also be discussed.
Conclusions: Together, our results suggest that aspects of subcortical DA processing are adult like by adolescence except in rewarded contexts where there is hyperconnectivity in adolescence. DA processing contributing to cortical integration, however, continues to strengthen through adolescence, mediating enhancements in HPC/PFC connectivity supporting maturation of cognitive processing. These findings support our EDAC model of development proposing that unique to the adolescent period is the integration of HPC memory systems and PFC value-based decision-making processes underlied by changes in DA. The relative maturational stages of these processes may be critical for adaptive processes of increased motivation and integration of experience supporting adult level executive function. Deviations from this developmental trajectory may inform impaired development and mechanisms of resilience.
Disclosure: Nothing to Disclose.

6.4 Neurocognitive Correlates and Consequences of Psychiatric Symptom Domains During Adolescence

Patricia Conrod

Université de Montréal, Montreal, Canada

Background: Most psychiatric conditions have their first symptom onset during adolescence, yet, there is little known about the neurodevelopmental consequences of early onset psychiatric symptoms on adolescent development. In this presentation, we present data on the structure of psychopathology in adolescence, the neurocognitive correlates of these latent dimensions of risk and the neurodevelopmental consequences of early onset symptoms on adolescent cognitive development.
Methods: The Co-Venture Trial is a large cluster-randomized trial of personality-targeted interventions on five-year adolescent substance use and cognitive outcomes. Approximately 3800 students consented to and completed personality, psychiatric, substance use and cognitive measures upon entry into high school in the 7th grade and annually until the end of school in the 11th grade. Schools were randomized to deliver the Preventure Programme, a selective cognitive-behavioural intervention targeting 45% of the sample reporting elevated personality risk for early onset substance misuse. Using the longitudinal data from this trial, we conducted hierarchical structural modeling to examine the structure of psychopathological symptoms at 12 and 16 years of age, when substance use significantly increases from under 5% to greater than 80% in this sample. Using multi-level modeling, we examined three different potential relationships between symptom dimensions and cognitive development: common vulnerability, pathoplastic and neurotoxic.
Results: Results indicate that the structure of psychopathology in this young sample is highly consistent with previous studies suggesting one general psychopathology factor and three sub-factors representing internalizing, externalizing and thought disorder dimensions. Multi-level regression models with time-lagged functions revealed common underlying vulnerability, pathoplastic and neurotoxic relationships between early onset of substance use and depression on specific neurocognitive outcomes. Depression showed neurotoxic effects on episodic memory, working memory and perceptual reasoning; cannabis on response inhibition, and despite some pathoplastic effects, alcohol was not shown to have neurotoxic effects on the measures assessed in this study.
Conclusions: Adolescent psychopathology has a complex structure involving symptom dimensions that are hierarchically organized. Furthermore, the early onset of such symptoms impacts on adolescent cognitive development, suggesting the need for selective or preventative interventions delivered early in adolescence to prevent potential neurocognitive consequences of mental health symptoms and problems.
Disclosure: Part 1: Janssen, Honoraria, AbInBev Foundation, Advisory Board.

Panel
7. Towards Translational Brain Imaging Biomarkers: Successes and Challenges

7.1 The Impact of Accelerated Neuroimaging on Neuroimaging Research

Kelvin Lim

University of Minnesota, Minneapolis, Minnesota, United States

Background: Accelerated magnetic resonance imaging techniques that were developed by the Human Connectome Project have greatly enriched the types of imaging information that can be collected. This presentation will provide background on the acceleration techniques and how they have been used to enhance structural, functional and diffusion imaging.

Methods: The basic strategy of acceleration will be explained and advantages described by example. By collecting imaging data from multiple slices simultaneously, routine accelerations of a factor of 8 can be routinely achieved. These have been applied across the different magnetic resonance modalities including structural MRI, functional MRI and diffusion MRI. Among the advantages for functional MRI has been improved spatial resolution to 2mm isotropic (improved from 4mm isotropic standard) and improved temporal resolution of 800 msec (improved from 2000 msec standard). Accelerated imaging approaches have been integrated into two large NIH supported initiatives: the NIDA ABCD study and the Human Connectome Project. The acquisition approaches used in both of the studies will be compared.
Results: The ability to acquire more data has especially helped diffusion MRI. More complex and advanced diffusion models beyond the standard diffusion tensor model had been limited by the long acquisition times using non-accelerated imaging methods, making them impractical for clinical populations. These new diffusion models provide unique information about tissue status that have potential for use as biomarkers. The diffusion tensor model will be contrasted with the multishell diffusion models now available such as diffusion kurtosis, q-space and NODDI.
Conclusions: Accelerated imaging has provided the greatest advance in neuroimaging since the introduction 25 years ago of echo planar imaging and shielded gradients which enabled functional MRI and diffusion MRI. New diffusion models have the potential to provide unique information about tissue characteristics.
Disclosure: Nothing to Disclose.

7.2 Network-Based Approaches for Computing Robust and Reliable Brain Imaging Markers of Illness From Functional and Structural MRI Data

Vince Calhoun

The Mind Research Network, Albuquerque, New Mexico, United States

Background: The advent of the 'big data' era and the prevalence of advanced analytic approaches has accelerated the search for brain imaging markers of illness. In particular, the use of brain imaging data combined with machine learning to perform individual subject prediction has gained increasing interest. However, there are also a number of challenges including the need to calibrate across different scanners and protocols/pulse sequences, the relevance of the prediction to clinical practice, and the interaction between the approach and the data collection approach. The use of multimodal data approaches has also proven a promising way to both leverage available information but also to calibrate across scanners. In this talk, I will discuss the analysis of large data sets with a focus on functional and multimodal imaging. Several strategies for identifying robust 'network-based' markers of mental illness in SZ, BP, and MD will be presented and discussed. I will also discuss strategies for harmonizing across different MRI scanners, using 'big data' approaches for assessing pulse sequences and scanner differences, and visualizing the data.
Methods: We focus on advanced approaches to extract multiple brain networks from resting fMRI of individual subjects (including a spatially constrained group independent component analysis), followed by the use of these networks and their temporal properties to predict mental illness. We present the use of this approach to classify individuals into categories of bipolar and major depression in a set of patients with mixed initial diagnoses. In addition, the use of a nonlinear data visualization approach to identify systematic differences between subjects, sites, and scanners will be shown. And finally, the use of multimodal data fusion approaches to address cross-site calibration issues will be shown in an analysis using both structural and functional MRI data.
Results: Prediction of correct diagnoses was over 90% based on follow-up information for originally misdiagnosed bipolar and major depression. The use of patterns (brain networks) as opposed to regions of interest shows potential for providing a generalizable measure of brain disease. The use of high-dimensional visualization shows systematic but correctable differences across scanner field strength. Multimodal fusion approaches appear to mitigate some of the effects of cross-site data acquisition.
Conclusions: The combination of network-based approaches leveraging higher order statistical information, advanced visualization approaches, and multimodal information show considerable promise in the development of brain-based markers of mental illness. Many challenges remain, but the path forward looks promising.
Disclosure: Nothing to Disclose.

7.3 Combinations of Features as Structural Imaging Biomarkers

Jessica Turner

Georgia State University, Atlanta, Georgia, United States

Background: The relationship between clinical outcomes and morphometric measures in schizophrenia is complex, with a broad literature both finding and failing to find links between volumetric or shape measurements and clinical symptoms or treatment response. Large-scale meta-analyses have confirmed regional gray matter relationships with overall positive and negative severity, and individual regions have been related to the severity of hallucinations and delusions, for example. The combination of regions, their covariation and interactions, are higher-order features and combinations of features which also can be used as potential biomarkers. However, are these features repeatable, and do they relate to the disorder in anyway? We report several studies using structural imaging to identify multivariate features that are robust across datasets; and related to symptom profiles or patterns. The analyses depend on the use of Independent Component Analysis (ICA) in large, aggregated datasets of individuals with schizophrenia and age- and gender-matched controls, scanned in a variety of scanners and protocols. ICA on structural images identifies spatial maps decomposing the data into independent dimensions, and each subject's data are represented as a linear combination of those dimensions or gray matter patterns, with a loading for each pattern. Differences in the loading coefficients are a feature which can be tested for the

effects of diagnosis group, prognosis, and symptom severity; or they can also be combined to form a higher-order feature for consideration.
Methods: T1-weighted images (N = ~600, individuals with schizophrenia (SZ) and age-matched healthy controls (HC)) were processed using a standard pipeline in SPM8, including co-registration to a template, segmentation and normalization followed by smoothing. The smoothed gray matter images are corrected for age, gender, and scanning site effects prior to the ICA. In the first study, we performed ICA on the structural images, and for those dimensions which differed between HC and SZ, we looked at their combination in a biclustering technique and compared the outcome to PANSS scores, to determine if the clusters differed by symptom severity. In the second study, we examined the clinical symptoms along with the images in a parallel ICA, identifying patterns of symptom measures which correlate with patterns of gray matter covariation.
Results: Diagnostic group (SZ vs HC) showed strong differences in the gray matter patterns covering the salience network and the frontal cortex, which were repeated across subsets of the data from different scanners. The biclustering technique identified that the two components singled out subjects at the extremes—subjects with extreme values on both components showed one pattern of symptom severities, while subjects with extreme values on only one but not the other showed a different pattern. In the pICA outcomes, gray matter patterns were correlated with specific patterns of symptom severities.
Conclusions: These results argue for a more holistic approach to structural imaging biomarkers—examining networks of gray matter in a data-driven method can be informative and reliable, and in combination with patterns of phenotypes can identify groups which would not be distinguishable using simpler measures. The caveats are as always the longitudinal nature of these results—these are cross-sectional analyses and cannot at the moment provide prognostic information, and almost all the subjects were medicated, confounding disease status, progression, and medication effects.
Disclosure: Nothing to Disclose.

7.4 Imaging Biomarkers in Clinical Trials of Schizophrenia and Major Depression

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Background: The marked heterogeneity of clinical response remains a critical challenge in optimizing treatment strategies for the major psychiatric disorders. Multiple potential biomarkers have been assessed, including plasma metabolites, electrophysiological measures and genetic variants, but to date the majority of clinical trials do not include a priori stratification variables to enhance potential efficacy signals. This has resulted in increasingly large sample sizes in clinical trials, with concomitant increased expenses and the potential introduction of additional heterogeneity as additional sites and personnel are needed to achieve sufficient power.
Methods: In addition to a focus on genetic biomarkers, we have introduced resting state MRI (rs-fMRI) into clinical trials in schizophrenia and depression. In a clinical trial comparing aripiprazole with risperidone in first episode schizophrenia ($n = 198$), we conducted rs-fMRI prior to treatment initiation and again following twelve weeks of treatment. We placed 12 striatal seeds and assessed connectivity across the brain at baseline and at 12 weeks in order to identify a priori predictors of response as well as the neural circuitry that changes as psychosis diminishes. In a clinical trial ($n = 16$) involving electroconvulsive therapy, we assessed rs-fMRI at baseline, following the first ECT administration, and at endpoint. The primary measure derived from rs-fMRI was fractional amplitude of low frequency fluctuation (fALFF), which provides an unbiased voxel-wise estimation of brain activity. We compared treatment-related changes in mood ratings with pre- and post-treatment fALFF and connectivity measures.
Results: In first episode schizophrenia, we detected a corticostriatal resting state biomarker that was associated with clinical efficacy following twelve weeks of treatment; these results were replicated in a naturalistic study of 40 inpatients hospitalized for psychosis and treated with antipsychotic agents. Moreover, connectivity was significantly influenced by the duration of untreated psychosis (DUP) of the subjects prior to entering the trial; DUP and corticostriatal connectivity interacted to significantly influence overall treatment response. With ECT, the subcallosal cingulate cortex (SCC) demonstrated higher fALFF at baseline in depressed patients, and SCC fALFF decreased over the course of treatment. The baseline level of fALFF of SCC predicted response to ECT. In addition, the connectivity of SCC with bilateral hippocampus, bilateral temporal pole, and ventromedial prefrontal cortex was significantly reduced over the course of ECT treatment.
Conclusions: Resting state MRI measures may provide unique information relating to treatment responsivity in major psychiatry disorders. Early data has been promising; however, challenges remain in developing drug-specific biomarkers, as well as developing methods for utilization in larger scale trials that utilize multiple sites and diverse MRI platforms. Future directions also include the use of multimodal imaging measures, as well as the incorporation of additional biomarker approaches (e.g. pharmacogenetic markers), to enhance these precision medicine strategies.
Disclosure: Part 1: Genomind Inc., Consultant, Concert Pharma, Consultant, InformedDNA, Advisory Board.

Study Group

8. Establishing Best Practice Guidelines to Improve the Rigor, Reproducibility, and Transparency of the Maternal Immune Activation (MIA) Animal Model of Neurodevelopmental Abnormalities

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Study Group Summary: Animal models complement a large body of epidemiological data showing that microbial

infections during pregnancy can have long-term effects on the neurodevelopment of the offspring. The maternal immune activation (MIA) model, which uses either live pathogens or (more commonly) chemical mimics of different types of infections, is used to define relevant cellular and molecular mechanisms. While a number of investigators use different MIA models with distinct advantages/pitfalls, there are very few training opportunities to consult and establish a consensus on the appropriate design, implementation and reporting of this work. Moreover, several issues reported in the literature remain unanswered when validating these models as a proxy for neuropsychiatric disorders. The goals of this study group are to (1) foster discussion on the best practices to be implemented in MIA studies; (2) identify the data that need to be collected to harmonize procedures; (3) establish an evidence-based practice design; and (4) inform the MIA research community of these findings to ensure that MIA-induced neurobehavioral outcomes can be reproduced and translated to humans.

Study Group Format: In this session, we will review the MIA models and discuss their strengths and weaknesses. The participants will address the importance of validating the different immunogens (e.g., poly (I:C), lipopolysaccharide (LPS) and attenuated (inactivated) microbes) and standardizing the general design of MIA models both within and across laboratories. The presenters will address the important issues related to animal species and strains, vendors, administration protocols, and assessment of consequences such as sickness or generalized inflammation. There will be a special emphasis on examining the influences of husbandry conditions such as caging systems, environmental enrichment, diet and microbiome, breeding on site vs. time pregnant, cross-fostering, standard litter size and group allocation.

We will also discuss important biological variables and endpoint considerations in study design (e.g., age of testing, sex and treatments/interventions) and reporting guidelines to ensure data transparency. The experts in clinical and epidemiological research will critically assess the MIA models in the context of human psychiatric disorders. The participants will also discuss new directions for future MIA studies.

We believe that our study group discussion will facilitate the development of best practice guidelines for standardized MIA models in order to improve the rigor, reproducibility, and transparency of this research.

Disclosure: Nothing to Disclose.

Mini Panel
9. Stressing the Balance: The Impact of Stress and Stress Mediators on Excitability and Synaptic Plasticity Throughout the Brain

9.1 Corticotropin Releasing Factor (CRF) Indirectly Activates M5 Receptors on Dopamine Terminals in the Nucleus Accumbens to Facilitate Approach Behaviors

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Background: Depressed individuals are more likely to withdraw from the environment and respond to acute mild stressors with avoidance while, for healthy individuals, acute mild stressors often serve as motivators and drive engagement. Therefore, understanding the mechanisms by which acute stressors motivate or demotivate individuals under different conditions is key to understanding a cardinal feature of depression. It was recently shown that the stress-associated neuropeptide, corticotropin releasing factor (CRF) potentiates dopamine transmission in the nucleus accumbens (NAc) to promote approach behavior such as exploration of a novel object. Interestingly, repeated stress disrupts CRF's ability to increase dopamine and switches the behavioral response to CRF from approach to avoidance. Understanding the precise mechanism by which CRF regulates dopaminergic neurotransmission, at the level of the presynaptic terminal, will bring us one step closer to understanding how this interaction becomes dysregulated after repeated stressor exposure.

Methods: Electrophysiology: 240 μm coronal sections were prepared and maintained in ACSF, and either cell-attached or whole cell patch clamp recordings were made from cholinergic interneurons. Fast Scan Cyclic Voltammetry: 240 μm coronal sections were prepared and maintained in ACSF. Carbon fiber electrodes (working electrodes) were hand cut to approximately 100-150 μm past the capillary tip. The potential at a carbon-fiber electrode was held at -0.4 V versus Ag/AgCl, ramped to +1.2 V and back to -0.4 V (400 V/s) every 100 ms a custom written Igor-based software. Behavior: Wildtype or M5KO mice were habituated to a 40 x 40 cm arena for 30 min. Following a 5-min interval in the homecage, animals were placed back in the arena where a novel object was placed in the center. Behavior was monitored and analyzed using Noldus Ethovision software.

Results: We now show that CRF produces a robust increase in the firing of cholinergic interneurons (CIN) within the dorsal striatum and nucleus accumbens (540% and 247% for CRF 100 nM respectively). This increase CIN firing is through a CRF R1, PKA pathway dependent mechanism. Moreover, inhibition of the Ih conductance by ZD 7288 significantly reduces the effect of CRF on CIN firing. We replicated our past findings demonstrating that CRF increases the peak dopamine transient measured using ex vivo fast scan cyclic voltammetry. This increase is not readily reversed by prolong wash-out with a CRF receptor antagonist. We have identified two components driving the CRF mediated potentiation of dopamine transients – muscarinic acetylcholine 5 (M5) receptor dependent and independent components. Interestingly, global M5-R deletion reduces novelty exploration similarly to intra-NAc CRF receptor antagonism, suggesting that M5 activation is critical for engaging in this approach behavior. We assessed the effect of repeated stress on CRF effects on both the cholinergic and dopaminergic systems within the NAc. Surprisingly, repeated stress had no effect on CRF's ability to increase CIN firing. However, it did significantly reduce CRF's effect on dopamine potentiation. Further analysis, demonstrated that the M5 dependent component remains intact, while the M5-independent component is disrupted.

Conclusions: The results of this study provide evidence of a novel action of CRF in the striatum. Here we show that CRF enhances cholinergic interneuron firing. Furthermore, one consequence of this CRF-mediated increase in

cholinergic firing is to potentiate dopamine transmission via activation of M5 receptors. Interestingly, repeated stressor exposure has a different effect on CRF receptor efficacy at cholinergic neurons compared to dopamine terminals suggesting that stress can modulate CRF receptor signaling differently based on cell type, even within the same region.
Disclosure: Nothing to Disclose.

9.2 Corticotropin Releasing Factor (CRF) Alters the Inhibitory Control of CRF Receptor 1 (CRF1+) Neurons in the Mouse Central Amygdala

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Background: The central nucleus of the amygdala (CeA) is a primarily GABAergic nucleus involved in alcohol dependence, anxiety, and depression. Changes in both the tonic and phasic forms of GABAA receptor inhibition in the CeA have been reported following acute and chronic ethanol exposure in rats and mice. Corticotropin releasing factor (CRF) and the CRF 1 receptor have been implicated in the effects of ethanol and stress, but how CRF selectively engages the specific circuitry of the CRF/CRF1 system in the CeA remains unclear.
Methods: Whole-cell voltage- and current-clamp recordings of CRF1+ CeA neurons from mice expressing green fluorescent protein (GFP) under the CRF1 promoter (CRF1:GFP) were performed to measure pharmacologically-isolated phasic and tonic GABAA receptor-mediated inhibitory transmission during focal application of CRF and the CRF1 antagonist R121919. To selectively label neurons that project out of the CeA, fluorescent microspheres were injected into the dorsolateral bed nucleus of the stria terminalis (dlBNST), the mice were allowed to recover for one week after surgery prior to electrophysiological recording to allow for retrograde transport to the CeA.
Results: CRF (200 nM) produced a significant increase in holding current and in spontaneous inhibitory postsynaptic current (sIPSC) frequency, but not sIPSC amplitude, in CRF1+ CeA neurons, indicative of a simultaneous increase in GABA release onto, and tonic inhibitory control of, CRF1+ CeA neurons. In contrast, R121919 (1 μM) produced a significant reduction in holding current and a trend for a decrease in sIPSC frequency, but not sIPSC amplitude, suggesting an overall reduction in phasic and tonic inhibitory control of CRF1+ neurons. In CRF1+ CeA neurons labeled with microspheres injected into the dlBNST, CRF produced similar increases in phasic and tonic inhibition to what was observed in unlabeled CRF1 + CeA neurons, indicating that CRF is exerting this inhibitory control over CRF1+ neurons that project out of the CeA.
Conclusions: These data indicate that CRF increases phasic and tonic inhibition of CRF1+ CeA neurons through pre- and postsynaptic mechanisms. The increase in GABA release onto CRF1+ CeA neurons indicates that CRF1 receptors are on presynaptic terminals that synapse onto CRF1+ neurons in the CeA. While the origin of the CRF1-containing presynaptic terminals is not clear and could be from either extrinsic (e.g. basolateral amygdala) or intrinsic (local CeA neurons) sources, this finding suggests that the there is a direct connectivity between CRF1+ neurons in the amygdala, which has important consequences for how this system is engaged in times of stress or in the pathological activation of the stress system. As tonic inhibition is a strong determinant of excitability, it is also noteworthy that CRF was able to stimulate a postsynaptic tonic conductance in CRF1+ neurons in the CeA. The mechanism underlying the ability of CRF to stimulate this tonic conductance is not clear, but adds a new means by which CRF can exert its effects on CeA neurons. A CRF1 antagonist was shown to inhibit a tonic conductance in CRF1+ CeA neurons, suggesting the presence of a baseline tone in the CRF1 system that contributes to the ongoing tonic inhibition of CRF1+ CeA neurons. These data provide new information on how CRF can alter the activity of the CRF1 system in the CeA, which has relevance to the clinical conditions of drug and alcohol dependence, anxiety, and depression.
Disclosure: Nothing to Disclose.

9.3 Synaptic Imprinting Following Transmitted Stress

Jaideep Bains

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Background: Authentic stress is remembered by the brain through enduring changes in key neural circuits. Humans, primates, and rodents, respond to distress in conspecifics. These interactions console the distressed individual, but also transmit stress to the naïve individual. It is not known whether transmitted, or second order stress is captured in a persistent fashion by brain circuits.
Methods: All animal protocols were approved by the University of Calgary Animal Care and Use Committee. Male and female Crh-IRES-Cre; Ai14 mice in which CRH neurons express tdTomato fluorophore were used in the present study, at 4- to 8-weeks old. The study used a combination of electrophysiology, optogentic and behavioral approaches.
Results: Transmitted stress imprints synapses in a naïve partner mouse following social interaction with a stressed conspecific. These changes mimic the effects of the authentic stress experience and are transmitted through a putative alarm pheromone from the stressed individual to the partner. Silencing hypothalamic corticotropin-releasing hormone cells in the naïve partner during the interaction with the stressed individual, blocks the transmitted synaptic imprint. In the absence of stress, optogenetic activation of CRH neurons in one individual is sufficient to cue investigative behavior in the partner and generate synaptic imprints in both individuals.
Conclusions: Our findings reveal an unexpected role for PVN CRH neurons in transmitting distress signals and demonstrate that transmitted stress has the same lasting effects on the brain as authentic stress.
Disclosure: Nothing to Disclose.

Panel 10. Multimodal Strategies to Identify Precision Biomarkers of Treatment Response in Depression

10.1 Electrophysiological Neuromarkers in Understanding Mechanisms of Action of Different Antidepressant Modalities and Predicting Clinical Response in Major Depressive Disorder

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Background: Major depressive disorder (MDD) is a leading cause of disability in the world. An alarming 30% of MDD patients, that is over 100 million people worldwide, suffer from treatment-resistant depression (TRD) and fail to respond to pharmacological antidepressants (e.g., selective serotonin reuptake inhibitors (SSRIs)) or cognitive behavioral therapy (CBT). In such cases, electroconvulsive therapy (ECT) is the most effective treatment. It is understood that the electrical stimulation through ECT impacts distributed sets of brain networks disrupted in MDD. However, such non-specific stimulation can also affect brain networks involved in cognition, leading to severe side effects such as memory impairment. The effort to develop alternative treatments has resulted in a number of clinical trials demonstrating that ~30% of TRD respond to focal non-invasive brain stimulation, repetitive transcranial magnetic stimulation (rTMS). However, despite decades of research, it is still unclear why SSRIs and CBT are effective in some individuals and not others, why ECT remains to be the most effective treatment, or how to improve rTMS efficacy. This knowledge gap is in part due to lack of centralized initiatives that empower the possibility of monitoring the mechanisms of action of different antidepressant modalities in well-characterized cohort of patients using standardized methods of study design, data collection and analyses.
Methods: In association with Canadian Biomarker Integration Network in Depression (CAN-BIND), we formed an Electroencephalography (EEG) working group to address this knowledge gap. We collected high density EEG (> 60 channels) during resting-state in patients with MDD as they received a) escitalopram and aripiprazole ($n = 126$) in a clinical trial conducted across four institutes in Canada (i.e., University Health network, Centre for Addiction and Mental Health, Queen's University, and University of British Columbia), or b) cognitive behavioral therapy ($n = 40$) (at Centre for Addiction and Mental Health – on-going study that terminates in July 2017). Standardized electrophysiological recordings were conducted at baseline and then longitudinally at week 2 and week 8 of therapy; Week 16 data was collected for patients receiving CBT. Similar recordings were obtained in healthy controls ($n = 50$). Resting-state EEG was also collected in TRD before and after a course of ECT conducted at Centre for Addiction and Mental Health ($n = 20$). We created a standardized EEG analysis toolbox to pre-process and analyze EEG collected from all sites and across different antidepressant modalities (SSRI, CBT, ECT).
Results: Our preliminary results thus far indicate several unique as well as overlapping EEG markers that predict response to each antidepressant modality. Our preliminary analyses suggest that non-linear and network-specific metrics may be superior to linear metrics in predicting treatment response, and explaining the mechanisms of actions of antidepressants.
Conclusions: Several of our identified markers can be reliably recorded from less than 10 electrodes and thus likely through portable easy to set-up dry electrode EEG systems. Such systems can be easily deployed to clinics or utilized in replication trials with significantly less cost than high-density EEG. Collectively, through a multi-site, multi-investigator, and multi-clinical trial effort we provide promising evidence that support the practical application of EEG neuromarkers for understanding mechanisms of action of antidepressants and ultimately, upon replication studies, informing routine clinical care in MDD.
Disclosure: Nothing to Disclose.

10.2 Imaging Approaches to Clinical Staging of Treatment Resistant Depression

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Background: Treatment resistant depression (TRD) is currently defined clinically, based on failure to respond to two or more conventional antidepressant treatment strategies. Needed are classification strategies that consider quantitative neurometrics. We have previously defined brain-state patterns that predict differential remission and failure to antidepressant medication and cognitive behavioral therapy in MDD patients using both resting state fMRI (rs-fMRI) and FDG PET. As a next step, we examine the use of rs-fMRI functional connectivity to define patterns that classify different levels of TRD.
Methods: Baseline rs-fMRI scans from 3 MDD cohorts (total $n = 191$) with known past treatment histories and current episode treatment outcomes were compared: Cohort 1: combined CBT+medication failure but previously treatment naïve ($n = 25$); Cohort 2: combined CBT+medication failure but previously treatment responsive ($n = 11$); Cohort 3: multiple previous and current episode treatment failures including ECT ($n = 23$). All patients were scanned using an identical imaging protocol: 3 Tesla Tim-Trio MRI, high resolution T1 structural scan and 7-min eyes open, resting state functional scan. A standard pre-processing pipeline was applied and statistical analyses were performed in AFNI. A whole brain functional connectivity (FC) ANOVA using a bilateral subcallosal cingulate seed (SCC) was used to assess differential treatment resistant patterns as a function of past treatment exposure.
Results: Two TRD brain-state patterns were identified that differentiated the 3 TRD cohorts. Functional connectivity of the SCC to the left dorsolateral prefrontal cortex (DLPF BA46/10) distinguished the treatment naïve dual failures from both the previously treated and multiple treatment failure patients. In contrast, SCC-FC of the supplementary motor area (SMA6) and mid-cingulate cortex (MCC) distinguished the multiple treatment failure group from both

other cohorts. No pattern was shared by the multiple failure and treatment naïve groups.
Conclusions: Differential baseline abnormalities are identified in MDD patients with different levels of treatment resistance. These findings have potential implications for treatment selection strategies in both clinical practice and experimental treatment protocols.
Disclosure: Part 1: St. Jude Medical, Inc., Patent, **Part 2:** St. Jude Medical, Inc., Patent.

10.3 Transcriptomic Approaches Towards Biomarker Development in Depression

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Background: Increasing evidence supports the view that the heterogeneous syndrome of depression and its treatment by a variety of medications and psychotherapeutic approaches is associated with altered functioning across limbic brain regions.
Methods: Our laboratory and others are employing open-ended exploration of gene expression and chromatin abnormalities in these brain regions by use of several genome-wide approaches, including RNAseq and ChIPseq, to define the molecular basis of depression and antidepressant responses in mouse models, with findings in animals validated in postmortem human brain tissue.
Results: Our findings to date provide novel insight into the regions of the brain where traditional antidepressants (e.g., SSRIs) versus experimental antidepressants (e.g., ketamine) act to produce their therapeutic effects. They also define putative molecular mediators of antidepressant response as well as the molecular basis of non-response to these agents in mouse depression models. For example, treatment response seems to reflect a mixture of reversing pathologic changes seen in susceptible animals, inducing changes that mediate natural resilience, as well as additional mechanisms unassociated with stress responses. Likewise, treatment non-response is characterized by the failure to induce many of the changes seen in responders, but also with the induction of novel changes that could conceivably oppose treatment response. Parallel studies of blood indicate that most changes seen in brain are not recapitulated in blood; this is not surprising because most changes differ even across different regions of the brain. Nevertheless, some changes in brain are recapitulated in blood, and several other changes in blood, while not the same as those seen in brain, nonetheless might accurately reflect brain adaptations.
Conclusions: Further empirical research will confirm whether any of these peripheral changes could be used as bona fide biomarkers for depression and its treatment. This work also defines novel molecular pathways that is now being mined for the development of novel therapeutics for depression. Finally, our unbiased analyses demonstrate dramatic differences in the transcriptomic signatures of depression and its treatment in males versus females, which sets the stage for novel sex-specific approaches to developing biomarkers and treatments for depression.
Disclosure: Nothing to Disclose.

10.4 Short Non-Coding RNA and Antidepressant Response

Gustavo Turecki

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Background: Antidepressants (ADs) are the most common treatment for major depressive disorder (MDD). However, only about 30% of patients experience adequate response after a single AD trial, and this variability remains poorly understood. Genes can be regulated through the activity of several non-coding RNA (ncRNA) transcripts that act as fine-tuners and on-off switches of gene expression patterns. Among ncRNAs, small non-coding RNAs, including microRNA (miRNA), small nucleolar RNA (snoRNA) and P-element induced wimpy testis-interacting RNA (piRNA), are particularly interesting because they play a key role in the regulation of essential brain processes, seem to mediate psychiatric treatments, and circulate freely in blood, making them particularly good biomarker candidates. Here we will present a series of complementary studies investigating small non-coding RNAs as possible biomarkers of antidepressant response.
Methods: We used a series of different methods. We first performed small RNA-sequencing in a randomized placebo-controlled trial of duloxetine ($N = 516$) using blood samples collected before and eight weeks after treatment. We then confirmed our results in an independent clinical trial of antidepressant ($N = 122$), and subsequently, we investigated response to treatment in a well-characterized animal model of depression ($N = 26$). Further, we conducted a series of studies investigating postmortem brain samples and induced neurons expressing a serotonergic phenotype, in addition to RNA-IP, overexpression and downregulation experiments to test the interaction between small ncRNAs and their targets in vitro.
Results: The small RNA-sequencing in the randomized placebo-controlled trial of duloxetine revealed differential expression of several miRNA (miR-146a-5p, miR-146b-5p, miR-425-3p and miR-24-3p), 31 snoRNAs ($FDR < 0.05$) and 3 piRNA ($FDR < 0.05$). Interestingly, all the 31 significant snoRNA and 3 piRNA were upregulated according to response to treatment. The miRNA results were replicated in an independent clinical trial of depressed patients, and were further supported by AD treatment in the animal model of depression, and in postmortem human brain samples. Using bioinformatic analyses, we identified common target genes of the differentially expressed miRNAs, and found a significant dysregulation of genes involved in MAPK/Wnt signaling pathways. The snoRNA and piRNA were characterized in a series of follow up experiments, including in vitro work and functional assays with induced serotonergic neurons.
Conclusions: Together, our results postulate miR-146a-5p, miR-146b-5p, miR-425-3p and miR-24-3p, as well as SNORD11B and SNORD52 are potential biomarkers of AD response. These results provide important steps in the potential development of early diagnostic tools, preventive strategies, and effective pharmacological targets for MDD treatment.
Disclosure: Part 1: Pfizer, Grant, Janssen, Grant

Panel
11. Corticolimbic Trajectories From Neonatal Through Adolescent Critical Periods in Mental Illness Development: Implications for Early Detection, Outcomes and Treatment

11.1 Frontotemporal Gray and White Matter Structural Trajectories in Bipolar and Major Depressive Disorder: Associations With Suicide Risk and Outcomes

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Background: A peak in onset of mood disorders is observed during the mid to late adolescent period. Recent data suggests that early disease progression is associated with altered development of frontotemporal systems. Disease course however is not homogeneous and environmental and neuroanatomical factors can contribute to differences in disease trajectories and outcomes, i.e. suicide attempts. We therefore sought to identify alterations in frontotemporal structural development in youth with bipolar or major depressive disorders and identify baseline predictors and longitudinal trajectories associated with development of suicide behavior and substance use disorders (SUDs).
Methods: Longitudinal data from 137 adolescents/young adults (59% diagnosed with bipolar or major depressive disorder) who completed high-resolution structural MRI and diffusion tensor imaging at two time points will be presented. We will highlight recently published work identifying (1) differences in frontotemporal gray matter volume (GMV) and white matter trajectories in youth with bipolar disorder compared to healthy comparison participants, (2) neuroanatomical circuitry associated with suicide thoughts and behavior, and (3) neuroanatomical predictors of future SUDs. Additionally, we will present unpublished longitudinal data from 48 individuals diagnosed with bipolar or major depressive disorder and 25 healthy comparison adolescents/young adults (63% female, on average 18 years of age at time 1 and 3 years between assessments). Frontotemporal GMV and fractional anisotropy (FA) baseline predictors and trajectories (change overtime) associated with a future suicide attempt were assessed and relationships with clinical phenotype and childhood maltreatment investigated.
Results: Adolescents and young adults with bipolar disorder showed progressive decreases in frontotemporal GMV and white matter FA overtime ($p<0.005$). Magnitude of decreased frontotemporal GMV and white matter FA at baseline was associated with a past suicide attempt and greater risk for developing a SUD ($p<0.005$). When investigating development of suicide behavior, we observed 38% and 25% of youth with bipolar disorder or major depressive disorder respectively had an interim suicide attempt. Having a previous attempt or more hospitalizations was associated with greater risk for a future attempt ($p<0.05$). Magnitude of GMV decreases in orbitofrontal cortex (OFC) at baseline was predictive of a future attempt ($p<0.001$). A significant group by time interaction was observed in OFC white matter FA ($p<0.001$). Future attempt was associated with progressive decreases in FA overtime. Youth not making a future attempt and healthy comparison subjects showed increases in FA overtime. Childhood emotional abuse was associated with greater decreases in OFC FA overtime ($r=-0.48$, $p=0.01$).
Conclusions: Results support abnormalities in development of frontotemporal GMV and white matter FA in mood disorders. OFC GMV and white matter FA trajectories were associated with risk for, and development of, suicide thoughts and behavior. Exposure to childhood maltreatment may contribute to altered white matter development in mood disorders increasing risk for suicide. Findings aid in our understanding of the development of suicide thoughts and behavior in mood disorders and provide possible leads for early intervention and prevention strategies.
Disclosure: Nothing to Disclose.

11.2 Cortical Surface Area Development: A Neural Signature of Depression in Early Adolescence?

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Background: Our recent cross-sectional findings from the ENIGMA Major Depressive Disorder (MDD) consortium indicate that MDD is associated with structural alterations in cortical regions in a highly dynamic way, with different patterns of alterations in adolescence and adulthood. In adolescents with MDD we found pronounced and global surface area alterations, which were absent in adult MDD patients. New cross-sectional ENIGMA MDD data will be presented illustrating complex interactions between age and diagnosis on cortical surface area across the full life span. In addition, data from a longitudinal study that investigated the association between different trajectories of depressive symptoms and longitudinal changes in brain structure throughout adolescence.
Methods: Cortical surface area measures of 64 brain regions from 2,757 MDD patients and 4,513 healthy controls from 29 sites around the world participating in ENIGMA MDD (age range 10-85) were pooled and age by diagnosis interaction effects were examined. In addition, in the Adolescent Development Study, one-hundred-forty-nine adolescents were assessed on depressive symptoms and underwent structural MRI at age 12, and were followed-up multiple times until age 19. Three depressive symptom trajectories (Low-Stable [$n=97$], Early-Decreasing [$n=33$], Late-Increasing [$n=19$]) were identified, and effects of group and group-by-time on hippocampus and amygdala volume, and prefrontal cortical thickness and surface area were evaluated.
Results: ENIGMA MDD analyses showed lower surface area in MDD patients in early adolescence, with highest effect sizes observed in anterior cingulate cortex (ACC), orbitofrontal cortex (OFC), lateral prefrontal regions, insula, superior temporal gyrus and fusiform gyrus. These effects were less pronounced in late adolescence and early adulthood, absent in adults with MDD. Findings from the ADS study showed that the Early-Decreasing Symptoms group exhibited cortical surface area alterations compared to the Low-Stable and Late-Increasing Symptoms groups, moderated by sex. Specifically, females in the Early-Decreasing Symptoms group showed lower ACC and OFC surface areas

across adolescence compared to females in the other groups. Males in the Early-Decreasing Symptoms group showed less right OFC surface area expansion over time compared to males in the Low-Stable and Late-Increasing Symptoms groups. No effects were found for cortical thickness, or hippocampus and amygdala volume.
Conclusions: Alterations in cortical surface area were specifically observed in young people
experiencing depressive symptoms in early adolescence but not in those developing depressive symptoms later in adolescence. In addition, our results indicate that lower surface area can be observed in young people with elevated depressive symptoms of which not all might develop a full-threshold MDD diagnosis. Our findings suggest that early adolescence is a particularly sensitive period for cortical surface area abnormalities associated with depressive symptoms and may provide a critical window for treatment of (sub threshold) depressive symptoms.
Disclosure: Nothing to Disclose.

11.3 Understanding Remission in Attention Deficit Hyperactivity Disorder

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Background: While many children with ADHD show improvement in their symptoms as they grow into young adulthood, many show persistence of the full syndrome. Here we contrast two models of remission. The first posits that remission is due to the emergence of compensatory processes underpinned by neural re-organization. An alternate model holds that remission by adulthood might result from the amelioration of childhood, transient anomalies in brain structure and function. In prior studies of white matter microstructure and cortical architecture, we found evidence supporting the latter model. Here we extend this work to consider brain function.
Methods: We followed prospectively a cohort of 92 children with and 144 without ADHD into adulthood (mean age at study entry 10.2 (SD3) years; adult endpoint 24 (SD4) years). By adulthood, 37 (40%) showed persistent ADHD, the remainder no longer met criteria for ADHD. In young adulthood, multimodal imaging were acquired. (1) intrinsic functional connectivity, defined from task-free functional MRI and magnetoencaphalography- MEG; (2) brain activation (both fMRI and MEG) during a task of motor inhibition.
Results: Young adults whose ADHD has persisted from childhood show atypical neural activity, whereas remitters did not differ from those who were never affected. For intrinsic functional connectivity, those with persistent inattention lost the typical balance of connections within the default mode network (prominent during introspective thought) and connections between this network and those supporting attention and cognitive control. In MEG, these symptom related anomalies were most prominent in the theta and gamma bands. By contrast, adults whose childhood inattentive symptoms had resolved did not differ significantly from their never affected peers, both hemodynamically and electrophysiologically. Further, during a probe of motor inhibition, a core deficit in the disorder, we find remitters show typical hemodynamic and electrophysiological activity at both cortical (right inferior frontal and inferior parietal/precuneus) and cerebellar regions. Here, the persistent ADHD group showed under-activation, whereas the remitted ADHD group did not differ significantly from the never-affected group. MEG showed that the association between adult symptom severity and prefrontal neuronal activity was confined to the time window covering the act of inhibition (300-350 ms). The only exception occurred at the caudate, where anomalies reflected the childhood history of ADHD rather than adult outcome.
Conclusions: Combined, the findings suggest that remission of ADHD symptoms by early adulthood is associated with brain function that largely does not differ from never affected individuals. This finding held at cortical and cerebellar levels, both hemodynamically and electrophysiologically.
Disclosure: Nothing to Disclose.

11.4 Cortical Thickness, Surface Area and Cognitive Development in Very Young Children at Risk for Schizophrenia

John Gilmore

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Background: Adults with schizophrenia exhibit alterations in cortical morphology, including reduced cortical thickness. These alterations can be present in the prodromal phase of the illness as well as in unaffected siblings at genetic high risk. Given the evidence that early brain development is altered in schizophrenia, we studied cortical thickness and surface area in very young children at risk for schizophrenia and other psychiatric disorders.
Methods: Pregnant women with schizophrenia, bipolar illness, and no psychiatric illness were recruited; brain structure of offspring was studied with 3T MRI after birth and at ages 1 and 2 years. Cognitive development was assessed at 1 and 2 years with the Mullen Scales of Early Learning. Regional cortical thickness and surface area was determined using age-specific atlases. Usable scans were obtained in 63 high risk (schizophrenia N = 25; bipolar disorder N = 20; mood disorder NOS N = 18) and 173 control neonates. At age one there were 42 high risk (schizophrenia N = 23; bipolar disorder N = 6; mood disorder NOS N = 13) and 68 controls. At age two, there were 35 high-risk subjects (schizophrenia N = 26; bipolar disorder N = 5; mood disorder NOS N = 4) and 66 control subjects.
Results: Children at risk for schizophrenia had significantly lower Mullen composite scores at one ($p = 0.49$) and two years ($p = 0.0006$). High-risk neonates had significantly reduced total surface area ($p = 0.006$) that was also seen in the schizophrenia ($p = 0.043$) and bipolar groups ($p = 0.088$). Total surface area was significantly reduced in female high-risk neonates ($p = 0.001$), but not in males; total surface area was also significantly reduced in female offspring of mothers with schizophrenia ($p = 0.024$), bipolar disorder ($p = 0.028$) and mood disorder NOS ($p = 0.003$). Interestingly, there was a trend for increased mean cortical thickness in female high-risk offspring ($p = 0.052$), including those at

high risk for schizophrenia ($p = 0.070$). There were no significant differences in total surface area or mean cortical thickness between high risk and control groups at ages one or two years. Interestingly, females at risk for schizophrenia had a few regions of increased surface area at 1 and 2 years of age.
Conclusions: We found that children at risk for schizophrenia had significant delays in cognitive development. Females at genetic high risk for schizophrenia had decreased total cortical surface area compared to controls shortly after birth. At age 1 year, there was no difference in total surface area in female high-risk children, with some regions exhibiting an increase in surface area. These findings support previous studies of altered early child development in children at high risk for schizophrenia, and suggest that there are gender-specific alterations of cortical surface area at birth that tend to normalize in the first year of development.
Disclosure: Nothing to Disclose.

Panel
12. Exploring the Insula's Role in Substance Use Disorders

12.1 Individual Differences in Insula Function Impact Nicotine Dependence

Amy Janes

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Background: The insula's involvement in nicotine dependence has become clear as damage to this region disrupts nicotine use in both humans and rodents. Smoking cue exposure reliably activates the insula and, as we have shown, this region is involved in smoking-related memory retrieval. While the insula may generally play a role in nicotine dependence and smoking cue-reactivity, individual variability of this region's function may impact smoking behavior and treatment outcome. For instance, using data from the human connectome database we showed that nicotine dependence severity is correlated with insula reactivity to negatively valenced stimuli. Our prior work also showed that greater insula reactivity to smoking cues predicts poorer cessation outcome. However, this finding has not been confirmed which is critical given the call for reproducibility especially when using neuroscience to guide treatment development. Further, as the insula and dorsal anterior cingulate cortex (dACC) are primary nodes of the salience network, it is unclear if the functional communication between these regions impacts subsequent smoking cue-reactivity.
Methods: Using functional magnetic resonance imaging (fMRI) we evaluated the pre-cessation brain reactivity to smoking vs. neutral cues in nicotine dependent smokers who were and were not able to maintain subsequent abstinence. In an independent cohort, we correlated the strength of resting state insula-dACC coupling with subsequent brain reactivity to smoking cues.
Results: In cohort one, smokers who relapsed showed enhanced reactivity to smoking cues in the right insula and dorsal striatum showing significant overlap with our prior work despite methodological differences including the fact that our previous work only involved women. In the second cohort, individuals with the greatest insula-dACC coupling at rest had enhanced brain reactivity to smoking cues in regions associated with relapse vulnerability; the right insula and dorsal striatum. Stronger Insula-dACC coupling also was associated with smoking cue-induced ventral lateral prefrontal cortex (VLPFC) reactivity.
Conclusions: The current work supports our prior results and builds on the concept that the insula and dorsal striatum work in concert to maintain nicotine dependence. Specifically, dorsal striatal-mediated habitual responding may be triggered both by the external drug-associated cues, and the insula-mediated internal states that provide additional context motivating drug use. Additionally, insula-dACC functional coupling impacts subsequent brain reactivity to smoking cues enhancing these same brain regions as well as the VLPFC, which is involved in the detection of behaviorally relevant sensory events. Collectively, these findings indicate that individual variability in insula function influences cue reactivity and relapse supporting the need for personalized therapies that consider these neurobiological differences.
Disclosure: Nothing to Disclose.

12.2 A Role of the Insular Cortex in Interoceptive Inference and Decision-Making: Evidence From Computational Modeling

Xiaosi Gu

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Background: The insula has been associated with a wide range of functions including interoception, emotion, reward, risk, and cognitive control, making it difficult for researchers to reach a consensus about its actual function and thus, hindering our understanding of its role in psychopathology such as addiction. This is (at least partially) due to the lack of understanding of the insula at the algorithmic and computational levels.
Methods: In two studies, we investigated the algorithms implemented by the insula using computational modeling of behavioral and neuroimaging data in cigarette smokers. In Study 1, we used a Bayesian observe model and an olfactory cigarette cue exposure paradigm to quantify the computational mechanisms of cigarette craving. In Study 2, smokers were asked to perform a reward learning task in the scanner; we modeled their choice behavior using formal models of reinforcement learning and also measured their craving levels before and after scanning.
Results: In Study 1 ($n = 24$), we found that Bayesian craving prediction errors were encoded in the anterior insular cortex (AIC) as well as midbrain dopamine regions. Furthermore, AIC activation, but not midbrain activity, correlated with the number of cigarettes smoked in the previous 24 hours. In Study 2 ($n = 24$), we found that the AIC integrated both reinforcement learning and post-scanning craving signals in smokers.
Conclusions: Taken together, these results demonstrate that the AIC computes interoceptive information in a Bayesian

fashion; more crucially, the AIC also provides information about bodily states to exteroceptive (e.g. reward) decision-making processes. These results provide computational evidence supporting a key role of the AIC in interoceptive inference (i.e. the Bayesian inference of bodily states), and in translating interoceptive information into computational signals that guide choice behavior.
Disclosure: Nothing to Disclose.

12.3 Anterior Insula Projections Drive Compulsion-Like Alcohol Drinking

Frederic Hopf

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Background: Compulsion-like motivation is a critical contributor to harmful binge-patterned alcohol drinking and a central obstacle to successful treatment of Alcohol Use Disorder and its enormous social costs. We previously identified an anterior insular (aINS) circuit that specifically promotes compulsion-like alcohol drinking (CLAD), but has no role in "regular" (unpunished) alcohol consumption, in a rat model. In particular, CLAD requires aINS-NAcCore and mPFC-NAcCore inputs, as well as cortically activated non-canonical NMDARs in NAcCore. This agrees with ideas from clinical groups (Tiffany and Conklin, 2000; Bechara and Naqvi, 2010) that it is the conflict during intake (especially where intake persists despite negative consequences) which recruits specific cortical-subcortical circuits to successfully maintain compulsion-like behavior. In contrast, regular drinking does not involve conflict or need this circuitry.
Methods: Wistar rats drank 20% alcohol under intermittent access for ~3 months, then drank for 20 min/day 5 days/week. Compulsion-like drinking was determined by adulterating alcohol with quinine. Optogenetics and intracranial and systemic pharmacological inhibition were combined with lickometer analyses of patterns of responding during compulsion-like and regular alcohol intake.
Results: Since compulsion-like intake involves consumption despite conflict, which could be stressful, we examined whether CLAD would require the adaptive stress system, in particular the Locus Coeruleus (LC) and noradrenergic projections. Inhibition of alpha1-noradrenergic receptors with prazosin systemically or in the mPFC significantly and selectively reduced CLAD. In addition, aINS projections to the LC area also strongly mediated CLAD, with no role in regular alcohol or saccharin-quinine intake. Analysis of behavioral microstructure suggested that the animals were actively experiencing aversion during CLAD, with dramatic reductions in variability of responding suggesting a focus on more automaticity during compulsion-like intake.
Conclusions: Thus, adaptive stress responding may be a central part of the mechanism allowing the aINS to drive compulsion-like intake. Together, we seek to identify the central circuitry required for maintaining pathological intake, which could help develop better pharmacological and behavioral therapies to reduce the potent control that compulsion-like drives exert.
Disclosure: Nothing to Disclose.

12.4 Nicotine Abstinence Modulates Connectivity Between Insula Subdivisions and Network Nodes Related to Endogenous Processing

Elliot Stein

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Background: Smoking cessation attempts regularly fail, often within hours of abstinence, suggesting that the initial stages of a quit attempt is a key barrier to long-term abstinence. The insula is a node of integration between cognitive and affective processing, both of which are biased during abstinence. The insula plays a central role in adaptive cognitive control and the integration of visceral and homeostatic stimuli that influence salience detection. It interacts with constituent nodes of its own and other large-scale brain networks to influence attention orientation via a hypothesized tripartite network model, i.e. during interoceptive processing, the DMN is relatively more active while the ECN is relatively deactivated; exteroceptive processing leads to the reverse. As part of the Salience Network, the insula toggles between these networks. While enhanced insula-DMN connectivity is reported in acute abstinence, a downregulated insula-ECN connection has not, perhaps due to the functionally and structurally diverse nature of the insula.
Methods: Twenty healthy smoking cessation treatment-seeking participants signed NIDA-IRP IRB approved consents. Resting BOLD data were acquired during sated adlib smoking and after 48hr acute smoking abstinence. Withdrawal (WSWS) and cognitive (sustained attention) processing were assessed (RVIP) immediately prior to scanning. Functional connectivity strength differences between the 2 conditions were calculated using seeds from 3 insular subdivisions masked with 'targets' within the ECN, SN and DMN using dynamic (d-rsFC) and static rsFC (s-rsFC). We hypothesized increases in insula-DMN rsFC during abstinence in Post-Ins-DMN associated with interoceptive processing and decreases in Dorsal-Ins-ECN associated with attention processing.
Results: Abstinence caused a reduction in sustained attention, while 5 of 7 WSWS subscales showed significant subjective withdrawal. D-rsFC showed that the probability to remain in a given state ($p = .03$) increased in the abstinent condition. Four functional circuits showed significant effects of abstinence: rsFC increase between DOR-ins and inferior parietal lobule (within DMN); two circuits between the POS-ins and dorsal ACC and inferior frontal cortex, (both within SN), while a circuit between VEN-ins and the dlPFC (within ECN) decreased during abstinence. The VEN-ins-dlPFC circuit showed a negative correlation with the WSWS craving subscale ($p < .0001$), while POS-ins-dACC (SN) circuit showed a positive correlation with WSWS anger subscale when sated ($p < .001$).
Conclusions: Acute abstinence precipitates a less flexible network configuration, whereby connections from insula subdivisions show distinct patterns of modulation. As predicted by the tripartite model, divergent connections between insula subdivisions and both DMN and ECN were seen during abstinence. These divergent connections reflect an attention bias toward aversive affective processing and not away from exogenous cognitive processing. Thus, the insula does not modulate the exchange of an exogenous orientation during satiety for an endogenous orientation during

abstinence. Rather, abstinence modulates only connections between the insula and nodes more related to endogenous processing. Correlations between these oppositional changes in insula-seeded rsFC with subjective withdrawal ratings-but importantly not cognitive performance-are consistent with allostatic processes that promote continued drug abuse.
Disclosure: Nothing to Disclose.

Mini Panel
13. MicroRNAs in Clinical High Risk Subjects and Patients With Schizophrenia: Peripheral Blood and Cerebrospinal Fluid Analyses and Relationship to Cortical Development

13.1 Neuroimmunology and MicroRNAs

Clark Jeffries

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Background: Patterns in blood plasma analytes and leukocytic microRNAs distinguish patients presenting mental distress who convert to a diagnosis of schizophrenia within two years from other patients who do not convert. Several of the distinguishing analytes are prominent in the immune system (Perkins et al., Schiz Bull 2015). Likewise, certain leukocytic miRNAs can be used to predict risk of conversion (Jeffries et al., Transl Psychiatry 2016).
Methods: Integrating blood plasma analyte data with leukocytic miRNA data yields three distinct patterns of strong correlation networks for groups comprised of: unaffected comparison subjects, patients who converted to schizophrenia within two years, and patients who did not convert.
Results: All three types of networks (among blood analytes, among miRNAs, and between pairs of blood analytes and miRNAs may be analyzed with strict and rigorous attention to permutation tests, assuring that any resulting patterns are not explicable by chance.
Conclusions: Comparing the three types of networks results in patterns that distinguish them. Surprisingly, almost all strong protein v miRNA correlations are positive, contradicting the view that miRNAs inhibit protein expression. However, dynamical systems analysis explains this and moreover points to new understanding of miRNA functions. While miRNAs indeed cause downregulation of proteins in the short term, in homeostasis the proportions remain nearly constant. In fact, deviation from this rule is indicated in nonconverting subjects and all the more so in converting subjects.
Disclosure: Nothing to Disclose.

13.2 The Role of MicroRNA Expression in Cortical Development During Conversion to Psychosis

Amanda Zheutlin

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Background: An increased rate of cortical thinning has been observed in individuals at clinical high risk who convert to psychosis relative to non-converters and typically developing controls in the North American Prodromal Longitudinal Study (NAPLS). Levels of pro-inflammatory cytokines at baseline, which reflect increased systemic inflammation, were correlated with the rate of cortical thinning, suggesting this neurodevelopmental trajectory may be coordinated in part by increased inflammatory signaling. We investigated gene expression pathways within these cells to nominate cellular systems that may be important to cortical maturation during conversion to psychosis.
Methods: We used a forward stepwise regression algorithm to identify microRNA expression – hub regulators of mRNA – in peripheral leukocytes associated with annualized rate of reduction in gray matter in a subsample of NAPLS ($N = 74$).
Results: Nine microRNAs were included in a single classifier for cortical thinning, $p = 3.63$ x 10-08, $R2 = .358$, permutation-based $p = .039$, the mRNA targets of which were enriched for intracellular signaling pathways involved in coordinating inflammatory responses within immune cells ($p < .05$, Benjamini-Hochberg corrected). The classifier was also related to pro-inflammatory cytokine levels ($p = .038$).
Conclusions: MicroRNAs identified here have been previously linked to brain development, synaptic plasticity, immune function and/or schizophrenia, demonstrating some convergence across studies and methodologies. Perturbations in intracellular signaling pathways regulating inflammation may promote disease onset among those at high risk for psychosis via alterations in cortical maturation.
Disclosure: Nothing to Disclose.

13.3 MicroRNAs in Cerebrospinal Fluid and Plasma of Schizophrenia-Spectrum Disorder Patients and Healthy Volunteers

Juan Gallego

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Background: Genetic variants in miRNA genes and abnormalities in the concentration of microRNAs (miRNAs) in tissues and biological fluids have recently been associated with a diagnosis of schizophrenia. Most of these studies used post-mortem brain tissue or whole blood as the source of RNA. However, examination of microRNAs in cerebrospinal fluid (CSF) might provide an in vivo biomarker, more accurately reflecting expression level changes in the brain. To date, there are no studies that have investigated miRNA expression in CSF in patients with schizophrenia using small RNA-seq. In the past, our group had the opportunity of investigating the correlation between miRNA profiles in CSF and blood measured using microarray technology. Therefore, to expand our findings and use current cutting-edge technology, we measured miRNA profiles in CSF and plasma using small RNA-seq in a decently large sample of patients with schizophrenia-spectrum disorder (SSD) patients and healthy volunteers.
Methods: Twenty-two SSD patients and 17 healthy volunteers underwent a lumbar puncture and a blood draw. 15-25 cc of CSF and 5-10 cc of peripheral blood were obtained from each subject. CSF and peripheral blood samples were centrifuged. CSF and plasma samples were aliquoted into

1 mL cryovials, and stored at -80C degrees. Vesicular RNA was extracted from 1 mL of CSF and plasma samples following the protocol from the Qiagen exoRNA easy kit. The BioScientific NextFlex RNA sequencing kit was used for library construction. Sequencing was done on HiSeq2500. MiRNAs that were present in at least half of the schizophrenia patient samples or at least half of the control samples were included in the analysis. Samples that had at least 50,000 reads going to mature miRNA sequences were included. Differential expression analyses were conducted in R using the DESeq2 package in Bioconductor.

Results: In the overall sample cohort, subjects were male (66.7%), not Hispanic (79.5%) and black (48.7%). Mean age was 36.7 years (SD = 12.2), There were no differences in age, sex, ethnicity, race between diagnostic groups. In the patient group, 16 (72.2%) had schizophrenia, 5 (22.7%) had schizoaffective disorder and 1 (4.5%) had psychosis not otherwise specified. Differential expression (DE) analyses were conducted for 153 miRNAs in CSF and 510 miRNAs in plasma. After adjusting for multiple comparisons, DE analysis between patients and controls in CSF showed that miR-486-5 pm was significantly decreased in patients compared to controls (log2 Fold Change: -1.45, adjusted p-value: 0.02). Similar analysis in plasma showed that miR-942-5p (log2FC = -0.88, adj p-value: 0.02) and miR-204-5p (log2FC = -1.82, adj p-value: 0.02) were significantly lower in patients compared to controls. Principal component analysis showed a clear separation between CSF and peripheral blood samples. Out of 626 miRNAs used to examine the relationship between CSF and plasma, 167 (26.7%) were present only in CSF while 145 (23.2%) were present only in plasma.

Conclusions: miR-486-5p was downregulated in CSF of SSD patients compared to healthy volunteers whereas miR-942-5p and miR-204-5p were downregulated in plasma of SSD patients compared to healthy volunteers. MiRNA profiles in CSF and plasma have important quantitative and qualitative differences that may make them excellent, but different, candidate biofluids for biomarker discovery.

Disclosure: Nothing to Disclose.

Panel
14. Circuit and Synaptic Plasticity Mechanisms of Drug Relapse

14.1 Time-Dependent Changes in NMDA Receptor Transmission in the Nucleus Accumbens Core During the Incubation of Cocaine Craving

Marina Wolf

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Background: Cue-induced cocaine craving in rats intensifies ("incubates") during withdrawal from extended-access cocaine self-administration. After ~1 month of withdrawal, levels of GluA2-lacking, Ca2+-permeable AMPARs (CP-AMPARs) increase in the nucleus accumbens (NAc) core and thereafter their activation is required for expression of incubated craving. Little is known about whether CP-AMPAR plasticity in NAc core is accompanied by altered NMDAR transmission. However, the Lüscher lab has demonstrated that incorporation of GluN3- and GluN2B-containing NMDARs accompanies the increase in CP-AMPAR levels in the ventral tegmental area elicited by a single cocaine injection. In the incubation model, the Dong lab has demonstrated increased GluN2B-containing silent synapses shortly after discontinuing cocaine self-administration. Therefore, we were particularly interested in potential adaptations involving GluN2B- or GluN3-containing receptors.

Methods: We conducted whole-cell patch-clamp recordings in NAc medium spiny neurons (MSN) from rats that self-administered saline or cocaine. We measured evoked NMDAR-mediated synaptic responses across various membrane holding potentials (-80 to +40) and then characterized these responses using antagonists selective for GluN2B- or GluN3-containing receptors.

Results: As expected from prior studies, MSN from saline rats exhibited NMDAR currents sensitive to GluN2B- but not GluN3-selective antagonists. During the first week of withdrawal, we observed an increase in NMDAR currents at positive potentials in cocaine rats compared to saline controls that was attributable to NMDARs containing GluN2B but not GluN3. Between 2-3 weeks of withdrawal (still prior to detection of elevated CP-AMPAR levels), we observed increased NMDAR currents at both negative (-80 to -40mV) and positive (+40mV) holding potentials. The same results were obtained during a later period of withdrawal (>39 days) when cocaine craving is maximal. Pharmacological studies conducted in late withdrawal indicated that increased NMDAR currents reflected incorporation of NMDARs that contain both GluN2B and GluN3, as well as NMDARs containing GluN2B but not GluN3. Studies are in progress to determine if these sequential changes in NMDAR subunit composition are required for the elevation of CP-AMPAR levels and the incubation of cocaine craving.

Conclusions: Incubation of craving is accompanied by time-dependent changes in NMDAR subunit composition in MSN of the NAc core. The GluN3 plasticity observed at later withdrawal times is particularly significant as inclusion of this subunit results in atypical NMDARs that have very low Ca2+ permeability and low sensitivity to Mg2+ block. Together with the CP-AMPAR and group I mGluR plasticity that we have demonstrated previously, these changes will dramatically alter synaptic transmission and synaptic plasticity in NAc MSN.

Disclosure: Nothing to Disclose.

14.2 Cascades of Homeostatic Dysregulation Progressively Intensify Cocaine Seeking

Yan Dong

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Background: Homeostatic regulation and dysregulation after drug exposure have been long hypothesized as a key mechanism underlying the progression of the addictive state. However, how addiction-related homeostatic alterations are formed and evolve remains poorly understood. In

the nucleus accumbens (NAc), a brain region that has been critically implicated in the development and maintenance of addition-associated behaviors, the functional output of principle medium spiny neurons (MSNs) relies on the integration of the excitatory synaptic input and the membrane excitability.
Methods: Using a combination of slice electrophysiology, viral-mediated gene transfer, RNAi, and operant behavioral tests, this study characterizes a cascade of homeostatic dysregulation that contributes to the development of incubation of cocaine craving.
Results: Our results show that the excitatory synaptic input and membrane excitability of NAc MSNs are coordinated through a synapse-membrane homeostatic crosstalk (SMHC), through which an increase or decrease in the excitatory synaptic strength induced a homeostatic decrease or increase in the membrane excitability, and the vice versa, such that the overall functional output of NAc MSNs can be maintained relatively stable through internal and external disturbances. However, cocaine self-administration generated false SMHC signals at excitatory synapses, resulting in cascades of SMHC-based dysregulation to polarize the NAc function, while preventing the cascading of cocaine-induced homeostatic dysregulation in NAc MSNs prevented the progressive intensification of cocaine seeking after drug withdrawal.
Conclusions: Our results delineate a cellular mechanism through which cocaine-induced homeostatic dysregulation promotes drug relapse after withdrawal.
Disclosure: Nothing to Disclose.

14.3 Contribution of Cue-Elicited Glutamate Release Within Prefrontal Cortex to Cocaine-Craving

Karen Szumlinski

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Background: Drug-craving in response to drug-associated stimuli is associated with anomalous activity within prefrontal cortical regions governing volitional control over cognition and behavior. In both humans and laboratory rodents, the intensity of cue-elicited drug-craving intensifies with the passage of time in drug withdrawal and, at least in rodents, the manifestation of this "incubation of craving" is associated with cellular hyperactivity and glutamate release within the more ventromedial aspects of the prefrontal cortex (vmPFC). However, the functional relevance of excitatory neurotransmission within vmPFC subregions for incubated craving remains to be determined. Thus, this study determined the cause-effect relations between incubated cue-elicited glutamate release within the two major subdivisions of the vmPF – the infralimbic (IL) and prelimbic (PL) cortices – for cue-elicited drug-seeking and its incubation during protracted withdrawal.
Methods: To determine the functional relevance for endogenous glutamate within the PL versus IL in driving incubated drug-seeking, we employed a neuropharmacological strategy in which extracellular glutamate was raised and lowered, respectively, using the non-selective excitatory amino acid transporter inhibitor TBOA (300 μM) and the mGlu2/3 autoreceptor agonist LY379268 (20 mM). Adult male rats were fitted with intravenous catheters and stereotaxically implanted with bilateral guide cannulae for microinjection into the PL or IL. Rats were then trained to self-administer intravenous cocaine (0.25 mg/infusion) for 6 h/day over 10 days under an FR1 schedule of reinforcement (20-sec time-out) during which with each cocaine reinforcer delivery was paired with the presentation of a 20-sec tone-light stimulus complex. Rats were then subdivided into 3 or 30-day withdrawal groups, matching for cocaine experience. On their appropriate withdrawal day, rats were microinjected with vehicle, 50 μM APDC, or 300 μM TBOA (0.5 μl/min/side) into either the IL or PL, and then given a 30-minute cue-test in which responding resulted in the presentation of the tone-light stimulus previously paired with cocaine, but no cocaine delivery.
Results: As expected based on published work, vehicle-infused rats tested for cue-elicited drug-seeking at 30 days withdrawal exhibited incubated responding, relative to their counterparts tested at 3 days withdrawal. Surprisingly, infusion of neither drug into the IL or PL significantly influenced cue-elicited responding in rats tested at 3 days withdrawal. In contrast, an intra-IL TBOA infusion reduced, while intra-PL TBOA did not significantly affect, the elevated responding exhibited by rats tested at 30 days withdrawal. Oppositely, an intra-PL LY379268 infusion reduced, while intra-IL LY379268 did not affect, incubated responding.
Conclusions: The present results argue that endogenous glutamate within vmPFC is neither necessary nor sufficient to influence cue-elicited drug-seeking during early drug withdrawal in animals with a relatively extensive history of cocaine-taking. However, the observation that incubated drug-seeking can be blocked by either reducing or increasing endogenous glutamate within the PL and IL, respectively, bolsters the theory that the corticofugal projections from these two vmPFC subregions play opposite roles in regulating drug-seeking behavior in highly cocaine-experienced animals. Importantly, these data provide novel neuropharmacological evidence that the time-dependent or withdrawal-induced plasticity that occurs at glutamate terminals within the PFC are critical for regulating the manifestation of behavioral hyper-reactivity to drug-associated cues of relevance for relapse in addiction.
Disclosure: Nothing to Disclose.

14.4 The Anterior Insula-To-Central Amygdala Glutamatergic Pathway is Critical to Relapse After Contingency Management

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Background: We recently developed a rat model of relapse after choice-based voluntary abstinence that mimics human relapse after cessation of contingency management, a behavioral treatment that uses alternative non-drug rewards to maintain abstinence. Here, we studied the role of central amygdala (CeA) and its afferent projections in relapse after voluntary abstinence.
Methods: We trained rats to self-administer palatable food (6 d) and intravenous methamphetamine (14 d). We then

assessed relapse to methamphetamine seeking after 14 voluntary abstinence days (achieved via a discrete choice procedure between methamphetamine and palatable food).
Results: Relapse to methamphetamine seeking after voluntary abstinence was associated with increased expression of the activity marker Fos in CeA but not basolateral amygdala (BLA). Systemic injections of the dopamine Drd1-family receptor antagonist SCH39166 decreased relapse and CeA Fos expression; in situ hybridization showed higher co-labeling of Fos with Drd1 than with Drd2. CeA SCH39166 injections decreased relapse after voluntary abstinence; in contrast, BLA SCH39166 injections or CeA injections of the dopamine Drd2-family receptor antagonist raclopride were ineffective. Double-labeling of Fos with the retrograde tracer cholera toxin subunit-B (CTb, injected in CeA) demonstrated that relapse after voluntary abstinence was associated with selective activation of ventral anterior insula (AIV) → CeA projection. AIV inactivation with GABA receptor agonists or chemogenetic inactivation of AIV → CeA projection decreased relapse after voluntary abstinence. Electron microscopy data showed that AIV vGluT1-expressing projection-neurons form excitatory asymmetric synapses on CeA neurons.
Conclusions: Our data demonstrate a critical role of CeA Drd1 and the AIV → CeA glutamatergic projection in relapse after cessation of contingency management-induced voluntary abstinence.
Disclosure: Nothing to Disclose.

Study Group
15. The Role of Biomarkers in Drug Development for Psychotic Disorders

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Study Group Summary: Mechanistically innovative psychotropic drug development and devices for neuromodulation have been stalled by the lack of valid measures of target engagement and efficacy. Nuclear medical, MRI technology and quantitative electrophysiology offer promising strategies to facilitate this vital process and attract biotech and pharma to invest in treatment development for brain disorders affecting mental function and behavior. The task of biomarker development is made more challenging when they must be applied in the context of multi-center studies, and requiring durable and reliable methodologies and quality control. This study group will present leading edge work on promising methods and engage in the discussion of their applications at various stages of treatment development and FDA approval. It will also consider what evidence base will be necessary to establish for the FDA to accept biomarkers as valid measures of drug activity and efficacy.
Disclosure: Part 1: Alkermes, Grant, Clintara, Advisory Board, Intracellular Therapies, Board Member, Pierre Fabre, Advisory Board, Repligen, Patent, **Part 2:** Clintara, Advisory Board, **Part 4:** Alkermes, Grant.

Panel
16. Neuroimmunomodulation of Brain Glutamate in Neuropsychiatric Disorders

16.1 Defining Immune-Glutamate Dysregulation in Mood Disorders as a Clinically Relevant Biotype – Use of Multimodal Imaging Approaches

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Background: The primary goal of this presentation is to develop and validate a neural profile of inflammation-induced glutamate dysregulation as a distinct biotype of mood disorders. Previously, we had reported that 4-week administration of interferon-alpha to non-depressed, Hepatitis C infected individuals led to a severe depressive syndrome associated with significant increases in basal ganglia and anterior cingulate glutamate concentrations. Subsequently, using a different sample of medically healthy, depressed subjects we demonstrated similar associations between peripheral inflammation (indexed using plasma c-reactive protein or CRP) and basal ganglia glutamate, which was associated with the severity of psychomotor slowing and anhedonia. In this analysis, we extend our previous observations by examining the downstream consequences of inflammation-induced glutamate increases on neural activity and connectivity by employing a computationally driven multimodal imaging approach using parallel and sequential models that also tested the classificatory power of the models using clinically relevant metrics.
Methods: We used a model-agnostic, data-driven, hierarchical clustering algorithm that combined CRP levels with brain glutamate estimates [obtained using magnetic resonance spectroscopy] to classify a sample of 41 unmedicated depressed subjects into two groups with contrasting inflammatory and glutamate profiles (High CRP/Glu, $n = 22$ and Low CRP/Glu, $n = 19$). Functionally relevant, local brain activity signatures were computed using Regional Homogeneity (REHO) – a measure of network centrality and voxel-wise coherence - in the MRS voxel and anatomically defined striatal connectome. FreeSurfer-registered whole brain REHO maps were used to identify brain-wide REHO changes that discriminated between High and Low CRP/Glu groups and functional connectivity analysis was used study interactions between these nodes. The probability of achieving membership in High CRP/Glu group was estimated using Fisher's Linear Discriminant Function Analysis (DFA).
Results: Membership in the High CRP/Glu group was predicted by decreased REHO in the left basal ganglia region prescribed for MRS voxel ($p = 0.008$); by decreased REHO in the FreeSurfer-defined striatal, ventral medial prefrontal, superior frontal, insular, anterior and posterior cingulate regions (all p values < 0.001) from the whole-brain analysis; and decreased functional connectivity between ventral medial prefrontal and superior frontal areas ($p = 0.037$) from functional connectivity analysis. The classificatory power of the combined model was robust (sensitivity = 84%, specificity = 93%, accuracy = 88%, misclassification rate = 12.5%). Discriminant score loadings on High CRP/Glu significantly correlated with severity of anhedonia ($p = 0.023$), depression

($p = 0.022$) and motor slowing ($p = 0.032$) validating the clinically relevant classificatory power of the neural signatures.
Conclusions: We speculate that increases in extrasynaptic glutamate signaling induced by inflammation might result in decreased local signaling coherence (decreased REHO), impaired long distance connectivity and related behavioral symptoms. This association implies a clinically meaningful predictive value of these brain activity/connectivity biomarkers in classifying individuals with High CRP/Glu.
Disclosure: Nothing to Disclose.

16.2 Dysregulation of Glutamate Signaling at Cortico-Striatal Synapses in Huntington Disease

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Background: The inherited neurodegenerative disorder Huntington disease (HD) manifests in mid-life with frontal executive, motor and affective impairments. Aberrant signaling at glutamatergic cortico-striatal synapses precedes overt motor abnormalities and may contribute to prodromal cognitive impairments and increased vulnerability of striatal neurons to degeneration in mice models of HD. Previously, our lab showed increased expression and signaling of extrasynaptic NMDA-type glutamate receptors on striatal spiny projection neurons (SPNs) in premanifest HD, shifting the balance away from cell survival toward death pathways. Despite normal clearance of synaptically-released glutamate in HD striatum, these extrasynaptic NMDA receptors can still be activated during high frequency cortical afferent firing that promotes glutamate spill-over. In this regard, retrograde endocannabinoid (EC) signaling from striatal SPNs to presynaptic cortical cannabinoid receptor 1 (CB1), stimulated by high-frequency trains of cortical activity, mediates long-term depression (LTD) at these synapses, suppressing presynaptic glutamate release. Since a reduction in striatal CB1 expression has been reported in HD patients and mouse models, we investigated whether CB1-mediated modulation of cortical glutamate release onto striatal projection neurons is attenuated in the transgenic YAC128 HD mouse model.
Methods: Acute sagittal cortical-striatal brain slices were made from 1-2-month-old YAC128 and control mice (background strain, FVB/N). Field recordings of excitatory postsynaptic potentials (EPSPs) or whole-cell patch clamp recordings of excitatory postsynaptic currents (EPSCs) were made from neurons in dorsal striatum in response to stimulation of cortical afferents in the corpus callosum, as follows: High-frequency stimulation (HFS) – 100 Hz for 1 s, four times at 10 s intervals; or Depolarization-induced Suppression of Excitation (DSE), which paired cortical stimuli with postsynaptic depolarization to +30mV. CB1 agonists and antagonists were diluted >1000-fold from DMSO stock into aCSF with bovine serum albumin, and pre-incubated in the slice >30 min before recording.
Results: HFS-induced LTD associated with suppression of glutamate release via CB1 signaling, which relies on retrograde signaling by AEA, was markedly attenuated in YAC128 striatum, specifically at cortical synapses onto D2-expressing striatal SPNs. Strikingly, HFS-LTD could be restored by treatment with JZL184, an inhibitor of monoacylglycerol lipase that degrades 2-AG, thereby boosting levels of 2-AG, whereas an inhibitor of AEA degradation was ineffective.
Conclusions: HFS-LTD deficits at cortical synapses onto dorsal striatal spiny projection neurons observed in 2-month old YAC128 mice may contribute to early stage impairment of skilled motor learning, as well as increased vulnerability of D2 striatal neurons to degeneration as it is thought to be critical in protecting striatal neurons from excessive glutamate exposure by suppressing presynaptic release. Treatment with the compound JZL184 may improve cognitive changes and provide neuroprotection in early stage HD. Moreover, deficits in AEA have been linked to anxiety associated with chronic inflammation, an area of interest for future investigation in HD.
Disclosure: Nothing to Disclose.

16.3 Signaling Networks and Neuromodulation: Converging Pathways for Inflammation and Glutamate Uptake Mechanisms in Severe Mental Illness

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Background: Neuroimmune mechanisms have been implicated in the pathophysiology of schizophrenia. However, currently available biomarkers including plasma and PET-based tracers have yielded conflicting and contradictory results. Methodological difficulties in mapping immune biomarkers in the brain as opposed to other peripheral source such as plasma have contributed to this lack of clarity. New approaches to identifying neuroimmune changes in schizophrenia might be informed by postmortem studies. Intracellular cascades of kinases including AKT, nuclear factor(NF) kappa-B, janus kinases (JAK), and p38 MAP kinase form vital links in transducing and connecting immune signals with glutamate cycling and metabolism. We employed a kinome array platform to study alterations in immune signaling and glutamate signaling in postmortem brain from subjects with schizophrenia. We hypothesize that imbalances in kinase activity in brain cells of individuals with schizophrenia will propagate downstream through an interconnected network of intracellular signaling systems to amplify or impede neuroimmune and glutamate signaling in the brain that might result in the observed deficits in schizophrenia.
Methods: We established a bioinformatics workflow distinguishing schizophrenia-altered kinases in anterior cingulate cortex using a kinome array dataset. We compared schizophrenia-altered kinases to haloperidol-altered kinases, and identified systems, functions, and regulators predicted using pathway analyses. We used kinase inhibitors with the kinome array to test hypotheses about imbalance in signaling and conducted preliminary studies of kinase proteins, phosphoproteins, and activity for kinases of interest. We also performed parallel work in rodent models to confirm kinase nodes identified by our initial hypothesis generating work.
Results: We identified seven kinases over- or underrepresented in the anterior cingulate cortex in schizophrenia. Using these kinases as a starting point, we built an expanded

signaling network, identifying kinase "nodes" with the most interactions in schizophrenia. Using pathway analyses, we found that kinases associated with immune and glutamate signaling pathways were overrepresented. In haloperidol treated rats, we found alterations in signaling networks that contain immune and glutamate-regulating kinases, including ERK, JNK, DMPK and AKT, with expression profiles (decreased activity) in the opposite direction from our results in schizophrenia. Targeted confirmation studies using selective kinase inhibitors and individual kinase activity assays confirmed the suggested roles for AKT, ERK, and JNK in the schizophrenia signaling network. Divergence of these signaling network changes induced by antipsychotic treatment in rodents also provided a glimpse into immune signaling changes induced by treatment in schizophrenic patients.
Conclusions: Concurrent changes in the expression and signaling of kinases associated with immune and glutamate signaling indicate that subtle changes in kinase activity propagate along interlinked networks of psychopathological processes. It follows that the novel nodes identified in our model are putative targets to investigate the pathophysiology of inflammation and signaling defects in severe mental illness that may lead to development of novel biomarkers that reflect pathophysiological processes in schizophrenia more accurately.
Disclosure: Part 1: Allergan, Advisory Board, (Spouse), Jansen, Advisory Board, (Spouse).

16.4 Acute Treatment With the Glutamatergic Modulator Ketamine Regulates Adipose and Bone-Derived Immune Markers in Patients With Mood Disorders

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Background: Depression is associated with increased rates of obesity- and osteoporosis-related disorders in the context of an activated systemic inflammatory status. These phenotypic changes are often reflected by altered levels of peripheral immune biomarkers. This study examined the utility of several immune biomarkers as potential predictors of response to ketamine or as possible transducers of its therapeutic effects. Three key adipokines — adiponectin, resistin, and leptin — which play a key role in inflammatory response, energy metabolism, insulin sensitivity, and neuroplasticity, along with bone markers that play a key role in maintaining bone strength and modulating systemic inflammatory response - receptor activator of nuclear factor kappa-B ligand (RANKL, the principal osteoclastogenic factor), osteoprotegerin (OPG, a decoy receptor for RANKL) and osteopontin (OPN, which plays a significant role in bone strength) were studied before and after ketamine administration.
Methods: Pooled data from previous double-blind, randomized, placebo-controlled, crossover trials designed to test the antidepressant efficacy of acute ketamine infusion in individuals with major depressive disorder (MDD), bipolar disorder (BD), and healthy controls (HCs) was used. All subjects were medication-free, except for the BD patients who were maintained on therapeutic doses of lithium or valproate. All MDD and BD patients met DSM-IV criteria for a major depressive episode at the time of enrollment. Treatment-resistance was defined as a lack of current or past response to two or more adequate antidepressant or neuromodulatory trials. Subjects were randomized to receive a single subanesthetic ketamine infusion (0.5 mg/kg for 40 minutes). Plasma concentrations of adipokines and bone markers were measured at four time points: 60 minutes pre-ketamine infusion (baseline), 230 minutes post-ketamine infusion, on Day 1, and on Day 3 post-ketamine infusion. Linear mixed models with time as a fixed factor were used to examine changes over time. Bonferroni-adjusted posthoc tests were used to examine changes from baseline to each timepoint.
Results: Low plasma adiponectin levels predicted a positive acute antidepressant response to ketamine. In addition, lower adiponectin levels correlated with number of previous clinical depressive episodes, indirectly suggesting that non-response to current therapeutic options might have led to a protracted clinical course. In MDD subjects, acute ketamine administration restored OPG/RANKL ratio to levels seen in HCs, decreased RANKL levels and normalized the baseline decreases in OPN and OPN/RANKL. Bone inflammatory markers remained largely unaltered in HCs.
Conclusions: These results suggest that a single acute ketamine infusion not only improves depressive and cognitive symptoms, but may also correct critical abnormal peripheral markers associated with systemic illnesses in treatment-resistant subjects with mood disorders.
Disclosure: Nothing to Disclose.

Panel
17. Glutamate/GABA System Involvement in Depression and its Treatment

17.1 Increased Activity of Parvalbumin Interneurons in the Prefrontal Cortex Contributes to Anxiety- and Depressive-Like Behaviors in a Mouse Model of Stress-Related Mood Disorders

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Background: The prefrontal cortex (PFC) is highly sensitive to stress, a known risk factor for mood disorders including anxiety and depression. Hypoactivity of the PFC has been well described in humans displaying stress-induced depressive symptoms, and is now recognized to be a key feature of the depressed brain. However, little is known about the causes and mechanisms leading to altered PFC activity in the context of stress-related mood disorders. In addition, the contribution of this PFC hypoactivity to behavioral changes, specifically in females who have increased risk for stress-related mood disorders, remains poorly studied. Using a well-established mouse model of stress-related mood disorders, the unpredictable chronic mild stress (UCMS) paradigm, we previously observed that UCMS increases mRNA and protein expression of parvalbumin (PV) in the PFC. We now investigate whether this increased PV

expression reflects increased activity of PV interneurons (PV-I), and the extent to which this contributes to anxious and depressive phenotypes after stress.
Methods: First, adult male and female mice were exposed to UCMS for 4 weeks. Molecular approaches were used to determine (1) the extent to which UCMS leads to increased activity of PV-I in the PFC (measured by double immunofluorescence for PV and cFos) and (2) whether glutamatergic inputs onto prefrontal PV-I change as a function of UCMS (measured by immunofluorescent labelling of Vglut1 puncta onto PV-I). Then, a chemogenetic approach was used in PV:Cre mice to assess the impact of chronic activation of prefrontal PV-I, as observed during UCMS, on emotional behaviors (anxiety- and depressive-like behaviors). Finally, we used a prophylactic pharmacological approach with a single injection of ketamine prior to UCMS exposure to determine whether a stress-resilient phenotype induced by ketamine coincides with reduced levels of PV expression in the PFC.
Results: Exposure to UCMS increased the number of PV-I expressing cFos not only under resting conditions but also after exposure to an anxious stimulus, suggesting increased activity of this neuronal population. These changes are observed in both males and females, but are more predominant in the latter, and are associated with increased expression of VGlut1 puncta onto PV-I. In addition, chronic PV-I activation in the PFC of PV:Cre mice is sufficient to induce long-lasting increase in anxiety in a sex-specific manner, showing that increased PV-I activity, as observed following UCMS, contributes directly to changes in emotional behaviors. Finally, we observe that a prophylactic treatment with ketamine reduces PV expression in the PFC and prevents elevated emotional behaviors normally induced by UCMS, further demonstrating the link between high level of PV expression in the PFC after UCMS and increased emotional behaviors.
Conclusions: Together, we highlighted a novel mechanism by which increased GABAergic inhibitory prefrontal function, in the form of an overactive PV system resulting from exposure to chronic stress contributes causally to changes in emotional behaviors. This particular subtype of inhibitory interneurons could thus be targeted for potential therapeutic interventions against mood disorders. Furthermore, the heightened sensitivity to stress of the female prefrontal PV system when compared to that of males could explain their increased risk for stress-related mood disorders.
Disclosure: Nothing to Disclose.

17.2 Disinhibition With Negative Allosteric Modulators of GABA Receptors Containing Alpha 5 Subunits for Rapid Relief of Depression

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Background: There is an urgent need for antidepressants that provide rapid relief of symptoms with few side effects. Ketamine has engendered great excitement for its fast antidepressant actions, but it can induce several effects that render it poorly suited for wide-spread use in the treatment of depression. Based on evidence of excitatory circuit dysfunction in chronic stress models of anhedonia in rodents, we postulated that negative allosteric modulators of GABA receptors might exert ketamine-like rapid antidepressant actions and, if targeted to GABARs containing alpha 5 subunits, would exert few side effects.
Methods: We used male C57BL/6 J mice. Chronic multimodal stress, including restraint, white noise, and strobed light, daily for 4 hours over 14 days, was used to induce an anhedonic state. Other tests were performed using standard methods, as we have described elsewhere.
Results: Like ketamine, a single administration of the alpha5 selective GABA-NAM, MRK-016 (3 mg/kg) decreased immobility time in the forced swim test assayed 1 hr and 24 hrs after injection. Chronic stress induced an anhedonic state characterized by loss of sucrose preference and diminished interest in social interactions. A single administration of MRK-016 also restored sucrose preference, social interactions, and preference for sniffing female urine within 24 hrs, with effects persisting for up to 7 days. Unlike ketamine, there was no evidence of psychomotor activation in the open field test, motor impairment in the rotorod test, addiction liability in the condition place preference test, or psychotomimetic effects in the prepulse inhibition test. Electrophysiological, pharmacological, and behavioral evidence favoring a potentiation of excitatory synapses in the reward circuitry as a consequence of GABA-NAM induced disinhibition will be presented.
Conclusions: Negative allosteric modulators of GABARs that selectively target the alpha 5 subunit display a favorable combination of ketamine-like fast, persistent antidepressant and anti-anhedonic behavioral effects, with a side effect profile that appears far more favorable than ketamine. We conclude that this class of compounds has great potential as novel, fast-acting, effective, and clinically viable treatment for human depression.
Disclosure: Nothing to Disclose.

17.3 GLO1 Inhibitors Alter GABAergic Signaling and Exhibit Fast-Onset Antidepressant Properties

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Background: Current antidepressants exhibit slow onset, side effects, and limited efficacy. Thus, identification of novel fast-onset antidepressants represents a major unmet need. Glyoxylase 1 (GLO1) is a ubiquitous cellular enzyme responsible for the detoxification of the glycolytic by product methylglyoxal (MG). We previously reported that MG is a competitive partial agonist at GABA-A receptors.
Methods: We assessed the effects of both genetic and pharmacological inhibition of GLO1 in two antidepressant assays: the tail suspension test (TST) and the forced swim test (FST). Then, we examined the effects of short-term treatment with GLO1 inhibitors in three well validated models of antidepressant onset: the chronic FST (cFST), chronic mild stress (CMS) paradigm, and olfactory bulbectomy (OBX) paradigm. Finally, we determined the effects of short-term GLO1 inhibitor treatment on cyclic-AMP

response-binding protein (CREB) activation and brain-derived neurotrophic factor (BDNF) induction in the medial prefrontal cortex (mPFC) and hippocampus.
Results: Genetic knockdown of GLO1 or pharmacological inhibition using two structurally distinct GLO1 inhibitors, S-bromobenzylglutathione cyclopentyl diester (pBBG), or methyl-gerfelin (MeGFN), reduced immobility in the TST and FST assay models. Both GLO1 inhibitors also reduced immobility in the cFST after 5 days of treatment. Furthermore, 5 days of treatment with either GLO1 inhibitor blocked the depression-like effects induced by CMS on the FST and coat state, and attenuated OBX-induced locomotor hyperactivity. Finally, 5 days of treatment with pBBG induced BDNF and phosphorylated CREB in the hippocampus and mPFC. Conversely, 5 days of fluoxetine treatment did not produce these behavioral or molecular effects.
Conclusions: Our findings suggest that GLO1 inhibitors may provide a novel and fast-acting GABAergic pharmacotherapy for treating depression.
Disclosure: Nothing to Disclose.

17.4 Multimodal GABA and Default Mode Network Antidepressant Mechanisms of TMS

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Background: The antidepressant mechanisms of TMS are poorly understood but likely involve plasticity of the default mode network (DMN), which shows elevated functional connectivity during a depressive episode. Additionally, GABA levels are dysregulated during depression and normalize with antidepressant treatment, including SSRIs and electroconvulsive therapy. TMS targeting the left Dorsolateral Prefrontal Cortex (DLPFC) increases GABA levels in the Ventromedial Prefrontal Cortex (VMPFC) in individuals currently in a major depressive episode. Here, we perform an exploratory analysis whether subjects who had increases in GABA in the VMPFC from baseline to post-TMS were more likely to have reductions in functional connectivity between VMPFC and other DMN structures.
Methods: We treated 26 currently depressed, antidepressant-resistant patients with 25 daily treatments of 10 Hz TMS over a left DLPFC stimulation site and measured symptom severity before and after the course of treatment with the HAMD-24. MR-Spectroscopy and resting state fMRI scans were acquired within 3-days prior to beginning and 3-days after completing TMS treatment. We measured GABA/W levels in a single bilateral voxel covering the VMPFC as ratios to the voxel water (W) signal using the standard J-editing MRS technique on a 3T-GE scanner. Pairwise functional connectivity was measured between 58 nodes comprising the DMN subset of a 277-node atlas of brain structures.
Results: The mean reduction in HAMD-24 after TMS was 33.6% (± 25.7%). Mean VMPFC GABA level was higher post-TMS than at baseline ($2.99 \times 10\text{-}3 \pm 0.60 \times 10\text{-}3$ vs. $2.66 \times 10\text{-}3 \pm 0.62 \times 10\text{-}3$, mean change in GABA 12.5%, paired $t25 = 2.21$, $p = 0.034$). In precisely two pairs of nodes, GABA level was significantly correlated with functional connectivity both at baseline and post-TMS. Functional connectivity of pair #1, in the right VMPFC (MNI [4935–12]) and left VMPFC (MNI [-3 44 -9]), was positively correlated with GABA level at baseline ($r = .404$, $p = .041$), while the functional connectivity between these same nodes inversely correlated with GABA after TMS ($r = -0.397$, $p = .045$). A similar change in sign of correlation from positive to negative was observed between GABA level and functional connectivity of pair #2, the left VMPFC (MNI [-7 51 -1]) and the right angular gyrus (MNI [47–5029]) (baseline: $r = .399$, $p = 0.043$; post-TMS: $r = -.400$, $p = 0.041$).
Conclusions: Open label TMS over the left DLPFC increases GABA levels in the VMPFC. Further, GABA increases in the VMPFC were associated with a change in directionality of the correlation between GABA level and functional connectivity between pairs of DMN structures involving the VMPFC. This suggests that inhibitory-excitatory balance may regulate functional connectivity of large scale neural networks and that changes in both may contribute to the antidepressant mechanism of TMS.
Disclosure: Nothing to Disclose.

Mini Panel
18. Psychiatric Neurosurgery for Severe, Intractable Obsessive-Compulsive Disorder: Towards a Precision Medicine Approach

18.1 Analysis of Lesion Size and Location in Gamma Ventral Capsulotomy for Severe, Intractable OCD

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Background: Obsessive-compulsive disorder (OCD) affects two percent of the world population and is associated with major impairment and suffering. The World Health Organization ranks OCD as one of the 10 most disabling conditions by lost income and decreased quality of life. It is estimated that OCD is treatment refractory in over 20% of cases. Those individuals have chronic, severe, and disabling symptoms, despite all available conventional and augmenting treatments. For a subgroup of such OCD patients neurosurgery is a recognized therapeutic option. The objective of this study was to examine lesion characteristics in these patients in order to improve our understanding of neurocircuitry important for clinical improvement.
Methods: T1 MRI scans were taken from 26 OCD patients who had received GKC for severe, intractable OCD at either Brown University ($n = 12$) or the University of São Paulo ($n = 14$). Patients were assessed using the Yale-Brown Obsessive-Compulsive Scale (Y-BOCS) at baseline, and at clinical follow-up appointments. YBOCS was tracked as percent reduction from pre-operative levels. Pearson's correlation was used to examine the relationship between lesion volume and percent Y-BOCS reduction, covarying for post-operative time. Voxel-wise associations were tested between lesion location and percent Y-BOCS reduction. Tractography was performed using a Human Connectome Project diffusion atlas, using voxels associated with Y-BOCS reduction as a seed.

Results: Twenty-one of the 26 participants (80.8%) were full responders. The mean percent Y-BOCS reduction at follow-up relative to baseline was 52.1%, with reductions ranging from 0% to 96.7%. No significant association was found between Y-BOCS reduction and lesion volume ($p > 0.89$; $r = 0.02$). Lesions were primarily found in the anterior section of the anterior limb of the internal capsule, but they also extended into adjoining structures, including the caudate, putamen, and nucleus accumbens. In the permutation testing, a set of 128 voxels, located primarily in the anterior and medial aspect of the anterior limb of the right and left internal capsule, demonstrated a significant relationship between lesion and greater percent Y-BOCS reduction (corrected $p < 0.05$). Tractographic analysis suggested that these voxels involve frontal-thalamic connections.
Conclusions: This is one of the largest, as yet unpublished, neuroimaging studies of psychiatric neurosurgery for severe OCD. Results indicate placement in the anterior medial aspect of the internal capsule as most relevant for clinical improvement, though there was significant variability across patients. Further analysis of this individual variability, particularly with regard to structural connectivity, will be essential for improving targeting and clinical outcomes.
Disclosure: Nothing to Disclose.

18.2 Tractographic Analysis of the Anterior Limb of the Internal Capsule: Assessing Individual Variability in Connectivity

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Background: The anterior limb of internal capsule (ALIC) is a promising target for neurosurgical interventions for severe, treatment refractory OCD, including deep brain stimulation and anterior capsulotomy. However, there is controversy about optimal targets within the ALIC, as different locations appear to have differential efficacy in modulating relevant neural networks. Using diffusion tensor imaging (DTI) from a large national imaging database, we parcellated the ALIC based on frontal structural connectivity. We then compared the ALIC segmentations between individuals to evaluate regional patterns of anatomical variability.
Methods: ALIC segmentations were generated for 40 subjects from the Human Connectome Project (HCP) using connectivity-based seed classification in FSL. Voxels within the ALIC were treated as seed regions and frontal Brodmann areas (BAs) as independent targets. We combined individual-specific segmentations by assigning each ALIC voxel to the most frequently associated frontal BA in the 40 individual segmentations. We compared this combined individual-based parcellation to one similarly created using a template from HCP that averaged diffusion data from 842 subjects. Segmentations were compared to one another using the Sørensen-Dice Index of similarity (SDI).
Results: All 40 segmentations exhibited a posterior-superior to anterior-inferior axis of organization. On average, the frontal BA assignments of voxels in the group analysis were consistent with 66.2% of individuals' segmentations. In this analysis, the region assigned to BA11 (orbitofrontal cortex, OFC), exhibited the highest degree of consistency across individuals, with 12.1% of this region being assigned to BA11 in all 40 subjects. The mean SDI between the individual-based combined segmentation and the template-based segmentation regions was 0.28. The mean SDI between individual segmentation regions was 0.45. Regions assigned to BA11 were the most similar across individual segmentations, with a mean SDI of 0.714.
Conclusions: These results clarify the organization of the ALIC in humans. They also demonstrate the high variability in ALIC organization between individuals, with only a few loci of focal consistency. This high degree of variability suggests that patient-specific tractography is required to optimize target localization. Interestingly, the most consistent regions of the ALIC, those connecting to OFC, are the regions most frequently targeted by neuromodulatory procedures. Future "precision medicine" approaches will therefore be critical for planning neurosurgical procedures for refractory OCD.
Disclosure: Nothing to Disclose.

18.3 Adaptive Deep Brain Stimulation for Intractable Obsessive-Compulsive Disorder

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Background: Although Ventral Striatum (VS) DBS has shown promise for intractable cases of OCD, there is room for improvement in both clinical benefits and reduction of DBS-induced behavioral side effects, especially hypomania. In contrast to current devices, new generation adaptive DBS (aDBS) systems can record, stimulate and use signals (e.g., Local Field Potentials) from the brain to make responsive adjustments to the patient's behavioral state. We were recently awarded a grant from NIH BRAIN to develop aDBS for OCD. One of the challenges was to identify a label for classifiers that is objective, reliable and fits NIMH RDoCs constructs.
Methods: We applied an Automated Facial Affect Recognition (AFAR) developed by Jeff Cohn, PhD, to measure changes in facial muscle Action Units associated with different emotional expressions. For the purposes of this study, we concentrated on the ability of bilateral ventral striatum DBS to induce positive and negative valence affect.
Results: Five subjects undergoing bilateral ventral striatum DBS for OCD were videotaped intra-operatively during behavioral testing of various monopolar settings. AFAR showed high correlation with Likert scale ratings of mood and affect. AFAR was superior to clinician ratings in tracking moment-to-moment changes in valence and intensity of affective changes and showed a dose-response relationship with stimulation intensity.
Conclusions: Automated Facial Affect Recognition shows promise as a tool to provide a label for positive and negative valence classifiers induced by VS DBS in subjects with OCD. It provides an objective measure of emotional state that can be time-locked with other inputs such as LFPs, EEG, physiology and motion that can be used to build classifiers needed for developing aDBS.
Disclosure: Part 1: NIH, Grant, **Part 2:** Simons Foundation, Grant, **Part 3:** Medtronic, Grant.

Panel
19. Is Theta the New Gamma? Implications of low Frequency Oscillations for Altered Excitation/Inhibition (E/I) Balance in Schizophrenia (SZ)

19.1 Circuit Mechanisms of Hippocampal-Prefrontal Synchrony in Working Memory

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Background: The hippocampus and prefrontal cortex synchronize during working memory, mediated at least in part by direct hippocampal-prefrontal inputs. This synchrony occurs in multiple frequency bands, including theta (8-12 Hz) and gamma (40-70 Hz) frequencies. The circuit elements that subserve synchrony in these frequency ranges are thus far unclear.
Methods: We use a combination of optogenetic inhibition and multi-site, multi-electrode electrophysiological recordings in mice to examine the requirement for specific projections into and cell types within the medial prefrontal cortex (mPFC) in the generation of local oscillations as well as synchrony between the mPFC and its input structures. AAVs supporting the expression of eArch3.0 in a cre-dependent (for specific cell types) or non-cre-dependent (for projections) manner are used to induce expression. Optrodes combining fiberoptics and multiwire electrodes are used to record neural activity during performance of a T-maze delayed non-match to sample test of spatial working memory, in the presence and absence of optogenetic inhibition. C57Bl/6 strains are used throughout, and all experiments are done blind to condition.
Results: Inhibition of ventral hippocampus (vHPC) direct inputs to the mPFC decrease gamma-, but not theta-frequency synchrony between the vHPC and mPFC without affecting the strength of oscillations within either region; this inhibition impairs t-maze performance in a phase-specific manner. Inhibition of somatostatin-positive, but not parvalbumin-positive interneurons in the mPFC also disrupt synchrony and working memory behavior.
Conclusions: These data demonstrate a crucial role for vHPC inputs and parvalbumin-positive interneurons in the generation and/or maintenance of oscillatory synchrony between the mPFC and the vHPC during working memory behavior. Full characterization of the circuit components required for this synchrony may yield crucial information about how oscillatory synchrony can be modulated in vivo, with potential therapeutic benefits.
Disclosure: Nothing to Disclose.

19.2 Theta Phase Resetting Disruptions in Early Illness Schizophrenia and Youth at Clinical High Risk for Schizophrenia: Implications for the Excitation/Inhibition Balance

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Background: Across the animal kingdom, a basic mechanism allows all species to distinguish between sensations coming from others (e.g., pressure on a nematode's head from an approaching predator) and sensations resulting from self (e.g., pressure on its head from swimming forward). It is referred to as corollary discharge, tags sensations as "self," and minimizes the resources needed to process the sensations. Vocalization studies in primates show that responses in auditory cortex are dampened during vocalizing compared to passive listening, reflecting an excitation/inhibition (E/I) balance important for survival. In humans, it is seen in EEG-based N1 event-related potentials (ERP), which come from auditory cortex. Furthermore, this balance is likely driven by long-range synchrony of theta band activity between frontal and temporal lobes in healthy people. Not only is the E/I balance disrupted in people with schizophrenia and bipolar disease, but so is long-range theta synchrony. To extend our understanding of this, we analyzed EEG data in the time-frequency domain from people at clinical high-risk (CHR) for psychosis, schizophrenia patients early in their illness (ESZ), and healthy controls (HC).
Methods: EEG data from 102 HC, 84 ESZ, and 71 CHR were acquired as they said "ah" (Talk) and then listened to a recording of the sounds (Listen). N100 was the most negative peak occurring ~100ms after sound onset in both Talk and Listen conditions. A principal components analysis was used to extract a theta-band inter-trial coherence (ITC) factor, with a sweet spot that included 5-7 Hz activity in the N100 latency range.
Results: N1 amplitude and theta ITC suppression were modestly correlated and shared less than 20% variance. In HC, there was a significant condition effect, but it was greater for ITC (Cohen's $d = 1.45$) than for N100 (Cohen's $d = 0.63$), suggesting that ITC outperforms N100 in reflecting the E/I balance. While N1 suppression was not related to symptoms in ESZ, deficits in ITC suppression was correlated with delusions ($p = .01$). In CHR, N1 suppression deficits were correlated with unusual thought content ($p < .001$), and ITC suppression deficits were correlated with deficits in function ($p = .002$).
Conclusions: Theta phase oscillations reset at the onset of each sound during listening but less during talking. Similarly, N1 is elicited at the onset of each sound during listening but less during talking. This E/I balance may result when a corollary discharge of the expected sound cancels or dampens the actual sound. That theta ITC outperforms N1 suggests the mechanism underlying N1 suppression may be theta modulation, and deficits in the E/I balance in SZ may reflect failures of this modulation. Deficits in circuits that generate low frequency oscillations such as theta may be an important component of schizophrenia.
Disclosure: I (or my spouse/partner) do have financial interests to disclose.
Part 1: Boehringer-Ingelheim, Consultant, Spouse, Takeda, Consultant, Spouse, Alkermes, Consultant, Spouse, Upsher Smith, Consultant, Spouse.

19.3 Theta-Band Dysfunction as a Unifying Theme Across Sensory and Cognitive Modalities in Schizophrenia

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Background: Schizophrenia (Sz) is associated with deficits in sensory and cognitive function that represent a core feature of the disorder. In the auditory system, deficits in mismatch negativity (MMN) generation are among the best established biomarkers, and are linked to impaired role function in both prodromal and established Sz. Recently, we have demonstrated the underlying neural signature of MMN deficits, permitting assessment in translational rodent models. In the visual system, deficits in early potentials such as visual P1 component have been extensively replicated and correlate with subsequent attention-dependent cognitive impairments. Both auditory and visual deficits map extensively to impaired theta-band oscillatory activity, consistent with generalized theta-band impairment in Sz, reflecting local I/E dysregulation. This presentation focuses on translation and back-translation of these findings across human and rodent models.
Methods: Findings will be presented from rodent and human datasets. In rodents, auditory MMN was obtained during chronic treatment with the N-methyl-D-aspartate receptor (NMDAR) antagonist phencyclidine (PCP) with or without concurrent treatment with the NMDAR co-agonist glycine. Neurophysiological activity was analyzed using time-frequency approaches to isolate underlying neuro-oscillatory activity, and response profiles were observed to those recently obtained in humans with a convergent paradigm. In humans, visual ERP were obtained during both passive visual stimulation and a feature-attention/conjunction task, permitting parallel assessment of low (theta/delta) and high (gamma) frequency activity, and their relative contributions to cognitive performance.
Results: In rodents, auditory MMN mapped primarily to the theta-frequency band, consistent with recent findings in humans. Chronic treatment with PCP (15 mg/kg/d) led to significant impairment in duration MMN generation and associated theta band activity. Concurrent glycine treatment (16% by weight) significantly prevented these deficits, supporting a critical role for NMDAR in modulation of theta-band dysfunction in Sz. In the human visual system, passively presented stimuli elicit an initial P1 response that (like auditory MMN) maps primarily to the theta-frequency range. Stimulus-driven theta activity is impaired specifically to magnocellular biased stimuli. Stimuli presented during an active visual (feature detection) task elicit both attention-independent early theta and later, attention-dependent gamma activity. Path analysis suggests significant modulation of gamma activity by early theta, and significant modulation of task activity by late gamma, suggesting hierarchical contributions to cognitive impairments in Sz.
Conclusions: This presentation supports theta oscillatory deficits as key contributors to impaired cortical information processing in Sz across sensory modalities. The rodent findings demonstrate links to underlying NMDAR dysfunction and support SOM-interneuron contributions to impaired E/I interaction in Sz. Visual findings suggest impaired oscillatory hierarchical theta/gamma coupling within sensory and cognitive brain regions, consistent with impaired SOM/PV interactions across cortical regions.
Disclosure: Part 1: Glytech, Stock / Equity, Amino Acid Solutions, Stock / Equity, NeuroRx, Stock / Equity, Takeda, Consultant, Lundbeck, Consultant, Autifony, Consultant, FORUM, Consultant, Pfizer, Honoraria, **Part 2:** Glytech, Stock / Equity, **Part 3:** glytech, Stock / Equity, Self

19.4 Inhibitory Control of Mismatch Negativity (MMN) in a Genetic Mouse Model of Schizophrenia

Jordan Hamm

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Background: Sensorineural processing dysfunction in schizophrenia (SZ) undermines how affected individuals perceive and relate to a changing environment. Reduced mismatch negativity (MMN), typically recorded with EEG during auditory or visual "oddball" paradigms, is among the best replicated biomarkers of this aspect of the disease. Since SZ involves cortex-wide alterations to microcircuitry and local interneurons, it is likely that the MMN biomarker reflects a circuit-level pathophysiology. Translational research in mice combined with state of the art genetic and optical technologies provides a promising framework for studying the fine components of MMN with cell- and circuit-level precision.
Methods: Here we examined sensory cortical activity in awake mice, both wild-type (WT) and Df(16)A+/- mutant mice modeling the human 22q11.2 microdeletion, a highly penetrant risk genotype for schizophrenia. Mice were presented with various visual and auditory "oddball" stimulation paradigms based on human EEG studies, and responses to "redundant" (i.e. repetitive standard) stimuli were compared to responses to deviant (i.e. contextually novel) stimuli. Local field potentials (LFP) were recorded with 16-channel electrode arrays spanning layers 1-5 to quantify voltage, current-source density, and time-frequency measures of MMN. Fast two-photon calcium imaging (GCaMP6s; 30 Hz) was used to measure spiking of local V1/A1 neuronal populations with single neuron resolution and top-down inputs in V1 from mouse PFC with single axon resolution. Inhibitory circuit mechanisms were investigated via chemicogenetic suppression (D.R.E.A.D.D.s) of somatostatin- (SOM) and parvalbumin-containing (PV) interneurons.
Results: At the cell and circuit-level, two components of MMN were differentiated: stimulus specific adaptation (SSA; reduced LFP, gamma-band, and neuronal responses to "redundant" stimuli) and genuine deviance detection (DD; enhanced LFP, theta-band, and neuronal responses to "deviant" stimuli). Deviance detection, but not SSA, was selectively reduced in Df(16)A+/- mutants at the single neuron level. In WT, top-down inputs to V1 from PFC showed significant DD-related dynamics, but occurred at a later latency than V1 deviance detection. Selectively suppressing local somatostatin-containing interneurons (SOMs) with pharmacogenetics (D.R.E.A.D.D.s) dramatically eliminated the DD component across all timepoints and impaired theta/alpha-band oscillatory dynamics selectively, but left SSA and related gamma-band dynamics intact. Finally, in Df(16)A+/- mice, a potentially more fundamental deficit in reliable coactivity of local cortical ensembles was pervasive across paradigms (rest, passive stimulation, and oddball) yet was not fully recreated by acute manipulations in WT.

Conclusions: Sensory cortices of awake mice provide a rich model for understanding and relating human EEG biomarkers of deficient novelty processing to the underlying neuropathophysiology of schizophrenia. Here we link a translatable biomarker, the MMN and related theta/alpha oscillations, to the function of somatostatin-containing interneurons, a suspected cell-level pathophysiology present in some patients. Results also underscore the importance of top down inputs from prefrontal regions and the integrity of local neuronal ensembles. This set of findings in sum provide a promising roadmap for designing interventions to ameliorate electrophysiological and behavioral indices of sensory-cognitive deficits in psychiatric disease.
Disclosure: Nothing to Disclose.

Panel
20. Dynamic RNA Signaling in Substance Abuse

20.1 Modulation of Choroid Plexus Function and Altered Circulating MicroRNA Expression With Nicotine Self-Administration

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Background: Circulating microvesicles in the cerebrospinal fluid (CSF) have been recently identified to contain a variety of signaling factors, such as proteins, enzymes, and RNA species. While microvesicles, or exosomes, have been implicated in a few pathological conditions, such as with Alzheimer's disease and cancer, their importance in other aspects of physiological function is unknown. Interestingly, nicotine has been proposed to modulate choroid plexus function, and thus, nicotine-mediated release of factors into the CSF may underlie the drug's effects in the brain. In the current studies, we sought to determine whether nicotine acts directly on the choroid plexus and whether chronic nicotine self-administration would alter the expression of choroid plexus-derived circulating miRNAs.
Methods: In the first series of studies, choline acetyltransferase (ChAT) and nicotinic acetylcholine receptor (nAChR) subunit mRNA expression were examined in the choroid plexus at baseline conditions. Next, rats underwent chronic intravenous nicotine or saline self-administration, and choroid plexus tissue and CSF were assessed for changes in miRNAs and mRNA expression of nAChR subunits and the choroid plexus-specific protein transthyretin. Since differences were found in miRNA expression, we then performed studies to determine whether the circulating factors were localized in exosomes.
Results: First, we documented expression of ChAT and nAChR subunits in the lateral, dorsal third and fourth ventricle choroid plexus sites, indicating that endogenous cholinergic signaling mechanisms regulate the function of choroid plexus ependymal cells. Transthyretin expression was selectively increased in choroid plexus derived from the dorsal third ventricle, but not in tissue from the lateral or forth ventricles. Given the localization of the dorsal third ventricle adjacent to the habenula, these data suggest that nicotine can selectively alter signaling factors released into the CSF for potential integration into the habenula. Finally, miRNAs were found to be differentially expressed in the choroid plexus and exosome fractions of the CSF with chronic nicotine self-administration.
Conclusions: In conclusion, we provide evidence that nicotine acts directly on the choroid plexus to modulate the release of factors, including miRNAs, into the CSF. These data support the hypothesis that nicotine alters extracellular transfer of miRNAs, which could potentially lead to downstream changes in neuronal gene expression. In addition to nicotine dependence, findings from these studies may provide insight into normal physiological function and other pathological disease states.
Supported by the National Institute on Drug Abuse (NIH DA039658 to CDF)
Disclosure: Nothing to Disclose.

20.2 Coding/Non-Coding Pairs in the Neuregulin 3 Signaling Pathway and Nicotine Dependence

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Background: Smoking is the largest preventable cause of death and disease in the United States, with about 46 million U.S. adults currently smoking. Failed smoking cessation has genetic contributions; however, the majority of genetic polymorphisms associated with smoking cessation are located in intergenic or intronic regions, making it difficult to assign functional effects. Both nonsynonymous SNPs, which alter protein coding, and synonymous SNPs share similar likelihood and effect size for disease association. This indicates that while synonymous SNPs do not directly impact protein coding, they exert distinct measurable effects on health outcomes. For example, our lab has been examining the mechanistic effects of a recently implicated gene, Neuregulin 3 (NRG3), in nicotine dependence. NRG3 is a member of the EGF family that signals selectively through the receptor tyrosine kinase, ErbB4. We have previously published results demonstrating significant association of multiple single-nucleotide polymorphisms (SNPs) across the NRG3 gene with smoking cessation outcomes in two independent cohorts of smokers. Yet how these synonymous SNPs could result in altered NRG3 expression is not known. Recent studies have implicated a family of non-coding RNAs, long non-coding RNA (lncRNA), in the molecular etiology of addiction. Therefore, one potential consequence of these NRG3 SNPs may be altered regulation of its anti-sense long non-coding RNA partner, Nrg3os.
Methods: Mouse hippocampal primary neurons, mouse ex vivo hippocampal slices, and hippocampal samples from animals treated in vivo were assessed for changes in transcripts of NRG3 and its long non-coding paired transcript, Nrg3os, following nicotine treatment using ddPCR and dual single molecule FISH/IF.
Results: Bioinformatic analysis utilizing lncRNASNP, a database providing comprehensive resources of SNPs in human/mouse lncRNAs and their potential functional effects, suggests that the nicotine dependence-associated NRG3 SNPs we identified have the potential to alter the

antisense lncRNA NRG3as1. Additionally, our preliminary data demonstrates that expression of the homologous mouse lncRNA, NRG3os, is significantly reduced in a mouse model of nicotine dependence and withdrawal, which coincides with a corresponding increase in CREB-mediated NRG3 expression.
Conclusions: These data show that nicotine elicits increased CREB-mediated expression of NRG3 and results in a down-regulation of its anti-sense non-coding transcript, Nrg3os. These data support a model where synonymous SNPs in NRG3 linked to nicotine dependence may result in altered Nrg3os expression, thereby increasing the expression of the NRG3 transcript and its translation, thereby resulting in exacerbated nicotine withdrawal symptoms. To further evaluate these possibilities, current studies are evaluating the functional consequences of these variants on NRG3/Nrg3os expression in vitro and in vivo.
Disclosure: Nothing to Disclose.

20.3 HuD Regulation of Circular RNAs Expression and Localization During Cocaine Seeking Behaviors

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Background: Substance use disorder (SUD) is a chronic disease characterized by compulsive drug seeking and use. SUD is thought to be mediated by aberrant expression of proteins regulating synaptic plasticity in neurons. The main post-transcriptional mechanisms controlling gene expression at synapses involve the function of RNA-binding proteins (RBPs). Several RBPs have been previously associated with addiction including neuronal HuD. Specifically, HuD regulates the expression of many genes listed in the Knowledgebase of Addiction related genes (KARG) database such as c-FOS, BDNF, GAP43, and CAMK2A. At the molecular level HuD binds to cis-acting elements in RNA molecules governing their processing, subcellular localization, translation and degradation. One such group of RNA molecules that could be regulated by HuD are circular RNAs (circRNAs), a novel category of non-coding RNAs that are derived from the back-splicing and covalent joining of exons. Due to their abundant expression in the brain and their localization at synapses, circRNAs could be of importance to neuronal development and neuroplasticity. Notably, circRNAs contain numerous consensus regulatory sequences for RBPs including HuD. Thus, we aimed to explore whether HuD interaction with circRNAs may be involved in the control of synaptic plasticity during drug addiction.
Methods: For these studies, we used a combination of state-of-the-art methodologies including RNA-immunoprecipitation (RIP) assays, cirRNA arrays, synaptoneurosome RNA isolation and qRT-PCR validations along with bioinformatics algorithms for RBP binding site analyses developed in our laboratory.
Results: Bioinformatics analyses revealed that 29% of mouse circRNAs bear consensus HuD binding motifs. A subset of circRNAs showed altered expression in the nucleus accumbens (NAc) during cocaine-conditioned place preference (CPP). Among these, we decided to focus on those cirRNAs derived from host genes associated with synaptic plasticity. Using RIP assays followed by qRT-PCR, we validated the interaction of HuD with several cirRNAs. To assess whether HuD binding could alter target circRNAs localization, we also evaluated circRNAs levels in synaptoneurosomes and found that specific circRNAs, including circHomer1, were significantly enriched in this compartment in HuD over-expressing mice (HuD-OE) compared to wild-type mice. Furthermore, we found that the levels of both cirHomer1 and Homer1 mRNA, which is also a known target of HuD, were increased in the NAc of HuD-OE.
Conclusions: These results are consistent with previous studies showing changes in HuD and Homer1 mRNA and protein levels after cocaine administration. Since HuD levels are increased after CPP and HuD-OE show increased cocaine CPP, we propose that HuD-circRNAs interactions may regulate cocaine-induced neuroplasticity and behavior.
Disclosure: Nothing to Disclose.

20.4 Long Non-Coding RNAs in Addiction

Claes Wahlestedt

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Background: Recent high-throughput sequencing studies have uncovered tens of thousands of RNA transcripts with little or no protein-coding potential. Long noncoding RNAs (lncRNAs), defined as noncoding transcripts greater than 200 nucleotide bases in length. A prominent sub-class of lncRNAs, Natural Antisense Transcripts (NATs) are transcribed from the opposite DNA strand of other RNA transcripts with which they share sequence complementarities. Of the proposed functional mechanisms of NATs, regulation of chromatin architecture and epigenetic memory have received much attention, as some antisense transcripts may provide a scaffold by which proteins can interact with DNA and chromatin in a locus-specific manner. However, it is currently unclear if NAT-based mechanisms also play a role in drug-induced neuroadaptations.
Methods: Male C57BL/6 mice (8-10 weeks old) were used. Saline or cocaine (20 mg/kg) was given i.p once a day for 10 consecutive days for repeated exposure experiments and a single injection of saline or cocaine (20 mg/kg) for acute studies. Neuro-2a (N2a) and other cell lines were used.
We utilized a previously published bioinformatics algorithm to identify Natural Antisense Transcripts (NATs) from AceView transcriptome database. The open-access algorithm and the manual are available at (https://github.com/Dmitry Vel/NATs). Briefly, the algorithm takes an input list of genes and first identifies all transcripts expressed from the antisense strand of the target locus or the gene promoter (1 kb downstream from the transcription start site). Next, transcripts containing a significant open reading frame according to the annotation on AceView are filtered out to retain noncoding NATs. We utilized this pipeline to investigate the expression of antisense transcripts overlapping with 545 genes. This gene list was based on previously published genes that have been implicated in cocaine-induced neuroadaptations and from the top gene searches related to neuroplasticity. The pipeline output

yielded the existence of an antisense overlap, the position of the overlap, the NAT Aceview name, the NAT length and the overlap start and stop sites. PANTHER (http://www.pantherdb.org/) was used to categorize NAT-containing genes based on protein class.
Results: Aberrant regulation of gene expression is one critical factor underlying the long-lasting behavioral abnormalities that characterize substance use disorder, and it is possible that some drug-induced transcriptional responses are mediated, in part, by perturbations in NAT activity. To test this hypothesis, we used an automated algorithm that mines the NCBI AceView transcriptomics database to identify NAT overlapping genes linked to addiction. found that 22% of the genes examined contain clearly identifyable NATs and that, for example, expression of Homer1 natural antisense transcript (Homer1-AS) was altered in the nucleus accumbens following repeated cocaine administration. In *in vitro* studies, depletion of Homer1-AS lead to an increase in the corresponding sense gene expression, indicating a potential regulatory mechanisms of Homer1 by its corresponding antisense transcript.
Conclusions: NATs and other lncRNAs are altered by drugs of abuse. Many known addiction-related protein coding genes are likely regulated by lncRNAs.
Disclosure: Nothing to Disclose.

Panel
21. Translational Studies of Inter-Relationships of Homeostatic Regulation of Daily Rhythms of Sleep, Activity, Eating and Emotions

21.1 Mobile Technology Assessment of Homeostatic Regulation of Rhythms of Motor Activity and Related Domains: Implications for Cross-Species Translation

Kathleen Merikangas

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Background: There is growing recognition of the role of physical activity in human health. Physical activity is related to numerous human functional processes including cognition, sleep, and weight regulation, and is impaired in diseases such as Parkinson's disease and rheumatoid arthritis. Emerging evidence that dysregulation of the rhythms of motor activity may also underlie bipolar disorder (BPD), a mental disorder that has substantial individual and societal impact, and is associated with premature mortality. The emergence of mobile technologies has provided an unprecedented opportunity to obtain objective assessment of motor activity and its relationship to multiple homeostatic regulatory systems in real-time.
Methods: The sample included 242 adults from a large community-based family study of probands enriched for lifetime history mood disorders [10.3% Bipolar I (BPI); 12% Bipolar II (BPII); 37.6% Major Depressive Disorder (MDD); 40.1 Other/Controls] who were evaluated at the NIH Clinical Center. Actigraphy monitors (Respironics®, Actiwatch Score®) that collect minute-by-minute physical activity counts were worn on the non-dominant wrist for two weeks. Jointly with actigraphy, participants were signaled 4x/day on a mobile device that was programmed to administer brief electronic interviews regarding activities and environmental context at that particular moment, as well as 7-point Likert scales on mood and energy. Signals occurred at fixed intervals each day, with approximately a 4-hour delay between signals.
Results: In the full sample, time-lagged associations revealed a bi-directional association between motor activity with subjective energy, and with sleep, and a uni-directional influence of motor activity and energy on subsequent mood. Individuals with a history of BPD had significantly greater inter-relationships across all systems, suggesting increased cross-domain reactivity.
Conclusions: Dysregulation of motor activity and energy, and significantly greater cross-domain reactivity in the subset of the sample with bipolar I disorder (BPD) confirms growing evidence that BPD may reflect a primary disturbance of rhythms of activation. In vivo multi-system tracking in humans may provide new models of these regulatory systems and their interrelationships suitable for cross-species studies that can inform their underlying etiologic processes, and define potential targets and timing of interventions.
Disclosure: Nothing to Disclose.

21.2 Sleep Spindles and Schizophrenia Genetics

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Background: Sleep spindles – short bursts of 11 to 15 Hz oscillations during NREM sleep driven by thalamo-cortical circuits – are associated with various aspects of learning and memory and are potential biomarkers of neuropsychiatric disease, including schizophrenia. Although twin studies indicate that spindle activity is partially heritable, specific genes are yet to be identified. Towards this goal, here we detect and characterize spindle phenotypes in 11,630 individuals (aged 5 to 95), confirm the heritable basis of spindle activity, initiate genome-wide association analyses to map individual genes, and investigate the genetic links between spindles and risk of schizophrenia.
Methods: We compiled whole-night polysomnography, demographic and medical data from the US National Sleep Research Resource (NSRR), applying automated artifact rejection and wavelet analyses to detect spindles from two central electrodes. Univariate heritabilities and genetic correlations were estimated using within-family intraclass correlations and variance components models for the genome-wide single nucleotide polymorphism (SNP) data. Polygenic risk score analyses were used to model the relationships between spindle attributes and the genetic risk of schizophrenia.
Results: Spindle and spectral phenotypes demonstrated high test-retest reliabilities, based on over 4,000 individuals with repeated polysomnograms. In 730 individuals from the Cleveland Family Study (CFS), canonical spindle density was highly heritable in both white ($h2 = 0.45$, $p = 8x10\text{-}6$) and black individuals ($h2 = 0.43$, $p = 3x10\text{-}6$), adjusting for age and sex. Fast (15 Hz) and slow (11 Hz) spindles showed

significant heritabilities but no evidence for genetic overlap with each other ($p = 0.31$ and 0.18 for whites and blacks respectively), suggesting that distinct sets of genes underlie these two classes. We observed a significant associations between polygenic risk for schizophrenia and multiple spindle attributes, and are currently expanding the genetic analyses to include additional NSRR samples.
Conclusions: We observed evidence for robust genetic influences on spindle phenotypes, controlling for a range of demographic and clinical covariates. We also observed evidence for a partially shared genetic architecture between spindles and schizophrenia, consistent with the hypothesis that polygenic risk for schizophrenia is in part mediated by spindle activity. This work informs studies that aim to understand better the genetic architecture of spindles and their relation to neuropsychiatric disease.
Disclosure: Nothing to Disclose.

21.3 Neural Circuits Underlying Light Effects on Circadian Timing, Mood and Learning

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Background: Light exerts profound effects on circadian timing, mood and learning through intrinsically photosensitive retinal ganglion cells (ipRGCs). Diverse brain regions receive photic input from ipRGCs that reside in the retina located at the back of the eye. A primary target, the suprachiasmatic nucleus (SCN), has been considered central to the effects of light on circadian timing, mood and learning. Here we show, however, that light-mediated effects on mood require a distinct neural circuit from circadian timing and learning.
Methods: We used mouse genetics, chemogenetics, neural tracing, molecular, cellular and neural tracing methods to determine how light influences circadian timing, mood and learning.
Results: Using a combination of genetic and tracing methods, we show that ipRGCs innervate a region in the thalamus abutting the habenula, the perihabenular region (PHb), which contains a circadian clock, receives direct light input from ipRGCs and projects to the medial prefrontal cortex (mPFC). Light conditions that cause mood alterations in mice disrupt the circadian clock within the PHb and induce morphological changes in its cortical target, the mPFC. Using mouse-genetics and chemogenetic manipulations of PHb innervation and activity, we demonstrate that the PHb is both necessary and sufficient to drive the effects of light on mood. In contrast, we find that the SCN is sufficient for the effects of light on circadian timing and learning.
Conclusions: We establish a novel retino-thalamo-cortical pathway with a unique role in mood regulation. This shows that light influences mood and learning through distinct parallel circuits. These advances have important implications for elucidating the mechanisms for the influence on light on mood and mood disorders in humans.
Disclosure: Nothing to Disclose.

21.4 Precision of Actigraphy Phenotypes in Animal Models

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Background: Actigraphy is a classic method for phenotyping of circadian rhythms and has been a critical circadian measure for automated assessment in animal models. In rodents, circadian activity is robust, precise and amenable to automation. These features have enabled forward genetic screens for circadian phenotypes in the mouse and these screens have led to the discovery of core genes involved in the circadian clock mechanism in mammals.
Methods: To explore the diversity of circadian activity phenotypes in the laboratory mouse, we have systematically studied circadian activity rhythms in over 30 inbred mouse strains using wheel-running activity. Mice were monitored in light-dark cycles, constant darkness and constant light conditions.
Results: There was dramatic diversity in the circadian activity patterns, period, entrainment and level of activity in different inbred strains of mice. Some of these divergent phenotypes can be attributed to the species origin of the mouse strains.
Conclusions: Circadian activity rhythms can be extremely robust, however, there are marked quantitative differences in circadian phenotypes across different inbred mouse strains. The phenotypic diversity in mouse strains can model the individual differences observed in human actigraphy studies and provide evidence that phenotypic diversity in actigraphy has a strong genetic underpinning.
Disclosure: Part 1: Reset Therapeutics, Inc., Board Member, **Part 2:** Reset Therapeutics, Inc., Advisory Board, Reset Therapeutics, Inc., Stock / Equity.

Panel
22. Perinatal Adversity, Inflammatory Mechanisms and Biomarkers, and Neurodevelopment Relevant to Psychiatric Disorders

22.1 Prenatal Maternal Inflammatory and Steroid-Related Serum Biomarkers and Metabolic Syndrome Exposure Among Faster Offspring With Autism Spectrum Disorder

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Background: Prenatal metabolic syndrome (PNMS) may influence a child's vulnerability to neuropsychiatric impairment by affecting fetal brain development. PNMS' impact on fetal neurodevelopment occurs along a continuum involving several facets of childhood mental health with autism spectrum disorder (ASD) representing an extreme manifestation. This study's objective is to investigate the association between PNMS and ASD.
Methods: A population-based ASD Master Case List of Utah residents with ASD ($N = 31,137$) was established for the purpose of identifying ASD prenatal risk factors and underlying mechanisms. This case list was populated by

research sources and multiple public health registries. The ASD Master Case List was matched to offspring of the First and Second Trimester Evaluation of Risk (FASTER) study, which collected prospective medical information, maternal serum samples, and ultrasound reports on 7,964 pregnant women living along Utah's Wasatch Front between 1998 and 2002. Birth certificate records provided pregnancy data to identify PNMS-associated conditions (pregnancy induced hypertension, gestational diabetes, pre-/eclampsia, diabetes and chronic hypertension). Inflammatory and sex steroid-related markers (e.g. cytokines, C-reactive protein, sex hormone binding globulin, DHEA/S, estrogen, progesterone, testosterone, cortisol) in prenatal maternal serum drawn at 16 weeks gestation were compared between 9 term offspring with and 11 term offspring without PNMS exposure. None of these 19 children had ASD. A biomarker composite variable was constructed using dichotomous variable transformation. Comparisons of the frequency of PNMS in mothers of children with and without ASD were conducted using chi-square tests and differences in biomarker composite variable mean scores between mothers with and without PNMS were conducted using t-tests.

Results: The number of ASD cases ($n = 153$) among FASTER offspring reflects the expected ASD prevalence (1.9%). PNMS was significantly more likely ($p = 0.004$) in pregnancies of offspring with ASD than matched offspring ($n = 6104$) without ASD ($n = 27$, 17.7%; and $n = 638$, 10.5%, respectively). Inflammatory and steroid-related markers in serum at 16 weeks gestation predicted pregnancies that would subsequently become complicated by symptomatic PNMS ($p < 0.001$).

Conclusions: PNMS is associated with significantly elevated ASD risk. In pregnancies carried to term for children without ASD, PNMS can be predicted at 16 weeks gestation, at least 8 weeks before the onset of maternal symptoms. Of note, 16 weeks gestation falls within the critical period of neurodevelopment – neuronal migration. The ability to predict PNMS at 16 weeks and the link between PNMS and ASD collectively indicate the potential for primary prevention of ASD. We are currently analyzing 16 week maternal prenatal serum of children with ASD to determine how well this biomarker pattern fits pregnancies associated with ASD among offspring and will adjust the composite score accordingly. This biomarker composite score will inform the study of childhood neuropsychiatric functioning across Research Domain Criteria groups in the context of PNMS exposure.

Disclosure: Part 1: BioMarin Pharmaceuticals, Advisory Board, Audentes Therapeutics, Advisory Board, **Part 2:** Audentes Therapeutics, Advisory Board, **Part 3:** Audentes Therapeutics, Advisory Board.

22.2 The Impact of Preeclampsia on Neurodevelopment and Inflammatory Mediators Within a Vasopressin-Based Mouse Model

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Background: Preeclampsia is a risk factor for altered child neurodevelopment. Disrupted verbal ability, altered infant temperament, and an increased risk of autism spectrum disorder in offspring have been linked with exposure to prenatal preeclampsia. Despite the high 5-7% rate of preeclampsia, the mechanisms for its consequences on offspring are not understood. Animal models of preeclampsia which could allow mechanisms to be closely investigated have been limited in their criterion validity. A recently-developed preeclampsia model created with continuous vasopressin (AVP) infusion demonstrates multiple clinical preeclampsia signs–pregnancy-specific hypertension, renal glomerular endotheliosis, and proteinuria–and is ideally suited for understanding how brain development is altered.

Methods: In C57-Bl6J female mice, AVP (24 ng/hr) or saline was continuously infused through a subcutaneous implanted pump (Alzet). Breedings with C57-Bl6J males were monitored for vaginal plugs, preeclampsia was verified by proteinuria, and offspring brain tissue was collected at embryonic, neonatal and postnatal time points. To test involvement of inflammatory mechanisms, we assessed cytokine levels in placental and fetal tissues using ELISA and assessed offspring microglia morphologies in the neocortex using neurostereology with digital microscopy (Zeiss/Microbrightfield). We also assessed the trajectory of neocortical morphogenesis. Lastly, we examined behavior on Y-maze, elevated plus maze, open field, rotarod, social approach, and object recognition tasks in adult offspring.

Results: The AVP-based preeclampsia model altered levels of the pro-inflammatory cytokine IL-17 at E18 in placental and fetal tissues. Exposure to AVP-based preeclampsia shifted offspring cortical microglia to more activated morphologies embryonically. In these exposed offspring, growth of the cortical plate and neocortex was reduced in late embryogenesis into early postnatal life but normalized into adulthood. Preeclampsia-exposed adult males exhibited increased anxiety-like behavior and altered social behavior, while preeclampsia-exposed adult females exhibited impaired procedural learning and working memory.

Conclusions: Our results showing cytokine and microglia changes in fetal offspring of this AVP-based preeclampsia model suggest that preeclampsia induces a pro-inflammatory phenotype during fetal development. Exposure to this pro-inflammatory preeclampsia state resulted in a reduction in cerebral cortex volume beginning in late gestation and only recovering in adulthood. Despite this recovery of overall cortical structure, we found that AVP-based preeclampsia exposed offspring exhibited a constellation of aberrant behavior, suggestive together of altered cortical functioning. By using this model to understand the maternal physiological and fetal brain mechanisms underlying these altered offspring phenotypes, possible interventions during and after preeclampsia can be developed to support healthy neurodevelopment.

Disclosure: Part 1: Oakstone Publishing, Honoraria.

22.3 Links to the Past: Differential Impacts of Maternal Exposure to Early Childhood Adversity and Prenatal Stress Across Stress Response Systems

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Background: Current hypotheses related to the psychosocial mechanisms of the intergenerational transmission of maternal adversity include prenatal stress, maternal psychopathology, and maternal chronic stress. However, few studies have examined the impact of prenatal stress on concurrently measured stress response systems concurrently and fewer studies have included both prenatal stress and maternal exposure to her own early life adversity. Given evidence of the impact of both early adversity and prenatal stress on multiple outcomes exploration of transgenerational effects across stress response systems is warranted.
Methods: Data from two cohorts is presented 1) A cohort of Black youth recruited from charter schools ages 4-15 as part of a dental health study (New Orleans Stress Physiology and Children, NSPAC). 2) A cohort of mothers, 66% Black and 33% White, recruited prenatally as part of a study exploring the impact of maternal prenatal and early life stress on infant neurodevelopment. For the youth cohort data on maternal prenatal stress and ACE was assessed retrospectively and nasal swabs, an indicator of current immune activation were collected at a study visit on site. Nasal swabs were examined using light microscopy and H&E staining to provide a direct assessment of current inflammation with smears with predominant neutrophilic population suggesting a Th-17/ bacterial activation while smears with eosinophilic population would suggest a Th-2 mediated response with elevated pro-inflammatory cytokines. For the prenatal cohort (Infant Development Study, IDS) mothers were interviewed prenatally for their own exposure to early adversity and prenatal stress. A dyadic assessment was completed at 4 and 12 months of age. Cortisol reactivity was obtained at 4 and 12 months of age and RSA was also obtained at 4 months of age during an age-appropriate dyadic stressor. Telomere length (TL) was measured, using quantitative PCR, from DNA collected across visits from the infant. For the infant cohort a composite prenatal stress index was created from six scales: Edinburgh Depression, perceived stress, pregnancy related anxiety, prenatal life events, subjective social status, and chronic strain.9-13 The upper quartile (social status reverse coded) was defined as high on each scale and a cumulative sum score across scales was calculated. These scales loaded onto one primary factor that explained 38% of the variance with all factors loading at.5 or higher. Mixed effects modeling, linear regression and generalized estimating equations were used for analyses, all analyses was done in SAS. Maternal early adversity was captured using the Adverse Childhood Experience Scale (ACE) in both studies
Results: In NSPAC maternal retrospective report of prenatal stress was significantly associated with cellular predominance in youth ($b = -.07$, CI -.13, -.005) $p = .03$). In IDS prenatal stress was positively correlated with maternal ACE and decreased RSA at four months of age. When both were included in the model both ACE ($\beta = .02$, $t = -1.9$, $p = 0.05$) and prenatal stress ($\beta = .04$, $t = -2.2$, $p = 0.03$) remained significant predictors of infant RSA. Maternal ACE was a significant predictor of cortisol at four months however prenatal stress was not. At 12 months maternal ACE ($\beta = .53$, $t = 2.3$, $p = 0.04$) and prenatal stress ($\beta = .28$, $t = 1.9$, $p = 0.05$) were significant independent predictors of infant cortisol. When included in the same model there was a loss of the direct association between maternal ACE and infant cortisol reactivity, although prenatal risk remained significant. Relative to TL trajectory from 4 to 18 months, maternal ACE and prenatal stress were independent predictors of TL trajectory, but only prenatal stress remaining significant in the combined model ($\beta = .36$, $t = 2.04$, $p = 0.04$).
Conclusions: Thus, the impact of maternal ACE differs across stress response systems, is differentially moderated by prenatal stress, and has changing developmental trajectories.
Disclosure: Nothing to Disclose.

22.4 Placental Mediated Mechanisms of Impaired Neurodevelopment

Lauren Jantzie

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Background: Minimizing central nervous system (CNS) injury from preterm birth depends on identification of critical pathways underlying essential neurodevelopmental programs and CNS pathophysiology. While chorioamnionitis (CHORIO) is a known precursor to brain injury in preterm infants, the precise mechanisms linking prenatal injury and long-term CNS damage are unknown. We hypothesized that the chemokine CXC-ligand 1 (CXCL1) and its receptor CXCR2, would be dysregulated in CNS injury concomitant with a persistent immune response, including neutrophil trafficking, in our preclinical model of CNS injury associated with preterm birth.
Methods: Using an established model of CHORIO in rats that results in deficits in the mature CNS that mimic those of children born preterm, uterine arteries were occluded for 60 min, and lipopolysaccharide (LPS) was injected into amniotic sacs on embryonic day 18 (E18). Pups were born at term (E22). SB225002, a CXCR2 antagonist, was administered intraperitoneally from postnatal day 1 (P1)-P5 (3 mg/kg). Brain, serum and placenta were collected from E19 to P15 and analyzed using multiplex electrochemiluminescence, Western blot, qPCR and flow cytometry (FC) with $n = 4$-8/group (t-test or ANOVA with Bonferroni correction with $p < 0.05$ indicating statistical significance). Executive function and cognition was assessed through translational touchscreen measures.
Results: Compared to shams, CHORIO increased placental CXCL1 (153 ± 23 vs 94 ± 15 pg/100 μg, $p < 0.01$) and yielded sustained serum CXCL1 elevation through P15 ($p < 0.05$). Notably, brain CXCL1 remained elevated in CHORIO rats compared to sham from P2 ($P < 0.05$) through P7 ($p < 0.001$). FC in brain lysate showed increased CXCR2+ neutrophils and microglia in CHORIO compared to shams (all $p < 0.05$). CHORIO rats were able to perform touchscreen testing of visual discrimination but demonstrated a lack of cognitive flexibility and deficits of executive function when performing reversal tasks. Specifically, CHORIO animals have longer latency to paradigm learning, commit significantly more errors ($p = 0.03$) and exhibit more preservative behavior ($p = 0.008$) than shams. The CXCR2 antagonist, SB225002, reduced cerebral neutrophil activation, gliosis, and αII-spectrin cleavage at P7, indicative of improved neural cell health ($p < 0.05$).
Conclusions: These data show a sustained systemic and neuroinflammatory response following CHORIO in rats, and

for the first time we demonstrate impaired executive function using a touchscreen platform, a highly translatable method of preclinical cognitive assessment. Further characterization of neuroinflammatory signaling pathways will facilitate identification of brain and serum injury biomarkers and advance new therapeutic strategies and stratification of newborns with CNS injury to existing therapies. Additional touchscreen testing will allow dissection of deficits in distinct pillars of cognition following in utero injury.
Disclosure: Nothing to Disclose.

Mini Panel
23. Toward Reproducible Transdiagnostic Markers of Psychopathology in the Developing Brain

23.1 Common Neuroanatomic and Functional Substrates of Psychopathology Across Clinical Diagnostic Categories in Youth

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Background: Co-morbidity among psychiatric disorders is extremely common, and may be particularly challenging in youth, where clinical phenotypes are often less distinct. Such high comorbidity among neuropsychiatric disorders suggests common neurobiological origins, which may confer vulnerability to a range of symptoms and diagnoses. Here I will review a series of recent unpublished data that seek to delineate common anatomic and functional deficits across clinical diagnostic categories in a large sample of youth who received multi-modal neuroimaging as part of the Philadelphia Neurodevelopmental Cohort (PNC).
Methods: The sample included 1,042 youths (ages 11 to 23 years) who completed a clinical diagnostic interview and cross-sectional imaging as part of the PNC using a single 3T scanner. A dimensional measure of psychopathology was constructed using a bifactor model of item-level data from a psychiatric screening interview, which delineated four factors (fear, anxious-misery, psychosis, and behavioral symptoms) plus a general factor, which represented overall psychopathology present across disorders (the "p factor"). Regional brain volume was measured using T1-weighted imaging, cerebral blood flow (CBF) was measured using arterial spin-labeled MRI, and intrinsic functional connectivity was measured using a resting-state BOLD acquisition. Covariance networks were derived using non-negative matrix factorization, an advanced multivariate analysis technique. Group level analyses utilized generalized additive models with penalized splines to capture both linear and nonlinear developmental effects. Multiple comparisons were accounted for using the False Discovery Rate (FDR $Q < 0.05$).
Results: Overall psychopathology was dimensionally associated with a constellation of specific structural and functional abnormalities. These included prominent volume loss and reduced cortical thickness that was maximal in frontal and temporal cortex. Additionally, overall psychopathology was associated with elevated perfusion in several regions including the dorsal anterior cingulate cortex (ACC) and rostral ACC. Follow-up analyses of functional connectivity data revealed that the dorsal ACC had reduced connectivity to the bilateral caudate. Furthermore, specific dimensions of psychopathology demonstrated dissociable associations. For example, psychosis symptoms were related to reduced perfusion in the frontal operculum and insula, whereas fear symptoms were associated reduce perfusion of the subgenual ACC.
Conclusions: Taken together, these results delineate both common and dissociable abnormalities across neuropsychiatric disorders in youth. Shared anatomical and functional deficits revealed by multi-modal imaging suggest common mechanisms of vulnerability to psychopathology that cross clinical diagnostic boundaries.
Disclosure: Nothing to Disclose.

23.2 Establishing a Framework for Optimizing Reproducibility Among Studies of Typical and Atypical Brain Development

Michael Milham

Child Mind Institute, New York, New York, United States

Background: The attainment of reproducible findings is critical to efforts focused on the development of brain-based biomarkers for neurodevelopment. In this regard, the neuroimaging community has welcomed the rise of numerous large-scale imaging samples focused on the delineation of brain development in clinical and non-clinical populations, as they avoid established limitations of underpowered samples. Although promising, increasing the size of samples may not be sufficient to fully address issues of reproducibility. Across imaging samples, a range of differences in phenotyping protocols and sampling strategies exist, which if not taken into account, can compromise reproducibility – potentially more than the commonly cited differences in imaging protocols. I will describe a data-aggregation effort focused on bringing independently designed samples together to: 1) assess differences among imaging samples, 2) enable a mapping of phenotyping into a common space using the bifactor model (e.g., p-factor, fear, externalizing), and 3) establish optimal methods of maximizing reproducibility.
Methods: We reviewed the literature and publicized ongoing efforts to identify large-scale ($n > 200$) multimodal imaging samples of brain development in clinical and non-clinical populations. For samples not publicly available, we contacted the investigators to determine their willingness to share their data (phenotypic and imaging). Upon completion of this process, we reviewed phenotypic protocols for the samples included, identifying differences in key assessment domains, ranging from those focused on providing a categorical DSM-diagnostic label, to those focused on providing dimensional characterizations; for the latter, we differentiated between tools providing multidimensional characterizations and those focused on a specific domain (e.g., ADHD). We also reviewed sampling strategies, noting recruitment strategy time and intended sample composition. Finally, we looked at differences in imaging protocols.
Results: Developmentally-focused imaging samples containing functional and structural MRI included: NIH ABCD Study, Brazilian High Risk Cohort, Child Mind Institute

Healthy Brain Network, Chinese Color Nest Project, Human Connectome Lifespan Project, NKI-Rockland Sample, Philadelphia Neurodevelopmental Cohort, P.I.N.G., IMAGEN, and Saguenay Youth. A range of variation in phenotypic protocols existed. When present, the tools employed to characterize a DSM diagnosis varied (e.g., KSADS, DAWBA, GOASSESS). Even when looking at the same instrument (e.g., the K-SADS), differences in the specific implementation exist. The Childhood Behavioral Checklist was the most frequently used tool for providing multidimensional psychiatric characterizations. When focusing on more in-depth characterizations of a given domain of illness (e.g., ADHD), again marked variation was noted in the questionnaires employed and their psychometric properties. Similarly, differences were noted in the recruitment strategies used across studies, regardless of whether focusing on typically developing populations or those affected neurodevelopmental disorders (e.g., clinical referral, community ascertained, family-history based). Finally, as expected, we noted marked differences in the imaging sequences used to support each, resting state fMRI and diffusion imaging.

Conclusions: Taken together, our findings highlight a range of sources in variation across large-scale imaging studies focused on brain development. While the imaging field has made significant strides in the development of strategies for assessing and overcoming differences in imaging protocols, notably less attention has been given to the phenotypic protocols employed. The motivations for using the bifactor model to link phenotyping across differing samples will be discussed, as well as next steps for the initiative (i.e., imaging analysis).

Disclosure: Nothing to Disclose.

23.3 The Development of Dissociable and Common Aspects of Psychopathology From Childhood to Adolescence and Their Brain Structural Correlates

Giovanni Salum

Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil

Background: Psychiatric symptoms tend to aggregate into distinct clusters, presenting a dissociable aspect. However, symptoms across these clusters also tend to co-occur, presenting a common aspect. Bifactor models are statistical models that allow us to capture the dissociable and the common aspects of psychopathology. Here, I will review recent unpublished data that aim to investigate the longitudinal development of dissociable and common aspects of psychopathology, as well as their structural brain correlates using data from the Brazilian High Risk Cohort Study (HRC). I will also report initial findings from an effort focused on linking distinct phenotypic protocols from different samples together using the bifactor model.

Methods: The HRC sample included 2,511 children and adolescents (ages 6 to 14 years of age), of which 80% were followed after 3-years (ages 9 to 17). Psychopathology was measured by diagnostic (Development and Well-Being Behavior; DAWBA) and symptomatic (Child Behavioral Checklist; CBCL) instruments. A bifactor model with three dissociable symptomatic dimensions (fear, distress and externalizing) in combination with one common factor (the "p-factor") was fitted to the data. Growth mixed effects models investigated longitudinal trajectories taking advantage of the accelerated longitudinal design. Measures of cortical thickness, area and volume were measured using T1-weighted imaging processed using freesurfer in 720 participants. Associations with symptom dimensions were assessed by using general linear models using Bonferroni correction ($p < 0.00006$).

Results: First, the bifactor model with one common factor and three dissociable factors presented excellent fit to HRC data. Second, growth mixed models revealed inverted U quadratic trajectories with development for the distress factor (peaking at age 13) and for the fear factor (peaking at age 12), but not externalizing. For the p-factor, a significant trend-level ($p = 0.055$) Age X Sex interaction emerged, with an inverted U quadratic trajectory for boys (peaking at 11) and a linear increasing for girls. Third, brain structural analysis revealed the p-factor was associated with higher thickness in the cuneus, lingual gyrus and pericalcarine cortex bilaterally, and lower area in the left lingual and pericalcarine cortices. Higher p-factor was associated with lower area in the lateral and medial portions of the orbitofrontal cortex bilaterally. Fear and distress factors were associated with higher left putamen volume and right cuneus thickness. Finally, we found that the same bifactor model had an excellent fit with phenotypic protocols from other developmental imaging samples (i.e., NKI-Rockland Sample, CMI Healthy Brain Network and Chinese Color Nest Project).

Conclusions: These results demonstrate the potential of bifactor models to disentangle common and dissociable aspects of psychopathology in youth, revealing their developmental trajectories from childhood until adolescence and their brain structural correlates. These results reinforce both the importance to start looking for shared mechanisms for psychiatric disorders, but also to account for p-factor when looking for specific mechanisms of psychopathology. We also outline the potential of such models for data integration across different samples.

Disclosure: Nothing to Disclose.

Panel
24. Using Neuroimaging to Guide the Development of Novel Interventions

24.1 A Circuits-First Approach to Mental Illness

Amit Etkin

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Background: The way in which we have defined psychiatric diagnoses (i.e. based only on symptom clusters) and identified treatments (i.e. capitalizing on serendipity), has failed to substantially mitigate the disabling burden of mental illnesses. Not surprisingly, individual psychiatric diagnoses are highly clinically and biologically heterogeneous, with as much or greater variability within a diagnosis as between diagnoses, and typically only half of patients respond well in clinical trials. Neuroimaging, as the dominant tool in human

neuroscience, however, has been used largely for comparing these arbitrarily-defined diagnoses against healthy individuals not for robustly characterizing individual patients in objective biological terms. Imaging is also a purely observational method, and thus cannot by itself provide the causal understanding of circuitry that is necessary for transitioning from a descriptive to a circuit-based mechanistic understanding of mental illness that can directly guide novel interventions. I will outline in this presentation an approach, buttressed by new and mostly unpublished data, that by understanding causality in the brain circuits of individual patients we can pursue personalized diagnosis and treatment using individually-tailored plasticity-inducing neurostimulation, thereby establishing a direct linkage between circuits and clinical outcome.

Methods: I will present on studies using resting-state fMRI, concurrent TMS/fMRI and/or concurrent TMS/EEG, including patients with depression or PTSD. Studies include: a longitudinal sham-controlled study of repetitive TMS (rTMS) for depression, a longitudinal wait-list controlled study of psychotherapy for PTSD, and cross-sectional investigations of PTSD in two independent cohorts.

Results: We found for rTMS treatment in depression that better clinical outcome is seen in patients for whom the function (at rest or as measured by TMS/EEG stimulation probes) of the dorsolateral prefrontal target for rTMS was more perturbed. This signal can furthermore be used to optimize the stimulation location and coil positioning angle even in individual patients. In PTSD, we found that otherwise symptomatically indistinguishable patients can be divided into two broad categories based on behavior and resting-state fMRI connectivity, and that these categories predict dramatically different outcomes with psychotherapy. Moreover, resting-state connectivity was correlated with TMS/EEG evoked responses at specific prefrontal locations, establishing a circuit signature that can in turn be used as a target for treatment development. Finally, using concurrent TMS/fMRI we found a prefrontal region which can causally regulate amygdala activity, and that this causal influence was deficient in PTSD, providing a novel target for development of emotion circuit-directed rTMS treatments.

Conclusions: These findings suggest a path forward towards a circuit-based approach for diagnosis and treatment in psychiatry, centered primarily around tools for mapping causal influence in brain circuits (i.e. TMS/EEG, TMS/fMRI) that transcends the arbitrariness and heterogeneity of traditional diagnoses, the limitations of group-level imaging analyses and current trial-and-error approaches to treatment planning. In a simplistic formulation this would involve identification of abnormalities in circuits, as revealed through targeted imaging-coupled neurostimulation, and remediating these abnormalities through rTMS. Realization of this potential, however, will require extensive work to establish and validate the concepts and their implementation. Particular future challenges include robustly applying circuit-based decision rules to individuals and optimization of rTMS interventions for the identified circuit abnormalities.

Disclosure: Part 1: Takaeda, Consultant, **Part 2:** Akili Interactive, Advisory Board, Mindstrong, Advisory Board.

24.2 How Surprising Imaging Findings Refine New Approaches to Cognitive Training in Schizophrenia

Sophia Vinogradov

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Background: In recent years, neuroscience-informed cognitive training methods that target precisely defined aspects of impaired functioning in auditory systems have been developed for schizophrenia. After intensive training, improvement in untrained neuropsychological outcome measures is observed, and magneto-encephalographic and functional MRI studies reveal evidence of widespread cortical plasticity; certain neural system changes correlate with cognitive gains and with improvements in real-world functioning at 6-month follow-up. However, approximately 35-40% of individuals do not respond to such training, suggesting that there are as-of-yet unidentified factors that constrain (or amplify) the ability to engage with the treatment target and to respond to a neuroplasticity-based intervention.

Methods: Post-hoc analyses of MEG findings suggested that two distinct patterns of cortical plasticity in superior temporal vs. ventral visual cortical areas were seen in participants after auditory system training. Based on these imaging findings, we combined 4 data sets derived from our previous randomized controlled trials and applied PCA and topological data analysis to cognitive and symptom data. Our aim was to determine the existence of distinct longitudinal response patterns to the active auditory system training condition vs. the visual computer games control condition.

Results: PCA on symptoms and cognition at baseline ($N = 363$) revealed three distinct multidimensional domains that were stable at the post-training time-point. In contrast, PCA on change scores ($N = 296$) revealed four distinct response patterns, three of which were consistent with cross-sectional analyses, and a fourth that showed unique features not present at either cross-sectional time point. All four patterns showed a significant differential response to targeted auditory system training vs. visual computer games. The fourth newly emerging pattern indicated competitive interference between auditory and visual system plasticity in schizophrenia. Mapping individual patients by their scores on these response patterns using TDA separated patients into four distinct clusters with unique responses to the training interventions within these new multidimensional domains, suggesting the possibility of precision psychiatry based on cognitive profiles.

Conclusions: People with schizophrenia who undergo 40-50 hours of auditory system training vs. visual computer games show four discrete response patterns in their symptoms and cognitive measures, and each response pattern shows a differential association with the auditory vs. visual intervention condition. The implications for the development of cognitive treatments and for precision psychiatry in schizophrenia and related disorders will be discussed.

Disclosure: Part 1: Posit Science, Grant, Otsuka-Lundbeck, Honoraria, **Part 4:** Posit Science Inc., Grant.

24.3 Novel Mechanism-Based Treatments for Disruptive Mood Dysregulation Disorder

Melissa Brotman

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Background: Irritability has been conceptualized as aberrant responses to threat and frustration. Disruptive Mood Dysregulation Disorder (DMDD) is a new diagnosis in the DSM-5 that captures youth with chronic, severe irritability. The addition of DMDD to the DSM-5 reflects a recognition of the lack of evidence-based treatments for severe irritability in youth. Here, we used fMRI data to identify behavioral and neural mechanisms of irritability and develop circuitry-based targets for novel interventions.
Methods: During fMRI, we studied the associations among irritability, behavioral performance, and neural functioning while probing threat and frustration in two separate paradigms. In both fMRI studies, we included youth with an anxiety disorder, DMDD, and/or attention deficit hyperactivity disorder, as well as healthy volunteer children. First, to study threat, we used an implicit (gender) labeling task with multiple face emotions (happy, angry, fearful) across various intensities in N = 115 youth. Second, we examined the associations between irritability and the impact of frustration on neural functioning. To do so, we used a modified Affective Posner fMRI task in N = 195 youth. This paradigm models frustrative non-reward by first inducing and then violating the expectation of reward in the context of an attention orienting task.
Results: During the implicit labeling face emotion processing task, we found that irritability and anxiety influenced amygdala connectivity to the prefrontal and temporal cortex. Specifically, during viewing of intensely angry faces, decreased amygdala-medial prefrontal cortex connectivity was associated with high levels of anxiety and irritability; increased connectivity was associated with high levels of anxiety, but low levels of irritability, supporting the role of aberrant threat processing in both irritability and anxiety. During the Affective Posner task, following frustration, when subjects completed an attentional task, higher irritability was related to increased activation in multiple frontal regions, including bilateral cingulate gyrus and dorsolateral prefrontal cortex.
Based on behavioral and pathophysiological dysfunction in irritability, we explored two innovative mechanism-based treatments for DMDD. Interpretation bias training (IBT) is a computer-based training that targets aberrant responses to threat. We also developed a novel translational psychotherapy (cognitive behavioral therapy, CBT) targeting aberrant responses to frustration using exposure. Because both anger and fear are mediated by threat circuitry encompassing the amygdala and prefrontal cortex, we propose that extinction learning will occur as patients are exposed to frustrating situations in a graded fashion and develop increased frustration tolerance. We present work from both mechanism-based treatments indicating preliminary efficacy.
Conclusions: Severe irritability is one of the most common reasons that children present for mental health care; however, few effective treatments are available. Leveraging behavioral and neural dysfunction in DMDD based on aberrant responses to threat and frustration, we present the rationale for two novel mechanism-based treatment approaches.
Disclosure: Nothing to Disclose.

24.4 Predictive Biomarkers for Interventions in Anxiety and Depression

Martin Paulus

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Background: Positive and negative affect systems are dimensions of psychopathology identified by the RDoC work groups (Health, 2011. High negative affect is common to anxiety and depression (Brown et al, 1998; Chorpita, 2002; Prenoveau et al, 2010) and comorbid anxiety and depression is associated with more negative affect than each disorder alone (Weinstock and Whisman, 2006). Moreover, in the cognitive domain neuropsychological research provides evidence for subtle deficits in executive functions, including response inhibition and attention regulation, particularly for individuals with posttraumatic stress disorder (PTSD) (Aupperle et al, 2012; Polak et al, 2012). Executive functions are considered important for supporting optimal social and occupational functioning and likely contribute to emotion regulation abilities (Barkley and Murphy, 2010; Zelazo and Cunningham, 2007). We aimed to determine whether processing dysfunctions in the positive and negative valence domains and during inhibitory processing could contribute to the prediction of response to standardized behavioral interventions.
Methods: In the first study, 10 women with IPV-related PTSD (mean age = 34.60; SD = 9.40) and 12 healthy control women (HCW; mean age = 35.25; SD = 6.44) with no trauma history completed the Stop Signal Task during functional magnetic resonance imaging (fMRI). Linear mixed models were used to investigate group differences in activation (Stop minus NonStop). In the second study, 37 women with PTSD related to intimate partner violence and 34 age-matched healthy control women (HCW) completed fMRI to examine neural responses during anticipation of negative (ANI) and positive (API) emotional images. The Clinician-Administered PTSD Scale (CAPS) was used to characterize PTSD symptom severity. Wechsler Adult Intelligence Scale (WAIS-III) Digit Symbol test, Delis-Kaplan Executive Function System (D-KEFS) Color-Word Test, and Wisconsin Card Sorting Test were used to characterize neuropsychological performance. Fourteen of the women with PTSD subsequently completed cognitive trauma therapy for battered women (CTT-BW).
Results: (1) although women with IPV PTSD did not perform more poorly on the stop signal task, these individuals exhibited greater differential activation to Stop-NonStop in right dorsolateral PFC and anterior insula and less differential activation in several default mode network regions (i.e., precuneus, medial PFC). (2) women with PTSD performed worse on complex visuomotor processing speed (Digit Symbol) and executive function (D-KEFS Color-Word Inhibition/Switching) measures compared to HCW. PTSD

was associated with greater anterior insula and attenuated lateral prefrontal (PFC) activation during emotional anticipation. Greater dorsolateral PFC (dlPFC) activation (ANI-API) was associated with lower PTSD symptom severity and better visuomotor processing speed and executive functioning. Furthermore, greater pre-treatment ACC activation during anticipation related to better CTT-BW treatment response.
Conclusions: These results are consistent with the hypothesis that individuals with PTSD have separable affective and cognitive dysfunctions, which can be probed on several units of analysis (symptoms, behavior, and circuits) to develop new and more targeted interventions.
Disclosure: Nothing to Disclose.

Panel
25. Re-Energizing the Development of Pain Therapeutics in Light of the Opioid Epidemic

25.1 Biased Opioid Agonists as Safer Analgesics

Laura Bohn

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Background: The rate opioid induced overdose is rapidly increasing across the country. While efforts are underway to restrict opioid prescription availability, pain remains. The mu opioid receptor (MOR) is a G protein coupled receptor (GPCR) and as such, has become to be recognized for its ability to signal through multiple signaling pathways. In mice lacking a signaling scaffold protein, βarrestin2, morphine produces antinociception without respiratory suppression. These studies prompted hypothesis that if one could develop an agonist that activates MOR signaling through G proteins yet does not promote βarrestin recruitment, it may be a way to dissociate the side effects from the pain relieving properties (a G protein biased agonist). The first agonist reported to be a MOR biased agonist is currently in phase 3 clinical trials and it is showing a modest improvement in separating the analgesic properties from respiratory suppression in a post-surgical setting. Herein we ask whether improving the bias (the separation between G protein signaling and βarrestin recruitment) can predict an even greater separation between antinociception and respiratory suppression.
Methods: Cell based signaling assays are used to assess pharmacological parameters of morphine, fentanyl, and new small molecule compounds for their relative potency and efficacy for stimulating G protein signaling or βarrestin2 recruitment. The operational model is applied to calculate the "bias factor" and compounds are selected to represent different degrees of biased agonism. Pharmacokinetic data are collected to confirm a plasma and brain presence. Compounds were tested for antinociceptive responses in C57Bl/6 J mice via the hot plate and warm water tail immersion tests and for respiratory parameters using a pulse oximeter designed for rodents. Full dose response studies were carried out and ED50 parameters were calculated. Therapeutic windows were calculated comparing respiratory ED50s/ antinociceptive ED50s. Correlation analyses were performed to determine the coefficient of determination comparing bias factor and therapeutic window.
Results: We identify several new compounds, all structurally related, with increasing degrees of bias for G protein signaling. Within the chemical scaffold, we also identify a βarrestin-biased agonist and we show that fentanyl also shows bias towards βarrestin2 recruitment In vivo, we find that all of the agonists induce antinociception with ED50 values similar to morphine (with some more potent, such as fentanyl). However, as bias for G protein signaling increases, there is less respiratory suppression. Analysis for correlation produces a coefficient of determination of 0.97 for bias factor and therapeutic index. Further, compounds that show bias for recruiting βarrestin2, such as fentanyl, also fall within this correlation- producing a more narrow therapeutic window than that observed with morphine.
Conclusions: These results demonstrate a correlation between biased agonism detected in cell based assays with the separation of the therapeutic window in vivo; with G protein biased agonists showing improved therapeutic profiles and βarrestin2 biased agonist performing with a more narrow safety window. These findings are in concert with the hypothesis that the development of MOR agonists that are biased towards G protein coupling rather than βarrestin2 may provide a way to develop safer opioid analgesics for the treatment of pain.
Disclosure: Nothing to Disclose.

25.2 Novel Mu Opioid Receptor Targets for the Treatment of Pain

Gavril Pasternak

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Background: Most opioids used to treat pain act through mu opioid receptors. The cloning of the mu opioid receptor gene Oprm1 soon led to the identification of a large number of splice variants with similar splicing patterns in mice, rats and humans. These can be broken into three classifications based upon their structures. The largest group are traditional, 7 transmembrane G-protein coupled receptors (7TM) generated by the exon 1 promoter that differ from each other only in the tip of the intracellular C-terminal. A second group generated through the exon 1 promoter is comprised of only a single transmembrane domain. The third group utilizes a different promoter associated with exon 11 to produce truncated proteins possessing only 6 transmembrane domains (6TM). All three sets of variants are functionally relevant.
Methods: To assess the functional role of alternative splicing, a series of knockout mice were generated with the selective loss of full length, traditional 7TM variants, truncated 6TM variants or both and used to define the actions of a series of opioids. Drugs were classified based upon their dependence upon either 7TM, 6TM or both sets of Oprm1 variants. Confirmation of the classification was confirmed by rescue studies using lentivirus expressing select splice variants.
Results: Knockdowns or knockouts removing 7TM receptors eliminate morphine analgesia, establishing its dependence upon traditional 7TM receptors. Loss of 6TM variants in an

exon 11 knockout model (E11 KO) does not influence morphine analgesia, respiratory depression or reward in conditioned place preference studies, consistent with a 7TM mechanism of action. However, morphine locomotor activity and hyperalgesia was lost in the E11 KO mice and tolerance developed more slowly. IBNtxA, a novel opioid that targets a 6TM-dependent binding site in brain, is a potent analgesic. Unlike morphine, IBNtxA analgesia is lost in the E11 KO mice and retained in a exon 1 KO, implying that its analgesic actions are totally dependent upon 6TM variants. This was confirmed in rescue studies in which a 6TM variant was expressed through a lentivirus vector in knockout mice. The expression of the 6TM variant restored IBNtxA analgesia, but not morphine analgesia. Further characterization of IBNtxA revealed that it lacks respiratory depression and reward in conditioned place preference. It shows no cross tolerance to morphine and chronic administration fails to produce physical dependence.
Conclusions: These findings illustrate the complexity of mu opioid receptor action. Most opioids are morphine-like, producing analgesia through traditional 7TM variants and possessing similar side-effect profiles. IBNtxA illustrates the ability to target 6TM variants to obtain analgesia lacking many of the problematic side-effects and suggests that safer, effective analgesics are possible.
Disclosure: Nothing to Disclose.

25.3 New Approaches to the Management of Chronic Pain

Allan Basbaum

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Background: With a view to replacing morphine and other opiates in the management of chronic non-cancer pain, recent clinical trials using antibodies to nerve growth factor have proven remarkably effective in many osteoarthritis conditions and are being tested for back pain, the most prevalent chronic, non-cancer pain condition. Antibodies to calcitonin-gene related peptide are also proving incredibly promising not only for the treatment but also for the prophylaxis of migraine. A very different opportunity to produce a highly selective, opioid-free approach to chronic pain control comes from the development of blockers of the NaV1.7 subtype of voltage-gated Na channel, loss of function of which results in a condition of insensitivity to pain. Finally, I will describe two recent successful preclinical approaches that illustrate additional reasons for optimism that alternatives to opiates in the management of chronic pain are on the horizon. These studies involve viral-based gene therapies that selectively target "pain" transmission neurons as well as spinal cord transplants of GABAergic inhibitory interneuron progenitor cells in models of neuropathic pain.
Methods: NA
Results: NA
Conclusions: NA
Disclosure: Part 1: Neurona Therapeutics, Advisory Board, SwitchBio, Stock / Equity, **Part 2:** SwitchBio, Stock / Equity, Neurona Therapeutics, Advisory Board, **Part 4:** Toray Industries, Grant.

25.4 Functional Connectivity-Based Biomarkers for Chronic Musculoskeletal Pain

Robert Edwards

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Background: Chronic pain constitutes a public health epidemic and a serious therapeutic challenge. Inter-patient variability in analgesic outcomes is enormous, and there is tremendous interest in characterizing patient phenotypes that are associated with better or worse outcomes of chronic pain treatments (e.g., the degree of opioid analgesia, the development of opioid tolerance, or medication misuse behaviors). Understanding these phenotypes promises to facilitate the identification of pain mechanisms and biomarkers that can speed the development of novel therapeutics, and contribute to advances in personalized pain medicine in which treatments are optimized on the basis of patient phenotype.
Methods: In several studies of patients with chronic musculoskeletal pain (e.g., fibromyalgia, chronic low back pain), our group is employing 3T functional MRI to study patterns of brain connectivity that may serve as mechanistic endophenotypes linking psychosocial factors (anxiety, pain-related catastrophizing) and pain-modulatory processes (e.g., indices of endogenous pain inhibition and pain facilitation) with critical pain-related outcomes. Applying methods of seed-based connectivity to BOLD data collected during the application of a tonic musculoskeletal pain stimulus, we compare groups of chronic pain patients to demographically-matched healthy controls, and quantify inter-patient variability in pain-induced patterns of functional connectivity.
Results: Patients with chronic musculoskeletal pain (fibromyalgia, low back pain) report elevated levels of psychosocial distress and maladaptive changes in pain-modulatory processes (i.e., impaired pain inhibition and amplified pain facilitation) compared to pain-free controls. In addition, the patient groups differ from control groups on indices of both resting and pain-induced functional brain connectivity between regions of the anterior insula and several networks, including the default mode network (DMN) and somatosensory network. Enhanced connectivity between anterior insula and these networks is linked to negative affective processes, maladaptive pain-modulatory profiles, and poor outcomes of treatment for chronic pain. Mediational analysis suggests that altered patterns of connectivity serve as a mechanism linking established biopsychosocial phenotypes with treatment outcomes in chronic musculoskeletal pain.
Conclusions: Indices of fMRI-assessed brain connectivity show promise as biomarkers linking patients' psychosocial and psychophysical phenotypes with long-term pain-related outcomes. Such work has the potential to advance the science of personalized pain medicine, and facilitate the identification of pain mechanisms and biomarkers that can speed the development of novel therapeutics. The most substantial challenge to effective chronic pain treatment is the enormous inter-patient variability in response to a given therapy. We

hope that this work can contribute to a personalized pain management paradigm in which treatments are optimized for individual patients on the basis of their phenotypic characteristics.
Disclosure: Nothing to Disclose.

Study Group
26. Phenotype Harmonization for Neuropsychiatric Genomics Research

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Study Group Summary: Efforts to identify genetic associations to psychiatric phenotypes are beginning to evaluate variants across the allele frequency spectrum and to incorporate increasingly diverse study populations. As these projects attain global scales they face challenges arising from variation in phenotype definition and measurement across languages, cultures, sites, instruments, and diagnostic entities. Phenotype harmonization standards are necessary to justify combining measures across studies, to compare individuals, to track changes over time, and to establish valid, cross-culturally replicable genetic associations. This ACNP Study Group focuses on the NIMH Whole Genome Sequencing in Psychiatric Disorders (WGSPD) Consortium. Harmonization of diagnostic, symptom, and neurocognitive phenotypes is an acute issue for the Consortium. It is conducting WGS in ~20,000 individuals across five diagnostic categories (schizophrenia, schizoaffective disorder, bipolar disorder, major depressive disorder, and autism spectrum disorders), drawn from 10 countries over 5 continents, with great variation in inclusion/exclusion criteria and instruments used for phenotyping. The first 30 minutes of this Study Group will detail the project scope and frame questions for a 60-minute discussion, including:
- How should we define and model phenotypes? Given the absence of universally agreed or validated phenotype definitions, how best to address the tension between different psychometric modeling strategies? The latent variable approach can be contrasted with network and causal modeling strategies. Do latent class models support the traditional diagnostic taxonomy? Are there meaningful relations of symptoms to external variables including neurocognitive measures? Are mixture models needed to consider how different measurement models may be needed in different subpopulations?
- If we accept a priori the validity of certain latent variables or networks of symptoms or neurocognitive domains (e.g., "depression", "working memory"), or specific diagnostic entities (e.g., "Schizophrenia", "Bipolar Disorder"), what item-level measures enable the most efficient assessments? Do additional variables provide added meaningful phenotype variation? Can we prune redundant variables to increase efficiency?
- Do existing measures converge on constructs operationalized in the NIMH Research Domains Criteria (RDoC) initiative? Does this framework fit the empirical data better than traditionally-defined symptom, syndrome, or neurocognitive constructs?
- What is our measurement error tolerance for planned WGS analyses? How should we optimize the balance between precision in phenotype specification and the time needed to gather these data?
- Given the successes and challenges in phenotype harmonization so far, should we propose a focused study to provide a "Rosetta Stone" for translation, by assaying relevant phenotypes in a single sample?
Disclosure: Part 1: Cowen Healthcare, Honoraria, Forum Pharmaceuticals, Honoraria, Sunovion, Honoraria, ThinkNow, Inc., Honoraria, Lumos Labs, Honoraria, Snapchat, Honoraria, Elsevier, Honoraria, **Part 2:** Johnson & Johnson, Stock / Equity, Amgen, Stock / Equity, **Part 4:** Blackthorn, Grant.

Mini Panel
27. Connectivity and Psychosis: Emerging Data From a Neurodevelopmental Perspective

27.1 Neural Circuit Risk Pathways for Psychosis in Girls and Boys: Findings From a Population-Based Cohort

Aristotle Voineskos

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Background: Altered subcortical region volumes as well as cortico-thalamic-striatal-cortical (CTSC) circuit differences have been observed in schizophrenia. In this study, structural magnetic resonance imaging was used in a population-based sample of children and youth ($n = 1355$) including those with psychosis spectrum (PS) symptoms ($n = 350$) to promote early identification biomarkers.
Methods: All MRI data was acquired on the same 3T Siemens scanner. From T1-weighted images, subfields of the thalamus and striatum were identified using the segmentation tool MAGeT Brain. Correlations between these subfields and cortex-wide cortical thickness values were used to infer structural network connectivity. Boys and girls were analysed separately, since subregion volumes are sexually dimorphic during development.
Results: First, increased volumes for sensorimotor related striatal subregions ($t(614) = 3.21$, $pFDR = 0.003$) and sensory relay thalamic subregions ($t(614) = 3.71$, $pFDR = 0.0005$) were found in PS vs. nonPS boys. In contrast, multiple diversely connected anterior striatal subregions ($F(727) = 10.66$, $pFDR = 0.003$) and thalamic subregions ($F(727) = 6.95$, $pFDR = 0.01$) were found between PS and nonPS girls. For subcortical-cortical connectivity, a Fischer's r to z transformation revealed aberrant thalamic connections with the inferior frontal gyrus (IFG,RPS = -0.24, Rnon-PS = 0.12, $pFDR = 0.004$) and the insula (RPS = 0.15, Rnon-PS = -0.15, $pFDR = 0.02$) in boys, whereas striatal connections with the IFG (RPS = 0.13, Rnon-PS = -0.15, $pFDR = 0.06$) were implicated in girls.

Conclusions: Within the CTSC, these findings uncover potentially different psychosis vulnerability pathways in boys vs. girls. Finding these early indicators is a key strategy to provide insight into neural mechanisms underlying potentially sex-specific risk pathways for psychosis risk and treatment.
Disclosure: Nothing to Disclose.

27.2 Multimodal Evaluation of the Executive Network in Youth With Psychotic Spectrum Disorders

Katherine Karlsgodt

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Background: Executive function deficits are well established in individuals with schizophrenia, and due to their presence across illness phases and in unaffected relatives, are often considered to be an endophenotype for the disorder. Such deficits are difficult to treat, and have been associated with poorer long-term outcomes. However, beyond schizophrenia, the presence of executive dysfunction in individuals across the broader range of psychotic disorders has been less investigated. Here, we sought to examine the neural basis of executive function deficits across a sample of youth with a range of psychotic spectrum disorders.
Methods: 51 individuals with diagnoses across the psychosis spectrum (schizophrenia, schizoaffective, psychosis NOS, MDD with psychosis, bipolar with psychosis), and 53 age matched unaffected controls were evaluated with multimodal neuroimaging, cognitive testing, and clinical interviews. Participant ages ranged from 14-23, with the aim of assessing functional and structural differences present during the time period most commonly associated with illness onset.
Results: Our analyses included functional MRI, resting state MRI, and diffusion tensor imaging (DTI). Even in a sample with a wide range of symptom severity, we found multimodal disruptions in the central executive network (CEN). Patients showed task-based functional activation differences across the CEN during a Sternberg style item recognition task relative to controls, even when matched for performance. We then probed structural and functional connectivity of CEN networks. A set of fronto-parietal CEN regions of interest were functionally defined from the working memory task and applied to resting state fMRI data. An SVM analysis revealed that functional connectivity between the fronto-parietal nodes showed 70.29% classification accuracy for psychosis patients and controls. DTI analyses revealed that structural connections between these fronto-parietal nodes were also impaired.
Conclusions: Our evidence supports disruptions in the structure and function of the CEN in youth with early stage psychotic disorders, across a diagnostic spectrum. Thus, CEN function and working memory performance should be assessed in individuals not only with schizophrenia, but across the psychosis spectrum, and may be important targets for intervention.
Disclosure: Nothing to Disclose.

27.3 The Relationship Between DUP, Corticostriatal Connectivity, and the Outcome of Treatment of Early Phase Psychosis

Delbert Robinson

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Background: Duration of untreated psychosis (DUP), the period between the onset of psychotic symptoms and the initiation of antipsychotic treatment, has been consistently found to be a predictor of symptom outcomes for patients with early phase psychotic disorders. The neurobiological mechanisms underlying the association between DUP and outcome have been challenging to determine as DUP itself is influenced by environmental factors (e.g. access of treatment). We present novel data on the interaction between DUP, the type of treatment received and outcomes and on the relationship between DUP, corticostriatal connectivity and outcome.
Methods: The RAISE-ETP study compared 2 year outcomes between clinician-choice (CC) versus NAVIGATE, an integrated treatment program providing pharmacotherapy, individual therapy, family therapy and supported education/employment services, with 404 first episode subjects (mean age 23, 73% men) recruited from community treatment facilities in 21 US states.
We used seed-based resting state functional connectivity to examine the impact of DUP on corticostriatal circuitry with a separate cohort of 83 patients (mean age 22, 73% men) with early phase psychosis treated for 12 weeks with second generation antipsychotics. Mediation analyses examined a composite measure of corticostriatal connectivity on the relationship between DUP and treatment response.
Results: Mean DUP in the national RAISE-ETP cohort was 193 (SD = 262) weeks and the median DUP was 74 weeks. Participants in both conditions improved, but crucially there was differential improvement across conditions for short versus long DUP groups. Short ($<$ 74 weeks) and long ($>$74 weeks) DUP groups in CC and the long DUP group in the NAVIGATE condition improved to similar degrees but short DUP participants receiving NAVIGATE had significant and clinically meaningful greater change in total scores for both the Heinrichs-Carpenter Quality of Life (QLS) (treatment-by-time interaction, $p = 0.015$) and the PANSS (treatment-by-time interaction, $p = 0.016$).
Mean DUP was 102 weeks (SD = 77) and the median was 35 weeks in the imaging cohort. Thirty-one subjects were antipsychotic naïve at scanning; the median number of days on antipsychotics before scanning was 5. Forty-eight of the 83 subjects met predetermined response criteria. Longer DUP was significantly associated with poorer response rates ($p = 0.03$). Significant lower connectivity with longer DUP was found in 24 clusters across 12 seed regions reflecting connectivity of the striatum with a group of cortical regions. A principal components analysis revealed two significant factors. The first significantly predicted treatment response ($p = 0.02$). When the first factor was added as a mediator in the model of DUP and response, the effect of DUP on treatment response was no longer

significant ($p = 0.07$). The second factor was not related to treatment response.
Conclusions: Longer DUP is known to be associated with poorer treatment response and the RAISE-ETP outcomes suggest that besides globally predicting outcomes DUP may also predict differential response to treatment programs. Our imaging results suggest that the association between longer DUP and with poorer treatment response may be mediated by decreases in corticostriatal connectivity. Future studies using a longitudinal approach are indicated to examine the relationships between DUP, striatal circuitry and outcomes.
Disclosure: Part 1: Asubio, Consultant, Janssen, Consultant, Neurocrine, Consultant, Otsuka, Consultant, Shire, Consultant, **Part 4:** Otsuka, Grant.

Panel
28. The Adolescent Brain Cognitive Development (ABCD) Study: Goals, Methods, and Emerging Data

28.1 Assessment of Substance Use in a Nationally Representative Sample of 9 and 10 Year Olds

Mary Heitzeg

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Background: Adolescence is a time of increased risky behaviors, including initiation and escalation of substance use. One goal of the ABCD study is to examine the risk and protective factors influencing trajectories of substance use throughout adolescence and the impact of specific patterns of substance use on neurocognitive, health and psychosocial outcomes. Although the initiation of drinking and drug use typically begins during the teen years, data from state and community assessments suggest that youth may initiate first sipping or experimentation with substances in late childhood. Detailed data on substance use in pre-teens is sparse, however, as current national surveys begin assessment at ages 12-13 years. The ABCD study will be the first to provide detailed characterization of substance initiation, experimentation and use patterns in a large, diverse sample of 9 and 10-year olds across the US.
Methods: The ABCD study is planning to enroll 11,000+ children (ages 9-10) at baseline and follow them over ten years. Baseline assessments have been completed on 2,302 participants across 21 sites thus far. The substance use module assesses lifetime and recent quantity/frequency and patterns of use (including low-level use, first use, days of combined drug use) of all major drug categories. Important predictors of substance use are also captured, including peer use, intention to use, availability of substances, and parent rules about use. Additional risk and protective factors are captured in the culture and environment module of the ABCD protocol, including parental monitoring, family environment and conflict, school environment and neighborhood safety and crime. Baseline measures were selected to be developmentally-appropriate and gated to: 1) avoid exposing non- or low-using youth to substance use questions (using "heard of" questions in introduction); and 2) assess symptoms, craving and withdrawal in youth reporting use. Initial analyses are focused on comparisons between high-risk (44% of participants) and low-risk groups formed based on externalizing and negative affect symptoms.
Results: The majority of youth in the sample endorsed having heard of alcohol (97.5%), tobacco (94.4%) and marijuana (58.4%). The next most commonly "heard of" drug categories were prescription drug misuse (37.2%) and inhalants (27.8%). High-risk youth were more likely than low-risk youth to report having heard of marijuana (61.1% vs. 56.4%; $p = .025$) and inhalants (30.1 vs. 26.1%; $p = .034$), but not alcohol, tobacco or prescription drug misuse. Of youth having knowledge of alcohol, 26% reported having sipped alcohol in the past and 0.2% ($n = 5$) reported having a full drink; these rates did not differ by risk group. Of youth having knowledge of tobacco, 0.6% ($n = 14$) reported having tried a puff; this rate was significantly higher in high-risk than low-risk youth (1.4% vs. 0.08%; $p < .001$). High-risk youth also reported having friends who use alcohol and tobacco more than low-risk youth (alcohol: 4.4% vs. 1.9%, $p = .002$; cigarettes: 2.6% vs 1.2%, $p = .02$). Peer drinking was significantly associated with the endorsement of sipping ($p < .001$). Additional preliminary analyses will focus on associations between family, neighborhood and school environment and experimentation with substances.
Conclusions: The ABCD study will be able to provide detailed characterization of substance initiation, experimentation and use patterns in a large, diverse sample of 9 and 10-year olds. The collection of these data along with the assessment of risk and protective factors and psychological and neurobiological development over 10 years will allow the ABCD study to identify the effects of substance use trajectories on brain maturation, and behavioral, health, and psychological outcomes.
Disclosure: Nothing to Disclose.

28.2 Functional Neuroimaging Probes of Adolescent Neurodevelopment

Hugh Garavan

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Background: Adolescence is a period of substantial change in behaviors, attitudes, psychological and biological characteristics. Our understanding of the neurobiological development that accompanies these changes is very incomplete due, in part, to a shortage of large-sample, longitudinal studies that are broad both in numbers and sociodemographic diversity of their participants and in their breadth of assessments. In addition to a very extensive battery of psychological, biological, and genetic assessments, the ABCD study includes three functional neuroimaging task probes of psychological processes that are central to adolescent development. Furthermore, as adolescence is also a period in which mental health and behavioral problems often first emerge, the ABCD longitudinal design can help identify the antecedents and correlates of those behaviors. This information helps disentangle changes that may arise from those behaviors from those that preceded them and offers important clues into probable etiological mechanisms.

Methods: The 21 data acquisition sites of the ABCD study all employ multiband imaging on 3T platforms using acquisition protocols modeled on the HCP. The neuroimaging task battery assesses cognitive control, reward/reinforcement, working memory, and social processes. A motor inhibition STOP task assesses the cognitive control required to countermand a motor response and yields activation maps of both inhibitory control and error detection. A Monetary Incentive Delay task assesses reward and reinforcement processes yielding distinct activation maps for anticipation periods and outcomes with wins and losses of high and low value. An N-Back task assesses working memory abilities and contains both neutral and face stimuli. All tasks are assessed at baseline (ages 9-10) and every two years thereafter up to age 20. The sociodemographic richness and size of the ABCD sample ($n = 11{,}000+$) offers a unique and comprehensive insight into neurodevelopmental trajectories.
Results: By May 1, 2017, ABCD had recruited and tested 2,302 participants, making it the largest neuroimaging study yet conducted. The sample size is 54% male and diverse in race and ethnicity (56% White, 24% Hispanic, 7% African American, 3% Asian and 10% Other). Preliminary analysis of the neuroimaging tasks reveals high compliance by participants with performance measures as expected (e.g., performance declines with increasing NBack difficulty; increased accuracy on MID trials with wins/losses relative to neutral trials). The very high spatial and temporal resolution of the MRI acquisition allows surface-level analyses of the cortical data and anatomically-bounded high resolution volumetric analyses of the subcortical data thereby greatly increasing the accuracy of structural and functional neuroimaging metrics. Initial analyses will focus on cross-sectional comparisons of participants who are substance use naïve but are identified as being at elevated risk for substance use. Risk status is based on the presence of externalizing or negative affect symptoms as well as substance use in the home and a family history of substance use. Thus far, 44% of participants pass a relatively low threshold for high-risk status but analyses will focus on treating risk-status as a multi-dimensional continuous variable.
Conclusions: The ABCD study operates an open data model whereby this unique resource will be available to researchers globally ensuring that the myriad questions on adolescent neurodevelopment can be fully interrogated. The first data release of baseline assessments of approximately 5,000 participants will be in December 2017.
Disclosure: Nothing to Disclose.

28.3 An Early Examination of Examination of Functional Connectivity in the ABCD Study

Damien Fair

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Background: Adolescents and young adulthood is a unique period of development where an increase in risky behaviors, higher degrees of sensation seeking and impulsivity, greater sensitivity to rewards, and heightened reactivity to threat and punishment occur. In all aspects of development, in particular adolescents, a great deal of heterogeneity exists amongst typically and non-typically developing populations. In other words, particular characteristics may predispose certain subgroups of individuals more than others with a greater inclination toward risk. Importantly, our ability to understand this variance and how it relates to underlying brain has been limited, in part, because of the scarcity of samples large enough to tease out this variability. Within this context, the current research examines a "first look" at brain variability in the baseline scans of the ABCD study.
Methods: The ABCD study is planned to scan over 10,000 participants (9-10y) at baseline, and will be followed over the next 10 years into adolescents. From 21 unique data acquisition sites, 2,302 participants have been currently scanned, obtaining 15-20 minutes of resting-state fMRI data. The current study will apply new computational tools that integrate machine learning and graph theory to examine functional heterogeneity in the current sample.
Results: The very high spatial resolution and extended resting-state fMRI data has allowed for precise connectivity measurements across this large sample. New approaches to handle motion related artifacts in data with the high sampling rate of ABCD have been utilized. These include real-time motion monitoring (FIRMM), and specified filters to the motion estimates. Preliminary analyses suggest consistent connectivity patterns in the sample as viewed in the literature, highlighting the fidelity of the data. Initial heterogeneity analysis will focus on supervised comparisons of high and low risk adolescents. Thus far, 44% of participants might be high-risk. A secondary analysis will examine heterogeneity in an unsupervised fashion combing machine learning and graph theory on the functional data.
Conclusions: Research using human neuroimaging has revealed that much heterogeneity exists across individuals in brain physiology, a reality that complicates characterizing typical brain trajectories, as well as the multiple pathways that may confer risky behavior in adolescents. Here we examine and introduce new analytical tools that combine state-of-the-art methods for quantifying how spatially disparate brain regions coordinate activity (i.e., "brain connectivity maps") with an algorithm for classifying humans based on this brain physiology as assessed with functional MRI. The ABCD study, due to its size and breadth, provides an opportunity for the community to tackle these difficult issues.
Disclosure: Nothing to Disclose.

28.4 Assessing Psychopathology in the Adolescent Brain and Cognitive Development Study: Goals, Methods and Emerging Findings

Deanna Barch

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Background: The ABCD is a large and unprecedented study of children that will inform our understanding of the environmental, genetic, neurobiological and behavioral factors that promote health and which put children at risk for both physical and mental-health problems. Consequently, the ABCD incorporates a broad range of measures assessing

predictors and outcomes related to mental health in children and later in adolescence and young adulthood. We strove to develop a battery that would address a range of domains within the time constraints imposed by the need to assess many different areas within the ABCD.
Methods: A number of different considerations went into the choice of measures for the ABCD mental health battery, including: 1) assessing constructs important for understanding both healthy brain development and risk for substance use and psychopathology; 2) using assessments appropriate for 9- and 10-year-old children; 3) using measures that would "stand the test of time" in a longitudinal study; 4) strong psychometric evidence for reliability and validity; and 5) using measures that were also being used in other large-scale studies and/or recommended as common data elements by NIH initiatives. Parent reports include a diagnostic interview about the child using the new computerized version of the Kiddie Schedule for the Assessment of Schizophrenia and Depression (KSADS DSM 5), the Child Behavior Check List, and the Mania scale from the Parent General Behavior Inventory. Parents also report on their family history of psychopathology and their own mental health using the Achenbach Self-Report Inventory. Child reports include assessments of mood disorders, separation anxiety, social anxiety, generalized anxiety, sleep and suicidality using the KSADS DSM 5 (with research assistant support), as well as psychosis (Prodromal Questionnaire Brief Version modified for children), impulsivity (the Urgency, Perseverance, Premeditation, and Sensation Seeking - UPPS - scale), and the behavioral inhibition and behavioral activation scales.
Results: We analyzed the data from the first 1800+ children enrolled in the ABCD study to examine the relationship between risk status (children recruited at high and low risk for substance use) and initial psychopathology as assessed by both parent report and child report. These analyses demonstrated that children thought to be at high risk were rated by their parents as having higher symptoms of mania and as well as greater internalizing and externalizing symptoms (all ps $<.001$). Children at high risk for substance use rated themselves as having higher levels of both positive and negative urgency/impulsivity (ps $<.002$) as well as higher behavioral inhibition ($p = .045$). Further, children's self reports of psychosis were significantly related to parent's reports of hallucinations and delusions (ps $<.003$), as well as to parent's reports of depression and anxiety in children (ps $<.01$). For self-harm assessment, 9.8% of children reported current or past suicidal ideation or wishing they were better off dead, which was significantly correlated with child report of negative urgency on the UPPS impulsivity scale ($r = .14$, $p < .001$) and parent report of both internalizing ($r = .12$, $p < .001$) and externalizing ($r = .11$, $p < .001$) symptoms in the child.
Conclusions: These data illustrate the feasibility of in depth assessment of psychopathology in the ABCD study, speak to the initial validity of the designation of high risk for a subset of children, and support the validity of assessing pre-clinical symptoms of psychosis in this young child population. Our ability to validly assess early signs and symptoms of psychosis will be particularly important for our ability to track the dynamic relationship between the emergence of psychosis and the use of substances such as a marijuana over the course of development.
Disclosure: Nothing to Disclose.

Panel
29. Effects of Adversity on Brain Development and Mental Health: Converging Evidence From Rodents, Monkeys and Humans

29.1 Neurobiology of Infant Trauma: Specific Categories of Adversity Produce Diverse Neurobiological Signatures and Outcome

Regina Sullivan

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Background: We have known for decades that the timing and type of early life trauma produces vulnerability to myriad psychiatric disorders, with brain imaging studies and animal models indicating this is due to disruption of brain development. Importantly, more recently clinical evidence suggests that trauma experienced within a social context appears to have more profound neurobehavioral effects. Here we model this in rodents and questions whether the presence of the caregiver during the trauma impacts the brain's immediate response to trauma, as well as later infancy neurobehavioral response.
Methods: From 8 to 12 days old, rat pups were reared by an abusive mother (induced by reduced bedding for nest building). To assess what aspects of mother-infant dyad resulted in neurobehavioral abnormalities, we also gave pups mild electric shock (1 sec 0.5mA shock to the tail 8 times, every 4 min to mimic pain of abuse) with or without the mother present. Responses to shock and mother, as well as the amygdala were assessed during treatment. Testing occurred when pups were 13-14 days old, when the amygdala and social behavior with the mother were assessed. Techniques used to assess the amygdala during treatment and testing included LFP electrophysiology, c-Fos, and pharmacological manipulations of dopamine.
Results: Using naturalistic observations in the nest, all pups show attachment to the caregiver. However, in a more challenging test with an anesthetized mother, which assess only the pups contribution to interactions, shock-mom pups and rearing with a maltreating mother exhibited strong interactions with the mother but reduced responses compared to controls (no shock, control rearing): including reduced interaction with the mother, reduced response to the maternal odor, amygdala hyperactivity (fos) and reduced maternal control of pup LFP oscillations compared to controls. Pups receiving shock without the mother showed minimal disruption of social behavior with the mother but showed greater threat responses.
Conclusions: Shock with the mother and maltreatment from the mother both produce changes in social behavior and implicates the amygdala as partially responsible for the behavioral deficits. This effect of shock without the caregiver produced a distinct early onset anxiety-like phenotype.
Disclosure: Nothing to Disclose.

29.2 Adverse Maternal Care Affects the Development of Emotional and Stress Regulation and Underlying Neurocircuits in Nonhuman Primates: Associations With Telomere Length Shortening

Mar Sanchez

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Background: Poor maternal care is an early adverse experience linked to psychopathology and alterations in stress physiology. Using a translational macaque model of infant maltreatment, we have reported its developmental impact on social behavior, emotional and stress reactivity during infancy, as well as in brain systems that control these functions, particularly increased amygdala volumes. Here we study its long-term effects during the juvenile and adolescent periods, particularly on (1) emotional regulation using a fear and safety learning paradigm, (2) hypothalamic-pituitary-adrenal (HPA) axis function and (3) prefrontal(PFC)-amygdala connectivity critical for emotional and stress regulation. We also investigated molecular mechanisms that could link adversity exposure with poor neurobehavioral and physiological outcomes, in particular accelerated telomere length (TL) shortening.
Methods: Forty-two infant rhesus monkeys (Macaca mulatta) were studied using a unique crossfostering design with random assignment at birth to a competent (Control) or maltreating (MALT) mother to disentangle the effects of experience from heritability. In vivo, longitudinal, neuroimaging approaches were used to examine the development of PFC-amygdala functional connectivity (FC) using resting state functional MRI (rsfMRI). Longitudinal measures of the stress hormone cortisol and TL shortening -a molecular index of stress exposure and biological aging-, were also collected. And a fear-potentiated startle paradigm that assesses both fear and safety learning (AX+/BX-) was used to examine different aspects of emotional regulation during adolescence.
Results: MALT animals showed elevated levels of HPA axis activity throughout the infant and juvenile periods, demonstrated by higher cortisol secretion throughout the day and hair cortisol accumulation during the first months of life than Controls. Long-term impact of early adversity was also evidenced by increased fear-potentiated startle and impaired discrimination between safety and fear signals during the AX +/BX- paradigm. Weaker PFC-amygdala FC in MALT than Control animals detected during infancy and the juvenile period could explain the alterations reported in emotional and stress regulation. Furthermore, the maturation of some PFC-amygdala circuits seemed accelerated, which was paralleled by accelerated cellular aging (accelerated TL shortening). Shorter TL during infancy also predicted higher HPA axis activity at later ages, during the juvenile period.
Conclusions: Altogether, our findings suggest that infant maltreatment leads to weakened PFC-amygdala connectivity during primate development, which could underlie the increased activity of stress neuroendocrine systems and impaired regulation of fear responses. Similarly to reports in humans, early adversity accelerates the maturation of emotional regulatory neurocircuits in infant monkeys, in parallel to signs of accelerated cellular aging (i.e. TL shortening). This molecular impact predicts elevated activity of stress neuroendocrine systems. In sum, PFC-amygdala circuits and telomeres seem very sensitive to primates' early experience, potentially serving as both vulnerability and adaptive mechanisms shaping developmental trajectories.
Disclosure: Nothing to Disclose.

29.3 Differential Effects of Abuse and Neglect on Brain Development and Psychopathology in Adolescents Growing up in Poverty

Christopher Monk

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Background: In the U.S., 20% of children are growing up in poverty. These children face high risk for psychopathology, which often lasts a lifetime and perpetuates low socioeconomic status to future generations. Children of low-income families experience greater chronic stress, which may allow poverty to become biologically embedded by altering brain function. However, chronic stress encapsulates a heterogeneous set of events. To gain a more mechanistic understanding of how poverty leads to negative outcomes, it is crucial that the field identify how specific categories of poverty-related adverse events alter brain function and contribute to specific forms of psychopathology (McLaughlin, Sheridan & Lambert, 2014). Two prevalent and pernicious forms of poverty-related adversity are abuse and neglect. We hypothesized the following: 1) abuse would differentially impact threat circuitry (amygdala habituation) along with both anxiety and depression symptoms; 2) neglect would differentially affect reward circuitry (nucleus accumbens activation) and only depression symptoms.
Methods: We examined adolescents from The Fragile Families and Child Wellbeing Study (FFCWS), an ongoing study of children born to predominantly low-income families. The FFCWS is a population-based representative sample of youth growing up in U.S. cities. Children from the FFCWS were studied at birth, 1, 3, 5 and 9 years. We assessed 237 15-16 year olds from the FFCWS who are growing up in Detroit, Toledo and Chicago. To date, we have analyzed useable fMRI data from 118 adolescents. Full diagnostic assessments (KSADS, SCID) of the teen and mom as well as symptom measures of anxiety and depression were acquired. The Child Trauma Questionnaire provided abuse/neglect information. We collected fMRI data using an emotional faces task with threatening (angry and fear faces) and intrinsically rewarding social stimuli (happy faces). fMRI analyses utilized small volume corrections from regions of interest.
Results: 46% of teens experienced clinically significant maltreatment. Abuse and neglect were correlated ($r = .341$, $p < .001$), but not collinear (variance inflation factor = 1.000), indicating that abuse and neglect can be distinguished. More abuse predicted less right amygdala habituation to threat faces ($t = 3.09$, $p = .044$). Neglect did not relate to amygdala habituation. Greater abuse related to more anxiety symptoms ($r = .36$, $p < .01$) and depression symptoms ($r = .34$, $p < .001$). More neglect negatively related to less nucleus accumbens activation selectively to happy faces ($t = 3.27$, $p = .027$). Abuse did not relate to

nucleus accumbens activation. More neglect related to more depression symptoms (r = .23, $p = 003$), but not anxiety.
Conclusions: Abuse and neglect differentially affected brain function and psychopathology symptoms: 1) Abuse impacted threat circuitry as well as both anxiety and depression symptoms, but not reward circuitry; 2) Neglect affected reward circuitry and depression symptoms, but not threat circuitry or anxiety symptoms. Functional connectivity and longitudinal variables of abuse as well as neglect will be incorporated from the FFCWS to examine effects of developmental timing on threat and reward networks. These findings identify routes through which specific conditions that are prevalent in poverty alter brain function and increase psychopathology in adolescents.
Disclosure: Nothing to Disclose.

29.4 Separable Dimensions of Childhood Adversity Have Distinct Associations With Cognitive and Neural Function, Increasing Risk for Psychopathology

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Background: Numerous studies have demonstrated that childhood adversity is associated with increased risk for psychopathology, however, neurodevelopmental pathways underlying this risk remain poorly understood. The most commonly proposed model accounting for this association is a cumulative risk model, where exposure to increasing numbers of adversities is linked with increasing psychopathology. The cumulative risk model has been pivotal in identifying who among the children who are exposed should be treated, however, this model implicitly assumes that the same mechanism accounts for the impact of markedly different forms of adversity on risk for psychopathology. We have proposed a conceptual model which posits instead that childhood adversity can be deconstructed into at least two underlying dimensions—deprivation and threat—that are associated with distinct neural and cognitive consequences. The deprivation and threat model argues that deprivation- a lack of cognitive stimulation and learning opportunities- is associated with poor cognitive function, including cognitive control. In contrast, threat – the presence of trauma, the fear that oneself or a close other is in physical danger - is associated with difficulties in emotion regulation, including fear learning. We further propose that both threat or deprivation will be associated with psychopathology but that these exposures will increase risk through different neural and cognitive pathways.
Methods: We examine this hypothesis at multiple levels across three distinct samples. In Study 1 we examine the independent association of deprivation (low parental education and child neglect) and threat (physical, sexual, and emotional abuse, and community violence) with parent-reported problems with EF in adolescents (N = 169; 13-17 years) on the BRIEF. In Study 2, we examine the independent association between deprivation (low parental education) and threat (abuse exposure) with working memory (WM) performance and neural recruitment during high vs. low WM load encoding trials among adolescents (N = 51, 13-20 years). In Study 3 we use structural equation modeling in a large (N = 585) sample of children drawn from the Child Development Project (CDP; Dodge, Bates, & Pettit, 1990), a multi-year longitudinal project that has followed children for over a decade. Here we examine the indirect association between deprivation (HOME interview) and threat (maltreatment) at age 5 and 6 years and externalizing psychopathology at age 17 years through verbal abilities measured at 14 years.
Results: Across all three studies we demonstrate that deprivation predicts complex cognitive function, in particular, parent report of executive function, working memory task performance, and verbal abilities controlling for robust threat exposure in the form of maltreatment. In the final study, we demonstrate that the indirect impact of deprivation on externalizing psychopathology is through verbal abilities in a longitudinal study controlling for significant maltreatment exposure.
Conclusions: Together, these findings constitute strong preliminary evidence for a novel model of the neurodevelopmental consequences of childhood adversity. This data suggest that different dimensions of adversity independently impact different underlying neurocognitive constructs and that deprivation, specifically, may increase risk for externalizing psychopathology because of its impact on complex cognitive functions such as verbal abilities and executive function.
Disclosure: Nothing to Disclose.

Panel
30. Prepronociceptin Circuits in Motivational States

30.1 A Peri-VTA Prepronociceptin Neuronal System That Gates Motivation

Michael Bruchas

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Background: Nociceptin/Orphanin FQ Opioid Receptor (NOPR) and its endogenous ligand, nociceptin (NOC) are widely distributed throughout the brain and have been demonstrated to have a role in mediating pain, stress, anxiety, feeding, and reward behaviors. In particular, systemic NOPR stimulation has been shown to inhibit dopamine neuron firing and reduce drug-reward behavior. However, the endogenous sources and natural role of nociceptinergic circuits within these reward behaviors are not understood.
Methods: Here we used a novel knockin prepronociceptin (PNOC)-Cre mouse driver line, and conditional knockout mouse for the receptor, combined with behavioral measures of motivation, optogenetics, chemogenetics, fiber photometry, and retrograde tracing techniques. We discovered a population of prepronociceptin neurons located within the paranigral and paraintrafascicular nuclei of the ventral tegmental area (peri-VTA) that regulate motivated behaviors (progressive ratio task for sucrose and aversion).
Results: We found that these prepronociceptin neurons have projections onto dopamine cells and selective ablation of these cells enhanced sucrose seeking under fixed ratio three and progressive ratio operant tasks. In addition, we found that chemogenetic stimulation of peri-VTA nociceptin cells and

optogenetic photo-stimulation of peri-VTA prepronociceptin terminals reduced operant responding in the progressive ratio task and causes conditioned place aversion. These findings identify a previously unknown population of nociceptin-containing neurons, sitting just ventral to the VTA that are positioned to tonically suppress reward motivation via dopamine cell inhibition. To assess the functional activity of this prepronociceptin cell population during fixed and progressive ratio operant tasks, we used fiber photometry to record activity of prepronociceptin neurons during operant behavior. Our data suggest that these neuronal terminals are engaged (calcium transient activity) in specific time points during the reward consumption, and presentation.
Conclusions: Understanding how this discrete peri-VTA prepronociceptin containing nucleus may be critical to the regulation of motivation and reward seeking behaviors could provide insight into behaviors dysregulated during motivational states such as depression and addiction.
Disclosure: Nothing to Disclose.

30.2 Central Amygdala Prepronociceptin-Expressing Neurons Mediate Palatable Food Consumption and Reward

Thomas Kash

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Background: Food palatability is one of many sensory factors that drive food consumption, and the drive to feed for its rewarding properties is a key contributor to obesity and binge eating.
Methods: We have identified a population of cells expressing prepronociceptin, the gene encoding the opioid-like neuropeptide nociceptin, in the central amygdala (PnocCeA) that are activated by palatable food consumption.
Results: Ablation or chemogenetic inhibition of these cells reduces palatable food consumption. Further, ablation of PnocCeA cells reduces the increase bodyweight and adiposity accompanied by continuous high fat diet access. PnocCeA neurons project to the vBNST, PBN, and NTS, and activation of axons in the PBN and NTS produces reward behavior. These data suggest that the PnocCeA hindbrain network is critical for promoting the reinforcing and rewarding properties of palatable food.
Conclusions: This data is the first to demonstrate a genetically defined population of CeA neurons that respond to highly palatable foods and open the door to understanding how the CeA contributes to pathological feeding disorders.
Disclosure: Part 1: BlackThorn Therapeutics, Grant.

30.3 Nociceptin Neurons in the Bed Nucleus of the Stria Terminalis Regulate Anxiety

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Background: The neuropeptide Nociceptin/Orphanin FQ (N/OFQ) has been traditionally implicated in pain sensitivity. Prepronociceptin (Pnoc) is the genetic precursor of N/OFQ and is highly expressed in a subset of neurons within the bed nucleus of the stria terminalis (BNST). This region is essential for regulating both positive and negative motivational states, and due to the role of Pnoc neurons in pain sensitivity, we hypothesize they are involved in regulating negative motivational states within BNST.
Methods: We generated a Pnoc-ires-Cre mouse line that expresses Cre in Pnoc+ neurons. Using in situ hybridization (ISH) and immunohistochemistry (IHC), we characterized these neurons within anterodorsal BNST (BNST-Pnoc neurons). Next, we used patch-clamp electrophysiology to assess local connectivity. We then monitor single-cell dynamics of BNST-Pnoc neurons combining Cre-dependent expression of the genetically encoded calcium indicator GCaMP6s with head-mounted miniature microscopes. We used this to track the activity dynamics of BNST-Pnoc neurons while mice explore the elevated plus maze (EPM) or during exposure to predator odor (2,3,5-trimethyl-3-thiazoline; TMT). Using optogenetic tools, we then modulated these neurons during the EPM and a real time place preference assay (RTPP).
Results: Using ISH, we find that BNST-Pnoc neurons overlap with BNST-CaMKII (96%) and BNST-Vgat (97%) neurons, but less with BNST-Vglut2 (11%) neurons. Using IHC, we find BNST-Pnoc neurons have less overlap with BNST-Som (8%) or BNST-PKCδ (19%) neurons. Anatomical analysis showed that BNST-Pnoc neurons project locally and distally, predominantly to medial amygdala and medial preoptic area. Electrophysiology experiments reveal that BNST-Pnoc neuron excitation induces inhibitory post-synaptic currents in both Pnoc- (60%, $n=37$) and Pnoc+ (31%, $n=26$) neurons, but these cells do not elicit excitatory post-synaptic currents (0%, $n=63$). Using genetically encoded calcium indicators, we find that BNST-Pnoc neuronal activity significantly increases when mice transition from closed to open arms in the EPM ($p=0.01$) and significantly decreases with transitions from open to closed arms ($p=0.01$; $n=169$). Photoactivating BNST-Pnoc neurons with channelrhodopsin increases avoidance of open arms in the EPM ($p=0.0005$, $n=6$-7 per group), whereas photosilencing with halorhodopsin decreases avoidance of the open arms ($p=0.03$, $n=6$-9 per group). In contrast, photoactivation of these neurons during RTPP (in a non-threatening environment) does not induce avoidance behavior ($p=0.35$, $n=8$-10 per group). Furthermore, mice exposed to a distinct anxiogenic environment (TMT in corner of home cage) also show significant increases in both avoidance behavior ($p=0.009$) and activity of BNST-Pnoc neurons ($p=0.009$). In contrast, we find that exposure to peanut oil increases approach behavior ($p=0.03$) but does not alter activity of BNST-Pnoc neurons ($p=0.23$).
Conclusions: Together our findings demonstrate that BNST-Pnoc neurons consist of a GABAergic population necessary and sufficient to drive negative motivational states, particularly when mice are in threatening environments. Future studies should address how monosynaptic inputs modulate the activity of BNST-Pnoc neurons and also elucidate the genetic identity of Pnoc- neurons within BNST that receive input from BNST-Pnoc neurons to fully understand the underlying circuitry. Moreover, our data strongly suggest that targeting BNST-Pnoc neurons may be a novel and

potentially effective target for patients suffering from anxiety disorders.
Disclosure: Nothing to Disclose.

30.4 Behavioral and Physiological Characterization of BTRX-246040, a Novel Nociceptin Receptor Antagonist

Tanya Wallace

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Background: The nociceptin/orphanin FQ receptor (NOPR) is a G-protein coupled receptor with widespread expression throughout the brain that has been implicated in mediating stress, mood and feeding behaviors. Although the biochemical and physiological mechanisms underlying the role of NOPR in these functions is an area of active investigation, our current understanding suggests the development of selective receptor antagonists may be a promising approach for treating disorders associated with the aforementioned behaviors. Accordingly, we have identified BTRX-246040 (aka LY2940094), a selective and potent (Ki = 0.1 nM) NOPR antagonist with no significant cross-reactivity across a wide array of receptors, ion channels and kinases at physiologically relevant concentrations. BTRX-246040 is orally bioavailable and brain penetrant and exhibits centrally-mediated activity in vivo. Evidence from clinical studies has suggested BTRX246040 can affect mood in depressed patients (Post et al., 2016), and may have potential for treating alcohol-use disorder. Currently, we are extending the characterization of BTRX-246040 to better understand the role of NOPR in circuits underlying neurobehavioral disorders, and the results of these studies will be discussed.
Methods: We will use a combination of cellular, physiological and behavioral approaches to evaluate NOPR function using BTRX-246040. Specifically, we will: 1) investigate feeding behaviors using a binge eating paradigm in which whole brain activity will be measured by c-fos expression in nociceptin reporter mice treated with a high fat diet in the presence and absence BTRX-246040. Studies will be continued to identify the cellular and biophysical mechanisms of NOPR modulation of neurons in these brain regions using whole-cell neuronal recordings in brain slices from these mice; 2) characterize NOPR function within ventral tegmental area (VTA) neurons using whole cell recordings to determine if both dopamine and non-dopamine neurons in this region are sensitive to nociceptin and antagonism with BTRX-246040; and, 3) evaluate the effects of NOPR antagonism in paradigms of spatial working memory and decision making using behavioral and neurophysiological approaches in Rhesus macaques.
Results: The results of these studies will 1) aid in defining the brain region(s) that contribute to NOPR antagonist-mediated reduction of highly palatable food in models of binge eating disorder, and define the cellular and biophysical mechanisms of NOPR signaling in these brain regions. 2) identify the population or sub-population of midbrain ventral tegmental dopamine neurons involved in BTRX-246040 function. 3) characterize the effects of antagonizing the NOPR on cortical cognitive function.
Conclusions: The data from these studies will extend our understanding of the cellular, biophysical and behavioral effects of the novel and selective NOPR antagonist, BTRX-246040, in circuits underlying neurobehavioral disorders.
Disclosure: Part 1: BlackThorn Therapeutics, Stock / Equity, **Part 5:** BlackThorn Therapeutics, Employee.

Panel
31. D1 Dopamine Receptors: Novel Non-Catechol Agonist, Biased Signaling, Circuit Plasticity and Therapeutic Potential

31.1 Characterization of Novel Non-Catechol D1 Agonists are Orally Bioavailable and Have a Unique Signaling

Rouba Kozak

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Background: Selective activation of D1 receptors (D1R) is an attractive therapeutic strategy for many indications but efforts were historically hampered by rapid metabolism of all known selective D1R agonists. We recently discovered a novel series of non-catechol D1R agonists that are potent, highly selective, orally bioavailable, and exhibit excellent pharmacokinetic properties.
Methods: Putative compounds were characterized using multiple biochemical assays. Further in vivo testing in rats and monkeys was done using a variety of assays designed to assess neurotransmitter release (microdialysis), working memory (Ketamine/RAM, cTUNL, DSR) and motor performance (6-OHDA rotation and MPTP dyskenesia). In a final step, human clinical studies were performed to assess the pharmacodynamic response of this response.
Results: This non-catechol class of ligands strongly activates Gs-adenylyl cyclase-cAMP signaling but unlike the traditional catechol agonists, it does not trigger the recruitment of β-arrestin, a key mechanism leading to desensitization. At a functional level, similar to the catchols, these novel non-catechol agonists increase locomotor activity in mice and potentiate prefrontal cortex acetylcholine release in rats. Furthermore, working memory impairments induced by ketamine or high memory load are reversed in both rats and NHPs. The extent of this effect depends on the intrinsic activity of the selected compounds as well as the age of the animals. Finally, in the 6-OHDA rat and MPTP-treated NHP animal models of Parkinson's disease, the novel non-catechol D1R agonists reversed parkinsonian disabilities without significantly increasing dyskinesia and this response was maintained even after repeated dosing. Finally, human studies demonstrate a pharmacokinetic and safety profile
Conclusions: All together, this finding suggests that non-catechol class will allow finally testing of the therapeutic promise of this mechanism for diseases involving impaired or reduced dopaminergic signaling
Disclosure: Part 1: Pfizer, Inc., Employee, **Part 2:** Pfizer, Inc., Employee, **Part 3:** Pfizer, Inc., Employee, **Part 4:** Pfizer, Inc., Employee, **Part 5:** Pfizer, Inc., Employee.

31.2 The Novel D1R Agonist, PF-3628, Enhances Prefrontal Cortal Neuronal Firing in Aged Rhesus Monkeys

Min Wang

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Background: Dopamine D1 receptor (D1R) agonists have been considered a potential treatment for dorsolateral prefrontal cortical (dlPFC) cognitive deficits in schizophrenia. For example, D1R stimulation may lead to phosphorylation of NMDA receptors that maintains them within the post-synaptic density. However, treatment has been hampered by the inverted U dose-response of available D1R agonists, where higher doses have been shown to suppress dlPFC neuronal firing, reduce neuronal representation, and cause D1R internalization. In this study, we are tested a novel, non-catechol D1R agonist, PF-3628, in aged rhesus monkeys with naturally-occurring depletion of dopamine and reductions in dlPFC neuronal firing during a spatial working memory task.
Methods: The research utilized single unit recordings from dorsolateral prefrontal cortex in aged monkeys performing a working memory task coupled with iontophoresis of drug. We iontophoretically applied the novel D1R agonist onto Delay cells in the dlPFC of primates engaged in the oculomotor delayed response task, and compared results to a traditional D1R agonist, SKF81297.
Results: We found that iontophoresis of PF-3628 at 10-25nA strongly enhanced the delay-related firing of dlPFC neurons, and also enhanced their spatial tuning. These effects of PF-3628 were strikingly different from traditional D1R agonists, which sculpt and suppress dlPFC neuronal firing.
Conclusions: These results suggest that novel, non-catechol D1R agonists may provide promise as a new treatment strategy for dlPFC cognitive deficits in mental illness.
Disclosure: Nothing to Disclose.

31.3 In Vivo Characterization of an Agonist Dopamine D1 Receptors Tracer [18F] PF-8477 ([18F]-MNI-968) in Human and in Non-Human Primates

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inviCRO, New Haven, Connecticut, United States

Background: D1 receptors, which couple to inhibitory G-proteins, have been shown to regulate neuronal growth and development, mediate some behavioral responses. Its function has been shown to be altered in both neurologic and psychiatric disorders. To date, there is a lack of agonist PET tracers for the D1 receptors labeled with 18F with relevance in clinical studies. We report the evaluation in non-human primates of [18F]MNI-968 (PF-8477), a novel PET radiotracer of the D1 receptors.
Methods: Four brain PET studies, 2 baselines and 2 blockade studies using PF-2562, a D1 partial agonist compound, were conducted for 90 min in two rhesus monkeys with [18F] MNI-968 (169 ± 31 MBq). PF-2562 was administered at the same dose level for both monkeys as a bolus followed by a 2-hour infusion, with [18F]MNI-968 administered 30 min into the infusion.
Additionally, three brain PET studies were conducted over 180 min (317 ± 49 MBq) in 7 healthy human volunteers (4 test/retest and 3 test).
PET data were modeled with 2-tissue compartmental model (2T), Logan graphical analysis (LGA), and non-invasive Logan graphical analysis (NI-LGA) with cerebellar cortex as reference region to estimate total distribution volume VT, and binding potential BPND.
For the blockade studies in rhesus monkeys, occupancy was estimated from BPND at baseline and post blockade.
Results: In rhesus monkeys, [18F]MNI-968 ([18F]PF-8477), penetrated the brain with a peak whole-brain uptake up to ~3% of the injected dose at ~6 min post injection and showed a fast washout. The highest signal was found in the caudate, putamen, with moderate extrastriatal uptake. The lowest signal was in the cerebellum. BPND values were up to ~1.4 in the putamen. All three quantification methods (2T, LGA and NI-LGA) were in excellent agreement, with a similar estimated D1 receptors occupancy of PF-2562 of ~40% for both monkeys in the caudate and putamen.
In human, [18F]MNI-968 kinetics appeared to be faster compared to non-human primates, with a BPND in the putamen of ~0.8. Initial measurement of test-retest reproducibility was $\leq 7\%$ for BPND in the striatal regions.
Conclusions: Our work showed that [18F]MNI-968 ([18F] PF-8477), is a promising agonist PET radiotracer for imaging D1agnist receptors that can be quantified non-invasively. Studies are currently ongoing both in non-human and human primates to further characterize the tracer.
Disclosure: Part 1: MNI holdCo, Stock / Equity, MNH, Stock / Equity, Lundbeck, Consultant, inviCRO, Employee, **Part 2:** inviCRO, Employee, **Part 5:** inviCRO, Employee.

31.4 Dopamine D2/D3 Receptor Blockade Decreases Risk Aversion Through Linearization of Value and Probability Processing

Philippe Tobler

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Background: Many everyday decisions involve risk and require balancing the potential gains and losses with the probabilities that they will occur in different choice options. The dopaminergic system has been implicated in the processing of value under risk and a number of models posit that the balance of D1- and D2-receptor-mediated activity describes an individual's risk attitude. However, at least in humans these models remain largely untested and the precise effects of shifting D1/D2-balance on the components of risk aversion unclear.
Methods: We used the D2/D3 receptor antagonist amisulpride while participants made choices between risky options. Each choice provided a trade-off between risk and expected value. Using a novel model fitting procedure, we concurrently estimated the three parameters that define individual risk attitude according to an influential theoretical account of risky decision making.
Results: Compared to placebo, D2 receptor blockade resulted in more frequent choice of higher risk and higher expected

value options. The model fitting analysis revealed that the observed reduction in risk aversion under amisulpride was driven by increased sensitivity to reward magnitude and decreased distortion of outcome probability, resulting in more linear value coding.
Conclusions: Our data suggest that different components that govern individual risk attitude are under dopaminergic control, such that a D1-dominated state facilitates risk taking and expected value processing.
Disclosure: Part 4: Pfizer, Inc., Grant.

Mini Panel
32. Neuroinflammation, Neural Function, and Translational Neuroimaging: Establishing a Window to the Brain

32.1 Cognitive and Neurostructural Consequences of Cancer Treatments

Leah Pyter

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Background: Impaired cognitive function in cancer survivors is a common problem and typically attributed to the negative effects of cancer treatments on the brain. For example, approximately 90% of breast cancer patients survive and yet a broad range (17-60%) is plagued by persistent cognitive impairments (e.g., executive function, memory, and processing speed). Beyond diminishing quality-of-life, these cognitive impairments increase healthcare costs, morbidities, and ultimately, mortality. The underlying mechanisms by which cancer treatments cause "chemobrain" remain unspecified, however deteriorations in brain microstructure, correlated with poor cognitive performance, have been identified as a possibility. Specifically, cancer treatments reduce brain white and/or gray matter in the cortex and corpus callosum. The hypothesis for this project is that systemic taxol chemotherapy treatment impairs cognitive functions and induces long-term white matter damage using a novel tumor-resected mouse breast cancer survivor model.
Methods: To model typical breast cancer survivors, all female Balb/c mice were ovariectomized and a single mammary tumor was induced using murine 67NR cancer cells. Because the tumors are non-metastatic, after 2.5 weeks of palpable growth (tumors ~ 1 cm3), all tumors were surgically resected using a modified radical mastectomy procedure. Half of the mice received paclitaxel chemotherapy (30 mg/kg i.p.; every other day for 6 cycles); the other half received vehicle. All mice underwent brain diffusion tensor imaging (DTI), which was analyzed to assess diffusional properties of axons, either immediately after the last chemotherapy treatment or 2.5 months later. After imaging, brain immunohistochemistry was performed to measure oligodendrocyte and myelin biology. An additional cohort of mice underwent cognitive behavioral testing at the same time points.
Results: Immediately after the final dose, chemotherapy statistically significantly altered multiple aspects of anisotropy as assessed using DTI images in widespread brain regions. For example, taxol chemotherapy increased fractional anisotropy in the hippocampus, hypothalamus, thalamus, brainstem, and cerebellum, and inversely decreased the apparent diffusion coefficients relative to vehicle controls. Histologically, chemotherapy disrupted myelin integrity in the corpus callosum. These white matter imaging and histological changes were virtually resolved 2.5 months after chemotherapy. Corresponding cognitive analyses are currently underway.
Conclusions: Moderate taxol chemotherapy treatment is capable of inducing acutely overt brain white matter imaging and histological changes. Using models to relate imaging biomarkers to underlying pathology and cognitive changes can be translated clinically to identify patients at risk for developing cognitive deficits and to monitor progress or treatment efficacy of chemotherapy-induced brain changes.
Disclosure: Nothing to Disclose.

32.2 Diet-Induced Inflammation is Associated With Reward Neuroadaptations and Disinhibited Eating in Female Monkeys

Vasiliki Michopoulos

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Background: The availability of highly palatable, calorically dense diets (CDD) with increased amounts of saturated fats and sugar, promotes increased appetite, and is a significant contributor to obesity. CDD consumption induces dopamine (DA) release and activates rewards pathways similar to what has been described in drug addiction. Decreases in DA 2 receptor (D2R) levels and decreased functional connectivity in corticostriatal regions are predictive of an addictive phenotype and are present in obese individuals. While these neuroadaptations may contribute to the maintenance of increased calorie intake in an obesogenic dietary environment, the mechanisms by which CDD intake produces this phenotype remain unclear. A possible signal that could mediate diet-induced disruptions in reward circuitry is inflammation, as increased peripheral and central cytokines concentrations are associated with decreased central DA concentrations, decreased D2R levels in the striatum, and decreased FC between the prefrontal cortex (PFC) and nucleus accumbens (NAcc). More recently, neuroinflammation derived from activated microglia has also been implicated in reward pathway dysfunction. However, it remains unclear whether consumption of an obesogenic diet increases inflammation to disrupt corticostriatal reward pathways that facilitates further calorie intake. Using a non-human primate animal model, we hypothesized that diet-induced peripheral inflammation and neuroinflammation will predict increased calorie intake, altered DA concentrations, and decreased corticostriatal functional connectivity.
Methods: Socially housed adult female rhesus monkeys that were maintained in one of two dietary conditions were studied longitudinally. One cohort of females ($n = 18$) was fed a typical low calorie primate laboratory diet (LCD), and a second cohort ($n = 16$) had access to both the LCD and the more palatable CDD (high in saturated fat and sugar). All animals were fed ad lib using a previously validated automated feeding system. Resting state (rs)-fMRI scans were collected 12 months following the diet intervention. ROI analysis used FSL functions adapted for the rhesus brain, and images were corrected for artifacts, smoothed, and signal intensity normalized per experimental block. After

24 months of diet exposure, a subset of subjects received a PET scan using using fluorine [18F]-FEPPA to measure microglia activation. CSF concentrations of DA and its metabolite, homovanillic acid (HVA), and pro-inflammatory signals including C-reactive protein (CRP) and tumor necrosis factor alpha (TNFa) were also assessed.
Results: Female monkeys with access to a CDD in a dietary choice condition consumed more total calories ($p = 0.04$) and gained more weight than females with access to only a LCD ($p = 0.006$). Females with access to the diet choice showed greater peripheral concentrations of CRP ($p = 0.04$), lower CSF levels of DA ($p < 0.001$) and greater HVA:DA ratios ($p = 0.003$). In females with access to a diet choice, CRP concentrations were predictive decreased functional connectivity between the NAcc and the ventromedial PFC. However, increased HVA:DA ratios mediated the effect of CRP on NAcc-vmPFC connectivity ($p = 0.04$), suggesting that diet-induced inflammation decreases presynaptic DA to alter corticostriatial functional connectivity. Additionally, microglia activation within the oribitofrontal cortex (OFC, $p = 0.04$) and the dorsolateral prefrontal cortex (dlPFC, $p = 0.009$) were greater for females in the obesogenic diet condition compared to females with access to only a LCD. Heightened microglia activation in females in the diet choice condition predicted increased calorie intake ($r = 0.93$), greater peripheral ($r = 0.90$) and central (0.76) concentrations of TNFa, and decreased CSF concentrations of HVA ($r = -0.96$).
Conclusions: Taken together, these preliminary data indicate that consumption of an obesogeneic diet impacts reward pathways by decreasing DA concentrations, and attenuating functional connectivity between the PFC and NAcc. Our results suggest that increased peripheral and central inflammation may be critical signals that drive these alterations in reward pathways to facilitate increased caloric intake in an obesogenic dietary environment.
Disclosure: Nothing to Disclose.

32.3 Patterns and Predictors of Cognitive Impairment Among HIV-Infected Women

Leah Rubin

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Background: HIV-associated cognitive impairment (CI) remains a major clinical issue in HIV care and a high priority for HIV research. Although the incidence of dementia has markedly decreased in the era of combination antiretroviral therapy, 30-60% of individuals will exhibit CI at some point during their lifetime. Understanding the patterns, predictors, and mechanisms of CI among HIV-infected (HIV+) individuals is critical for developing adjunctive therapies to improve cognition. We have been focusing on these issues in women, in some cases compared to men, as they are a relatively understudied group and, for a multitude of reasons (e.g. high prevalence of psychological risk factors (PRF)), may be at greater risk for CI compared to HIV+ men.
Methods: We first examined sex differences in neuropsychological (NP) performance using women from the Women's Interagency HIV Study (WIHS; 419 HIV+, 291 HIV-) and a matched group of men from the Multicenter AIDS Cohort Study (MACS). Next, we examined patterns and predictors (PRFs) of HIV-associated CI over a 4-year study duration within WIHS (631 HIV+, 301 HIV-). Moving from epidemiological to mechanistic studies, we examined the role of inflammation and the hypothalamic-pituitary-adrenal (HPA) axis as potential mechanisms underlying CI. The first study examined cross-sectional associations between monocyte-driven inflammatory markers and NP performance in the WIHS. The second study used low dose hydrocortisone (LDH) as an experimental test of whether inflammation and the HPA axis underlie CI. The LDH study was a double-blind, placebo-controlled, cross-over study in HIV+ women ($n = 36$) randomized to either hydrocortisone (10 mg oral) or placebo. We investigated the acute (30 minutes) and delayed effects of LDH (~4 hours) on cognition. Cortisol and immune levels were measured in saliva and anxiety and mood were measured in questionnaires.
Results: A direct comparison of WIHS and MACS participants revealed that HIV+ women performed more poorly on select cognitive domains compared to HIV+ men. In a replication of our previous cross-sectional studies, we found CI in learning, memory, and attention and a decrease in motor skills over time in HIV+ compared to HIV- women. Although HIV+ and HIV- women had similar rates of PRFs, stress in particular was found to negatively influence verbal abilities in HIV+ compared to HIV- women. Mechanistically, in our cross-sectional study, higher levels of monocyte-driven inflammatory markers were associated with lower NP performance in HIV+ women. In our pharmacological challenge study, LDH enhanced learning and memory both 30-45 min after LDH administration and after 4 hours. Decreases in inflammatory markers with LDH were associated with better cognition in women.
Conclusions: HIV+ women are more cognitively vulnerable than HIV+ men in several cognitive domains. Prominent cognitive deficits in HIV+ women are evident in the domains of learning, memory, and attention. Mechanistic work implicates inflammation and alterations in the HPA axis as potential mechanisms underlying the stronger association between stress and cognitive impairment in HIV+ compared to HIV- women.
Disclosure: Nothing to Disclose.

Panel
33. Disentangling the Emotionally Dysregulated Brain: Novel Insights Into Neural Mechanisms of Mood Disorder Vulnerability to Provide Targets for New Therapeutic Interventions

33.1 Computational Models of Effort Based Decision-Making in Mood Disorders

Michael Treadway

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Background: Psychiatric symptoms related to impaired motivation are common across many disorders. Prior lesion studies in animal models have isolated the ventral striatum

and the dorsal anterior cingulate (dACC) as key regions that govern decisions regarding how much effort to invest for rewards. To date however, the role of dACC in both normal and abnormal cost-benefit decision-making remains unclear.
Methods: Data are presented from three different studies. First, we present new fMRI data in healthy volunteers ($n = 28$) using a novel effort-based decision-making task where reward and effort information are presented sequentially. Second, we present a new computational modeling approach to behavioral effort-based decision-making collected with the EEfRT, and reveal its characteristics in several previously collected samples of healthy volunteers and MDD patients (total $n = 96$). This model was validated on a separate sample of 450 participants (not included) using AIC comparisons against several null models. Finally, in a third study we present fMRI data MDD patients and matched controls ($n = 21$ per group). All fMRI results are analyzed in spm12 and results are corrected for multiple comparisons.
Results: Results: For study 1, consistent with other studies of value-based decision-making, we find that activity in ventromedial prefrontal cortex (vmPFC) tracks the selected value of the chosen option. However, using a novel subjective-value prediction model, we reveal that dACC signal reflects an update of expected subjective value in response to the sequential unfolding of cost and benefit information that is used to guide decisions. For study 2, modeling results indicate that control participants are better fit than MDD patients indicating that MDD patients may be more likely to deviate from subjective value comparisons. In addition, MDD patients were fit by higher effort parameters, indicating increased sensitivity to effort ($p = 0.02$). In study 3, we find that a computational model again shows that MDD patients exhibit greater effort sensitivity ($p = 0.04$), and further reveal that controls exhibit greater activity in the same dACC region identified by study 1 when choosing to expend greater effort (pFWE = 0.008, SVC for dACC ROI).
Conclusions: Together, these results help clarify the role of dACC in human effort-based decision-making, demonstrate that MDD is associated with greater sensitivity to effort, and suggest that alterations in dACC activity in particular may be associated with this deficit.
Disclosure: Part 1: Avanir Pharmaceuticals, Consultant, NeuroCog Trials, Consultant, Boston Consulting Group, Consultant, BlackThorn Therapeutics, Consultant, BlackThorn Therapeutics, Royalties, **Part 2:** BlackThorn Therapeutics, Royalties.

33.2 Impaired Prediction Error Encoding During Reward Learning in Depression: GABA and Dopaminergic Modulations

Poornima Kumar

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Background: Anhedonia (hyposensitivity to rewards) and negative bias (hypersensitivity to punishments) are core features of major depressive disorder (MDD). These deficits could be due an inability to learn about rewards and punishments, respectively, and modify behavior based on experience with the environment. Critically, animal studies have shown that dopamine (DA) signaling in the ventral tegmental area (VTA) and gamma-Aminobutyric acid (GABA) transmission from the habenula, as well as interactions between these systems, are implicated in reward and punishment learning. This interaction in turn modulates DA release to and GABA concentrations in the basal ganglia. Despite compelling evidence of involvement of DA and GABA systems during reinforcement learning in animals, very few studies have investigated these effects in humans.
Methods: This talk will present findings from three studies investigating reinforcement learning and effects of dopamine and GABA on learning in individuals with MDD. Study 1: We acquired fMRI data from 25 unmedicated participants with MDD and 26 healthy controls during a monetary reinforcement learning task. Study 2: Investigated social reinforcement learning in 30 unmedicated participants with MDD and 29 healthy controls after a single dose of placebo or 50 mg of the D2/D3 antagonist amisulpride (hypothesized to increase DA levels via autoreceptor blockade). Study 3: In an ongoing study, baseline GABA levels in the left and right striatum were measured using a novel multi-voxel Magnetic Resonance Spectroscopy Imaging technique from 12 unmedicated participants with MDD and 13 healthy controls. Participants then completed a social reinforcement learning task. In all these studies, reward prediction error (RPE) was calculated using temporal-difference algorithms and parametrically modulated with hemodynamic responses to identify brain regions that encode RPE signals.
Results: Study 1: Although the probability of choosing the correct stimulus generally increased throughout the task, relative to controls, the MDD group was characterized by reduced learning from gains (i.e., lower choices of the stimulus indicating a high probability of monetary gain; $p < 0.0001$), but no impairment in avoiding the stimulus associated with a high probability of monetary loss. RPE signals in the VTA did not differ between groups. However, we observed blunted RPE signal in the ventral striatum (VS; $p = 0.006$) and reduced VTA-VS connectivity during rewards in MDD vs controls ($p < 0.05$). In addition, for the control – but not MDD – group connectivity between VTA-VS correlated positively with RPE signals in the VS ($r = 0.57$, $p < 0.05$). Study 2: There was no behavioral performance difference between the groups. However, preliminary observations revealed that the drug modulated RPE signals in the VS across groups. Study 3: Preliminary data on 9 subjects revealed that the VS RPE signal correlated negatively with baseline GABA levels in the striatum ($p < 0.05$).
Conclusions: Collectively, these findings highlight important reward-related learning deficits in MDD and shade initial light on their underlying molecular mechanisms.
Disclosure: Nothing to Disclose.

33.3 Abnormalities in Reward-Related Neural Circuitry in Unipolar Depression Versus Bipolar Disorder

Robin Nusslock

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Background: There is growing evidence that risk for bipolar disorder and unipolar depression (without a history of hypomania/mania) are characterized by distinct and

opposite profiles of reward processing and reward-related neural activation. Specifically, bipolar disorder, and particularly risk for hypomania/mania, is characterized by a hypersensitivity to reward-relevant stimuli. By contrast, major depressive disorder (MDD) is characterized by a blunted sensitivity to reward-relevant stimuli and abnormally reduced reward-related neural activation.
Methods: Extending this literature, this talk presents results from two studies examining functional and structural connectivity within the cortico-striatal reward circuit among individuals with MDD and individuals at risk for bipolar disorder.
Results: Using analyses of functional connectivity (i.e., psychophysiological interaction analysis) among 25 participants with current MDD and 25 healthy controls, we report that MDD, but not healthy control participants, display a negative coupling between activation in the medial orbitofrontal cortex (OFC) and the nucleus accumbens (NAcc) during reward processing, $t(48) = 2.60$, $p = < .05$, FDR corrected. This suggests that unipolar depression may be characterized by an inability of the prefrontal cortex to dynamically engage sub-cortical reward processing regions when encountering rewarding stimuli. By contrast, using analyses of structural connectivity (i.e., diffusion tensor imaging), we report that individuals at elevated risk for bipolar disorder ($N = 54$) display enhanced white matter integrity between the NAcc and the medial OFC, $F(1, 52) = 5.32$, $r = 0.36$, $p = 0.03$. This suggests that individuals at risk for bipolar disorder may abnormally amplify subcortical reward processing via the prefrontal cortex.
Conclusions: These results have important implications for establishing biomarkers of differential risk for unipolar depression versus bipolar disorder. Results also have implications for unpacking the mechanisms of top-down (OFC-to-NAcc) versus bottom-up (NAcc-to-OFC) reward processing abnormalities in mood disorders.
Disclosure: Nothing to Disclose.

33.4 Toward Neural Biomarkers of Risk for Bipolar Spectrum Disorders and Neural Targets for Novel Neuromodulation Interventions

Mary Phillips

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Background: Onset of bipolar spectrum disorders (BPSD) peaks in early adulthood, yet it is extremely difficult to identify those young adults who are at specific risk for BPSD, as there are no objective biomarkers denoting risk for these disorders. High trait impulsive sensation seeking (ISS) is a risk factor for future BPSD in young adults. We reported in young adults a positive association between trait ISS and activity in distributed reward circuitry, including left ventrolateral prefrontal cortex (vlPFC) and ventral striatum (VS), to cues denoting uncertain future reward, i.e., to uncertain reward expectancy (RE). This parallels our previous finding of greater activity in left vlPFC and VS to uncertain RE in adults with BPSD vs. healthy adults. Elevated left vlPFC and VS activity to uncertain RE is thus a promising neural biomarker of BPSD. As the next stage in the search for neural biomarkers of BPSD, we are examining: 1. whether trait ISS is positively associated with functional connectivity (FC) in reward circuitry at rest in young adults; 2. The extent to which activity and FC in this circuitry to uncertain RE predicts future clinical outcome in young adults; 3. How such neural biomarkers can be used as targets for novel neuromodulation interventions in adults with BPSD.
Methods: Study 1: Resting state FC. In $n = 122$ (non-BPSD) 18-25 year-olds, we used Group Iterative Multiple Model Estimation, allowing computation of effective resting state FC in large scale neural networks, based on structural equation modeling and model selection. Study 2: Future outcome prediction. In $n = 27$ of the above 18-25 year-olds, we examined whether left vlPFC FC during uncertain RE predicted future worsening of hypo/mania vs. other symptoms at 6 months after scan. Study 3: Neuromodulation. In an ongoing study, $n = 9$ adults with BPSD and $n = 10$ healthy adults received left vlPFC cathodal (inhibitory) and control (left cathodal somatosensory) transcranial direct current stimulation (tDCS) concurrently with fMRI during reward task performance on two separate occasions, one week apart (counterbalanaced order across participants).
Results: Study 1: We show three subgroups of resting state FC patterns ($n = 52$, $n = 22$, $n = 49$), in which one pattern comprised significant FC from left VS to left vlPFC ($n = 22$). This subgroup had significantly higher Behavioral Activation System reward responsiveness scores vs. other subgroups ($F[2,114] = 4.2$, $p = 0.018$), accounting for age, gender, years of education, motion and group (seeking help for psychological distress or not), and indicate that greater resting state FC in left vlPFC-VS reward circuitry at rest is associated with a component of trait ISS, reward responsiveness. Study 2: There was a positive correlation between left vlPFC-left superior parietal cortical FC during uncertain RE and change specifically in mania, but not other symptom, severity from scan to 6 months later ($p < 0.001$, uncorrected; $R2 = 0.22$). Study 3: Adults with BPSD, but not healthy adults, showed significant reduction in bilateral VS activity during uncertain RE with left vlPFC cathodal vs. control tDCS ($t(8) = -3.43$, $p = 0.009$).
Conclusions: Together, our findings highlight the role of heightened left vlPFC-centered reward circuitry activity and FC during reward processing and rest as promising neural biomarkers of BPSD and BPSD risk. Our data also point to the left vlPFC as neural target for novel neuromodulation interventions for individuals with BPSD and those at risk for these debilitating disorders.
Disclosure: Nothing to Disclose.

Study Group

34. Isolated Populations and Psychiatric Research: The Importance of Well Assembled Global Collaborations to Mainstream Scientific Psychiatry

Javier Escobar*, Victoria Arango, Carlos Lopez-Jaramillo, Carrie Bearden, Gabriel de Erausquin

Rutgers-Robert Wood Johnson Medical School, New Brunswick, New Jersey, United States

Study Group Summary: Geographically stable, well-defined populations offer unique opportunities for neuroscience

research. Studies in the US (Amish) and outside the US (Island, Finland, Sardinia) have been of relevance but limited by small Ns of informative families. In Latin America, large, extended family systems offer unique opportunities (Venezuelan pedigree and Huntington's disease gene).

This study group will discuss two ongoing collaborations on special, isolated populations, residing in two Andean regions of Colombia and Argentina.

The "paisa" population includes large informative families residing near Medellin, Colombia for several generations. Francisco Lopera, a neurologist who leads the neurosciences group at Universidad de Antioquia first observed intriguing cases of early Alzheimer's disease, that were tracked to a presenilin-1 gene mutation in 25 extended family systems with > 5,000 affected individuals. This population is now the target of an intervention trial by an international consortium and Dr. Lopera will provide an update on this research.

Another collaboration targets bipolar disorder in the same population, following observations of a high prevalence of suicide and mood disorders in this region. Investigators at Universidad de Antioquia, UCLA, UCSF and Rutgers have been collaborating there for over a decade in NIH-funded projects. Dr. Lopez Jaramillo will give a detailed description of the "paisa" population and highlight its advantages for genetic studies of bipolar disorder and other conditions. Carrie Bearden, Ph.D., a Professor at UCLA and ACNP fellow, will discuss a phenotyping approach to these large multigenerational pedigrees genetically enriched for severe bipolar disorder that highlights heritable traits in several domains (neuroanatomy, temperament, cognition, circadian rhythms), as well as novel unpublished data related to brain structure and sleep/activity patterns.

Studies in Argentina led by Gabriel de Erausquin target a geographically isolated population of Quechua origin residing in the Andes Mountains of northern Argentina, with a high incidence of untreated psychoses. A collaboration of Universities in Argentina and Peru with Washington University, Harvard University and Rutgers University has been going for over a decade and has yielded important information on the natural history and neurological correlates of untreated psychoses that may share developmental and genetic pathways. Dr. de Erausquin and his mentor Dr. Cloninger, also a member of ACNP, will participate in the study group discussion and highlight major findings and advantages of this collaboration.

The co-chair, Victoria Arango, PhD, a fellow of ACNP, will highlight the relevance of these special populations to the study of suicide.

Several other issues to be raised in the discussion include:

Can these collaborations help elucidate the contribution of genomics to health disparities? (see McGlone West et al, JAMA 2017).

What makes these collaborations successful? – What are the essential elements for solid team building and the success in getting extramural funds?

What is the relevance of mentoring and research training grants?

How can we build sustainable research capacity for collaborative psychiatric genetics work in South and Central America?

Why is important that organizations like the ACNP reach out to and maintain links to investigators in other South and Central American Countries?

Disclosure: Nothing to Disclose.

Panel
35. Innovative Pharmacotherapies and Strategies to Address the Opioid Epidemic

35.1 Comparative Effectiveness of Extended-Release Naltrexone vs. Buprenorphine for Opioid Dependence Treatment – NIDA CTN-0051

John Rotrosen

New York University School of Medicine, New York, New York, United States

Background: With both agonist and antagonist medications available to treat opioid dependence, and with these differing markedly on a spectrum of parameters – including philosophy of treatment, need for detoxification to initiate treatment, ongoing dependence and withdrawal on stopping treatment, diversion risk, community acceptability and controlled substance restrictions – it's difficult to know which approach to take. What do we tell our patients? How should we choose? Is one approach better for some patients, the other for others? Does choice matter, do patients do better with their preferred treatment than with the alternative? CTN-0051 grows out of these questions and will establish the evidence-base to inform treatment decisions.

Methods: Close to a dozen designs, including no-medication treatment-as-usual conditions, SMART designs, and designs taking choice into consideration were considered. The final design was a randomized two-arm, head-to-head effectiveness trial comparing extended-release naltrexone to buprenorphine, both FDA approved treatments, in 570 patients across 8 sites. Recruitment was from inpatient detoxification and short term residential programs. We selected a flexible point of randomization to reflect community medication initiation practices. We predicted differential induction success, and included a mitigation plan to manage the detoxification hurdle. Treatment was for up to six months with follow up visits through nine months post-randomization. The primary outcome measure was time-to-relapse, defined as 7 consecutive use-days or 4 consecutive use-weeks. Secondary outcomes included successful induction, abstinence from opioids, alcohol, tobacco and other drugs, craving, subacute withdrawal, etc., and mediators and moderators of treatment response.

Results: Recruitment was completed in May 2016 with 722 participants consented and 570 randomized. Treatment visits were completed in November 2016 and follow up visits in February 2017. Women made up 30% of the population; 17% were Hispanic, 26% non-white; 69% were 25-45 years old, 15% were under 25; 57% had a high school education or less, 66% were never married, 63% were unemployed; 82% identified heroin as their primary abused opioid, 16% prescription drugs; 63% were IV users; 40% were high users defined as IV use of at least 6 bags a day, a stratification variable that we predicted to influence outcome; although all participants expressed willingness to take either medication,

29% indicated that they preferred extended-release naltrexone, and 33% buprenorphine. AEs and SAEs were those expected in this population. Data lock will be in May 2017 after which we will be able to look at outcomes data.
Conclusions: We expect to be able to present comparative effectiveness data on induction success, survival without relapse, time-to-relapse, abstinence from opioids and other drugs, successful completion of 24 weeks of treatment, craving, cognitive function, and adverse events including overdose during and after treatment. Findings on the role of choice in influencing outcome, and mediators and moderators of treatment success or failure will be presented. None of these data will be available until after data-lock, precluding more specific "conclusions" per se until early summer.
Disclosure: Nothing to Disclose.

35.2 Effectiveness of Extended Release Naltrexone Versus Daily Buprenorphine-Naloxone for Opioid Dependence - Norway Trial

Lars Tanum

Akershus University Hospital, Loerenskog, Norway

Background: Extended-release naltrexone (XR-NTX) has proven effective in reducing the use of heroin and other illicit opioid drugs in opioid dependent individuals. The medication has not previously been compared directly to any opioid agonist maintenance treatment (OMT) such as methadone or buprenorphine, the currently recommended treatment for opioid addiction. The objective of this study was to compare the effectiveness of intramuscular Extended-release natrexone (380 mg) with daily buprenorphine-naloxone (BP-NLX) in reducing heroin and other illicit drug use among adult opioid users.
Methods: This was a 12-week multi-center open-label pragmatic randomized controlled trial taking place at five urban addiction clinics in Norway. A total of 232 adult opioid dependent (DSM-IV) volunteers, 18-60 years of age, were recruited from outpatient addiction clinics and detoxification units and assessed for eligibility. Of these, 159 participants were randomized to study medication and eligible for ITT analyses. Participants were randomized to either daily oral flexible dose buprenorphine-naloxone (BP-NLX) (4-24 mg /day) or 4-weekly intramuscular extended-release naltrexone (XR-NTX) 380 mg. Main outcomes were days of non-use of illicit opioids (range 0-84), proportion of opioid-negative urine drug tests (range: 0-1), days of non-use of other illicit substances (range 0-84), number of participants completing the RCT, change in heroin craving and satisfaction with treatment.
Results: The treatment groups showed a similar retention in the study, 68.4 days versus 62.9 days (XR-NTX and BP-NLX resp.) and a similar decrease in the use of heroin and other illicit opioids from baseline. The treatment groups did equally well in reducing the use of heroin or other illicit opioids, heroin thoughts, heroin craving and on the proportion of opioid negative UDTs. Participants on XR-NTX reported higher satisfaction with treatment, but reported also more adverse effects than BP-NLX participants. Four participants in the XR-NTX and six in the BP-NLX group withdraw from the study due to non-serious adverse effects. Serious adverse events (SAE) were reported in six patients on XR-NTX and three patients on BP-LNX.
Conclusions: Maintaining opioid abstinence with XR-NTX is a feasible and safe alternative to daily opioid substitution with BP-NLX in abstinence motivated opioid dependent adults.
Disclosure: Nothing to Disclose.

35.3 Initial Efficacy of Interim Buprenorphine Dosing for Reducing Illicit Drug Use and Associated Risks Among Waitlisted Opioid-Dependent Adults

Stacey Sigmon

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Background: Despite the demonstrated efficacy of agonist maintenance for opioid use disorder, individuals can remain on treatment waitlists for months during which they continue to use illicit opioids and are at significant risk of infectious disease and premature death. Interim buprenorphine dosing, involving medication without counseling, may mitigate these risks during treatment delays.
Methods: In a 12-week randomized pilot study, we evaluated the efficacy of a novel Interim Buprenorphine Treatment for waitlisted opioid-dependent adults. Participants ($n = 50$) were randomized to receive IBT ($n = 25$) or continued Waitlist Control (WLC; $n = 25$). IBT consisted of bi-monthly visits for observed buprenorphine ingestion with the remaining doses dispensed via computerized device, daily monitoring via an Interactive Voice Response (IVR) phone system, and IVR-generated random call-backs. The primary outcome was percent of participants who provided illicit opioid-negative urine specimens at 4-, 8- and 12-week assessments, with secondary outcomes of injection drug use, non-opioid drug use, adherence and patient satisfaction.
Results: Compared to WLC, IBT participants submitted a greater percentage of illicit opioid-negative urine specimens at 4- (88% vs. 0%), 8- (84% vs. 0%), and 12-week (68% vs. 0%) assessments (p's $< .001$). They also demonstrated greater reductions in frequency of injection drug use ($p < .001$) and Addiction Severity Index Drug ($p < .001$) and Psychiatric ($p = .02$) composite scores. Finally, adherence with buprenorphine administration (99%), daily monitoring (96%) and random call-backs (96%) were high, as were ratings of treatment satisfaction (4.6+0.7 on a 5-point scale).
There was also a group by time interaction on the BDI-II ($F(3,125) = 11.26$, $p < .01$) whereby scores decreased among IBT participants ($F(3,125) = 26.62$, $p < .01$) and were lower at 4-, 8- and 12-week timepoints vs. intake (p's $< .01$; Figure 1). There was a group by time interaction on the BAI ($F(3,126) = 3.33$, $p < .05$), with scores decreasing in IBT participants ($F(3,125) = 12.68$, $p < .01$) and lower than intake at Weeks 4, 8 and 12 (p's $< .01$). The ASI Psychiatric subscale showed a similar pattern ($F(3,126) = 3.56$, $p < .05$), with scores decreasing in IBT participants ($F(3,125) = 9.25$, $p < .01$) and lower than intake at all timepoints (p's $< .01$). In contrast, there were no changes in BDI-II, BAI or ASI Psychiatric scores among WLC participants.
Conclusions: Despite the effectiveness of methadone and buprenorphine maintenance for opioid use disorder, many

opioid-dependent individuals encounter prolonged delays when seeking treatment. Individuals randomized to IBT demonstrated significantly greater reductions in illicit opioid use and injection use relative to waitlist controls. Provision of IBT without counseling was also associated with significant reductions in psychiatric distress among waitlisted opioid-dependent individuals. Taken together, providing IBT to waitlisted opioid-dependent adults may reduce individual and societal risks during delays to comprehensive treatment.
Disclosure: Nothing to Disclose.

35.4 Novel Formulations of Buprenorphine for the Treatment of Opioid Use Disorder

Sharon Walsh

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Background: Buprenorphine is an efficacious treatment for opioid use disorder; it blocks the effects of illicit opioids, suppress withdrawal and craving and reduces illicit opioid use. Despite these benefits, the daily sublingual products have been associated with significant diversion and pediatric overdoses. Long acting formulations should obviate these risks.
Methods: Phase II and Phase III clinical trials will be reviewed for three novel buprenorphine products, including a newly approved 6-month implant, a monthly fixed-dose, sustained-release injectable, and a weekly and monthly titratable sustained-release injectable.
Results: Supportive findings from each of the pivotal trials for these three novel formulations will be described in detail. Outcomes vary by study but will include opioid withdrawal suppression, opioid blockade, illicit opioid use, retention in treatment and other key secondary outcomes.
Conclusions: The newly approved implant product is available for use; the target patient population and the necessary resources for its implementation will be described. The potential utility of the sustained-release injectable formulations will be reviewed.
Disclosure: Part 1: Braeburn Pharmaceuticals, Consultant, Lightlake Therapeutics, Advisory Board, World Meds, Advisory Board, INSYS, Consultant, Daiichi Sankyo, Consultant, Neurocrine, Consultant, Astra Zeneca, Consultant, Cerecor, Advisory Board, Sun Pharma, Consultant, Kem Pharm, Consultant, Camurus, Consultant, **Part 2:** Braeburn Pharmaceuticals, Consultant, **Part 3:** Braeburn Pharmaceuticals, Consultant, **Part 4:** Cerecor, Grant, Braeburn Pharmaceuticals, Grant.

Mini Panel
36. Transcranial Electrical Stimulation for Improving Cognition in Schizophrenia: Preclinical and Clinical Insights

36.1 Convergent Neuroscience of Transcranial Alternating Current Stimulation

Flavio Frohlich

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Background: Transcranial alternating current stimulation (tACS) applies a weak sine-wave electric current to the scalp for non-invasive targeting of cortical brain rhythms. One promising target for tACS are alpha oscillations (8-12 Hz), a thalamo-cortical rhythm that represents a network substrate for long-range functional interactions in the cortex. Yet, it is unknown if tACS can modulate and restore alpha oscillations that are impaired in schizophrenia and other network dysconnectivity disorders. Here, we report on our convergent neuroscience approach that combines computational modeling of neuronal networks, animal electrophysiology, and a treatment clinical trial of tACS in schizophrenia. We present mechanistic insights on (1) target engagement mechanisms of tACS and (2) target validation of tACS in a randomized, placebo-controlled clinical trial for the treatment of auditory hallucinations in patients with schizophrenia.
Methods: Computer simulations: We simulated cortical networks using quadratic integrate-and-fire neurons to investigate how cortical networks respond to tACS and how deficits in lang-range input in the alpha frequency range can be restored through tACS.
In vivo electrophysiology: We implanted microelectrode arrays in both thalamus and multiple cortical areas of the ferret to examine how alpha oscillations emerge through circuit interactions and are modulated by tACS.
Clinical trial: We evaluated the efficacy of tACS in enhancing synchronization between the frontal and temporo-parietal areas of the left hemisphere. Behavioral measures (primary outcome: auditory hallucination rating scale, AHRS) were used to determine the efficacy of tACS and tDCS for the treatment of medication-refractory auditory hallucinations in patients with schizophrenia or schizoaffective disorder. The study had a parallel group design with three arms (sham, tACS, and tDCS). Participants received twice daily, 20-minute sessions of active sham, 10 Hz 2 mA tACS, or 2 mA tDCS over the course of 5 consecutive days. Resting state EEG and auditory steady-state responses (ASSR) were collected at baseline, completion of stimulation, and two follow-up visits (one week and one month after completion of tACS). Changes in alpha power, functional connectivity in the alpha frequency band, and ASSR were analyzed.
Results: The computer simulations showed that tACS can entrain and normalize alpha oscillations in a network that exhibits different, incompletely synchronized areas with different oscillation frequencies. Stimulation at the endogenous frequency was most effective in restoring and enhancing alpha oscillations. Successful target engagement was reflected in a shift of the oscillation frequency to the stimulation frequency, providing a marker for target engagement in the EEG data of our clinical trial.
Electrophysiological recordings in the awake ferret showed that the alpha oscillation is a state-dependent rhythm that strongly entrains neuronal firing. We examined an array of stimulation parameters (frequency and amplitude) to determine the target engagement principle. We found weak entrainment of cortical units (N = 2 ferrets).
In our clinical trial of tACS in schizophrenia (N = 25), we found that tACS had a larger raw effect size than tDCS or sham stimulation in terms of change to the AHRS both after 5 days of stimulation (1.31, 0.17, 1.06) and at the 1 week follow-up (0.78, 0.39, 0.71). Functional connectivity showed a clear shift towards the stimulation frequency, as predicted by

our preclinical work. Importantly, these changes to the organization of the network dynamics in the alpha frequency only occurred in the tACS group and not in the tDCS or sham group. Finally, 40 Hz ASSR was enhanced relative to baseline, correlated with clinical improvement measured by the AHRS, and only occurred in the tACS group.
Conclusions: We here employed a convergent neuroscience approach to demonstrate the underlying mechanism of target engagement by tACS. We found that cellular and network properties at the micro- and mesoscale interact to give rise to frequency-tuned target engagement by tACS in computer simulations and the ferret model. Importantly, we were able to demonstrate the predicted enhancement of alpha oscillations by tACS translated to patients with schizophrenia. Together, our work supports the further evaluation of tACS in clinical populations with impaired functional long-range connectivity.
Disclosure: Part 1: Pulvinar Neuro LLC, Stock / Equity, Pulvinar Neuro LLC, Consultant,
Part 4: Tal Medical, Grant.

36.2 Effects of Transcranial Stimulation on Cognitive Function and Brain Functional Changes in Schizophrenia

Robert Smith

New York University Medical School, Hewlett, New York, United States

Background: Cognitive deficits which persist in chronic schizophrenia (CSZ) after treatment of acute symptoms are implicated in persistent functional deficits. They have no generally effective treatments. Transcranial Direct Current Stimulation (tDCS) to the DLPFC has been shown to improve some aspects of working memory in normals, and our earlier study in a US sample showed that 5 sessions of active tDCS (AtDCS)compared to sham(SH) significantly improved cognitive function in CSZ measured using the MATRICS battery (MCCB). The generalizability of these findings and their relation to changes in brain function need to be investigated. We report a double-blind study of 10 sessions of tDCS on cognition and functional brain imaging changes in a larger sample of Chinese CSZ.
Methods: 40 Chinese patients with a diagnosis of schizophrenia or schizoaffective disorder were randomized to receive 10 sessions of AtDCS (2 mA for20 minutes) or SH tDCS (2ma for 40 seconds) over approximately a two-week period (anode L DLPFC, cathode R supraorbital ridge). Cognitive function was evaluated with a Chinese version of the MCCB, the Paced Auditory Serial Addition Task (PASAT), and subtests from the CogState battery, at baseline and at various times during and after tDCS treatment. Psychiatric symptoms were evaluated with the PANSS scale. Changes in brain function were evaluated with an fMRI protocol at baseline and after 10 tDCS sessions for resting state changes in brain connectivity, and changes in activation in DLPFC during a 0 and 2 back working memory task (WM).
Results: Preliminary analysis of data from the first 20 subjects, showed AtDCS vs SH significantly increased scores in MCCB domains of Speed of Processing ($P = .013$) and tended to improve Reasoning and Problem Solving, but did not improve MCCB overall composite score, or attention-vigilance and spatial working memory domain scores. There was a non-significant trend for AtDCS to improve accuracy on the PASAT (verbal WM). TDCS did not significantly change any PANSS scores. During the active WM task performed during fMRI the AtDCS group vs SH showed significant improvement in accuracy of 0 back condition after 10 sessions of tDCS ($p < 0.01$) but not 2-back task. In conjunction with this improvement in performance, task activation decreased in right middle frontal gyrus after 10 sessions of AtDCS for the 0 back condition, and there was a trend for increased activation in 2-back, A comparison of 2 back vs. 0 back task activation showed significant increased activation in bilateral middle frontal gyrus was evident after 10 sessions of AtDCS vs. SH.
Conclusions: In the preliminary analysis of the partial Chinese sample, the effects of AtDCS on cognition did not have the same strong effects on MCCB composite score, attention and working memory that we found in the US sample. However, AtDCS showed positive effects in other cognitive domains. The behavioral and activation patterns in the middle frontal gyrus during the 0 back task suggest AtDCS produced better performance with lower effort on the low-load 0 back task, closer to what we would expect in normals. The increased activation in the 2-back task suggests potential for increased cognitive reserve stimulated by tDCS.
Disclosure: Nothing to Disclose.

36.3 Combining TDCS With Cognitive Training to Promote Generalization of Learning in Patients With Schizophrenia

Tasha Nienow

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Background: Cognitive impairment associated with the onset of schizophrenia is severe, nearly ubiquitous, and associated with functional outcomes. The development of interventions to address this aspect of the illness has been ambitiously pursued. Cognitive training approaches that aim to restore abilities have shown some promise. However, gains have been modest, and the inconsistency with which learning generalizes to untrained cognitive tasks significantly limits the viability of cognitive training as a clinical intervention. Transcranial direct current stimulation (tDCS) is a neuromodulation technique that modulates neuro-excitablity and can be used in conjunction with cognitive training. To explore the efficacy of this combination, the neural system supporting working memory processes was targeted with repeated, concurrent administration of these interventions. Data from two studies will be presented to demonstrate evidence of target engagement and to quantify the magnitude of training gains and generalization when working memory training is combined with tDCS versus sham.
Methods: In study 1, 80 patients with schizophrenia were randomized to receive 48 hours of working memory training or a time-matched, active placebo condition. Working memory training consisted of adaptive training tasks selected

from commercial software as well as N-back tasks developed for this protocol. Outcomes were assessed with a training task (word N-back), a novel version of this training task (picture N-back), and untrained tasks of working memory and related cognitive abilities from the MATRICS Consensus Cognitive Battery (MCCB). A portion of the sample ($n = 26$) underwent fMRI pre- and post-intervention to examine change in brain connectivity attributable to working memory training. In study 2, 10 patients with schizophrenia completed the working memory training protocol from study 1 with concurrent tDCS ($n = 6$) or sham ($n = 4$) stimulation. Beginning in the third week of training, twice a week, 1 mA of anodal tDCS or sham stimulation was applied to the dorsal lateral prefrontal cortex (F3) concurrent with the first 20 min of training. Cognitive and symptom outcomes were assessed as well as tolerability of this combination of interventions.

Results: Training gains in study 1 demonstrated that change on the N-back training task (learning) was positively associated with generalization to an untrained task, r = .24, p = .05 (unpublished data). In study 2, patients who received working memory training with tDCS made significantly greater gains than patients in the sham condition on word, d = .48, and picture, d = .62, N-back training tasks and demonstrated greater generalization to untrained tasks. In a subset of the study 1 sample, performance gains on the N-back task were associated with increases in thalamo-corticial connectivity following working memory training.

Conclusions: Learning on training tasks is predictive of generalization to untrained tasks. Results indicate that tDCS is a safe and efficacious means of promoting learning and generalization of working memory training and suggests that cognitive training methods can be enhanced with tDCS. Intervention-induced change in working memory performance was associated with change in thalamo-cortical connectivity, suggesting that this network might be a fruitful target of intervention with behavioral and neuromodulatory techniques.

Disclosure: Nothing to Disclose.

Panel
37. In Control: Prefrontal Cortex Regulation of Motivation, Impulsivity and Reward

37.1 Projection Specific Encoding of Reward in the Prefrontal Cortext and Limbic Thalamus

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Background: The medial prefrontal cortex (PFC) is a critical hub for orchestrating motivated behaviors across mammalian species. In addition to intra-cortical connectivity, prefrontal projection neurons innervate subcortical structures that contribute to reward-seeking, such as the nucleus accumbens (NAc), ventral tegmental area, and midline thalamic areas such as the periventricular thalamus (PVT). While connectivity among these structures contributes to appetitive behaviors, how projection-specific prefrontal neurons encode reward-relevant information to guide aspects of behavior is largely unknown.

Methods: We used in vivo two-photon calcium imaging in awake and behaving mice to monitor the activity of dorsomedial prefrontal neurons and midline thalamic neurons in mice during a Pavlovian conditioning task. Over multiple conditioning sessions, mice were trained to associated the delivery of sucrose (appetitive) or quinine (aversive) with a neutral auditory tone. Cue-reward learning was indicated by the expression of anticipatory licking to the cue that predicts sucrose while mice generally withheld licking on trials when quinine was delivered. We quantified time-locked changes in calcium activity in multiple classes of PFC output neurons as well as PVT neurons that project to the NAc. To complement these experiments, we preformed pathway specific optogenetic manipulations of distinct projection neuron subtypes to determine whether time-locked activation or inhibition of these circuits could affect reward seeking behavior and decision-making.

Results: At the population level, PFC neurons display diverse activity patterns during the presentation of reward-predictive cues. However, recordings from PFC neurons with resolved projection targets reveal that individual PFC-NAc neurons show response tuning to reward-predictive cues, such that excitatory cue responses are amplified across learning. By contrast, PFC-PVT neurons gradually develop new, primarily inhibitory responses to reward-predictive cues across learning. Furthermore, bidirectional optogenetic manipulation of these neurons reveals that stimulation of PFC-NAc neurons promotes conditioned reward-seeking behavior after learning, while activity in PFC-PVT neurons suppresses both the acquisition and expression of conditioned reward seeking. With respect to PVT neurons, data reveal that while some of these cells have excitatory responses to the cue associated with sucrose after learning, other PVT neurons have inhibitory response profiles. In contrast, preliminary data suggest that PVT neurons do not develop excitatory or inhibitory responses to the stimulus that predicts the quinine.

Conclusions: These data show how PFC circuitry as well as neurons in the PVT can dynamically control reward-seeking behavior and decision making. Ongoing experiments that are still in progress, are investigating whether PFC-VTA neurons show similar or distinct encoding properties as those seen in PFC-NAc and PFC-PVT neurons. Collectively, the approach of systemically profiling and characterizing the activity dynamics of interconnected cell groups based on their project targets and gene expression profiles may provide important insight into the neurocircuit imbalances that occur in a wide variety of neuropsychiatric illnesses.

Disclosure: Nothing to Disclose.

37.2 Role of Subtypes of Cortical Cells in Behaviors Associated With Cocaine Addiction

Susan Ferguson

University of Washington/Seattle Children's Research Institute, Seattle, Washington, United States

Background: Drug addiction is a chronic relapsing disease that has costly social and economic impacts on our society,

in addition to the profound medical consequences faced by addicts. The cortico-basal ganglia-thalamic network is involved in decision-making, motivation and reward, and alterations within this circuit regulate the development of drug addiction. The prefrontal cortex serves as a key modulator of this circuit, providing strong glutamatergic drive to the striatum, as well as widespread input throughout the cortico-basal ganglia-thalamic system. Of note, cortical processing is crucial for the patterning of appropriate behavior and loss of top-down cortical control during drug use is thought to play a major role in the transition to addiction, as well as relapse. However, cortical pyramidal neurons can be subdivided into two major types with distinct inputs and projections targets, molecular and receptor profiles, morphologies and electrophysiological characteristics. Cortical neurons that have sparse apical tufts, minimal h-currents, and are regular spiking project bilaterally to striatum and contralateral cortex (IT) whereas cortical neurons that have thick apical tufts, prominent h-currents, and are burst firing send their main axon into the pyramidal tract with collateral projections to ipsilateral striatum and other subcortical structures (PT). As a result of the distinct connectivity patterns and cellular properties of these two neuronal populations, it has been hypothesized that they integrate and convey distinct signals for guiding decision-making processes and motivated behaviors. Nonetheless, the role of these two cell populations in the regulation of addiction behaviors has not been examined.

Methods: To begin to address this, we used Cre-recombinase dependent, combinatorial viral vector targeting approaches to express inhibitory Gi/o-coupled DREADDs (Designer Receptors Exclusively Activated by Designer Drugs) selectively in PT or IT cortical neurons. Activation of DREADDs by the otherwise inert ligand clozapine-n-oxide produces transient decreases in neuronal activity and allowed us to examine the effect of these targeted cellular manipulations on behaviors associated with addiction (drug self-administration, psychomotor sensitization, conditioned place preference).

Results: We found that decreasing activity of PT neurons significantly enhanced locomotor sensitization to cocaine. In addition, this manipulation also enhanced the rewarding properties of cocaine, as animals significantly preferred a chamber associated with PT inhibition plus cocaine compared to cocaine alone. This effect of PT inhibition was specific to cocaine, as it had no effect on a hedonic food reward-induced CPP, nor did it produce a place preference by itself. Interestingly, although PT inhibition had no effect on cocaine self-administration behavior in rats that showed escalation of drug intake, it did enhance drug-taking and drug-seeking in rats that showed stable cocaine intake. Although experiments examining the effect of IT inhibition are currently underway, we hypothesize that IT inhibition will have the opposite effect as PT inhibition, and attenuate cocaine sensitization, reward, drug-taking and drug-seeking behaviors.

Conclusions: These studies use cell-specific targeting and novel molecular tools to begin to isolate the contributions of specific cortical pyramidal cell populations in behaviors related to addiction. The findings from these studies support the hypothesis that activity within subtypes of cortical cells can regulate addiction behaviors. Ongoing studies will reveal if PT and IT cells do, indeed, play opposing roles as hypothesized.

Disclosure: Nothing to Disclose.

37.3 The Effect of Prefrontal Grip Deletion on Drug-Seeking Behavior and Physiology

Lisa Briand

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Background: Disrupted glutamatergic signaling is a component in the development and maintenance of addiction. The glutamatergic pathway from the prefrontal cortex (PFC) to the nucleus accumbens plays a critical role in extinction and reinstatement of drug-seeking. The majority of studies examining the trafficking of glutamate receptors have focused on the prefrontal inputs to the nucleus accumbens, rather than changes in the PFC itself. The goal of this work was to examine how disrupted AMPA receptor trafficking in the PFC affects addiction-like behavior. I will discuss work utilizing mice with an inducible deletion of glutamate receptor interacting protein, GRIP, a scaffolding protein necessary for the synaptic localization an aggregation of AMPA receptors.

Methods: Six weeks prior to any behavioral testing, mice received intracranial viral injections of either Cre recombinase or GFP into the medial prefrontal cortex. Following viral expression and knockdown, all mice were trained on a self-administration procedure. Briefly, mice were taught to perform an operant response to receive sucrose pellets. They next underwent an intravenous catheterization surgery, and after recovery, were allowed to perform the acquired operant response for an infusion of cocaine. We next used a progressive ratio schedule to measure motivation to work for the drug, before the mice entered a period of forced abstinence and the behavior was extinguished. Finally, mice experienced a cue reinstatement trial. Following behavioral testing, we took brains to record from either the prefrontal cortex or the nucleus accumbens to examine how GRIP deletion affected electrophysiological outcomes.

Results: Following prefrontal GRIP knockdown, mice exhibited higher breakpoints on a progressive ratio schedule, suggesting that impaired AMPA receptor trafficking in the mPFC increases motivation for drug-seeking behavior. Additionally, prefrontal GRIP KO mice exhibited greater rates of cue-induced reinstatement, indicating that impaired PFC AMPA receptor trafficking increases vulnerability to relapse. No differences were seen in acquisition of the operant task or food self-administration behavior. Prefrontal GRIP deletion alters glutamate transmission in both the PFC and the accumbens.

Conclusions: These findings demonstrate that disrupting AMPA receptor trafficking within the mPFC increases vulnerability to addictive-like behaviors. These findings could lead to an improved understanding of the biological alterations associated with addiction and relapse.

Disclosure: Nothing to Disclose.

37.4 Individual Differences in Neural Signatures of Reward Sensitivity and Inhibition

Harriet de Wit

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Background: Recent behavioral evidence suggests that people who are more sensitive to the rewarding effects of drugs may also exhibit poorer inhibitory control. In our own studies, subjects who report higher liking of a single oral dose of amphetamine also are less able to inhibit a prepotent response on the Stop Task. Both of these traits increase risk for drug abuse. It is not known whether these if overlap in the neural profile of these tendencies.
Methods: Healthy young adults participated in a functional magnetic imaging scans to examine brain activity during tasks designed to detect reward (anticipation/delivery) and inhibition.
Results: We have tested 80 healthy male and female volunteers with a reward task involving both wins and losses, and the Stop Task assessing inhibition. Reward (Wins > Loss) activated the ventral striatum/nucleus accumbens and the medial orbital frontal cortex/ventromedial prefrontal cortex, whereas the inhibitory task activated right prefrontal regions, as well as the anterior insula, cingulate gyrus, and supplementary motor area. We will examine the correlation between these measures in the presentation.
Conclusions: Based on our observed correlations between indices of reward and impulsive behavior we expect that the neural signatures of these processes will also be related.
Disclosure: Nothing to Disclose.

Panel
38. Dissecting Biological Mechanisms of Aggression: Implications for Neuropsychiatric Disease

38.1 Neural Mechanism of Female Aggression

Dayu Lin

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Background: Aggression is essential for competing over resources and defending against threats for all individuals although sexual selection often leads to enhanced aggression in males. Recent studies suggest that the sexually dimorphic aggressive behavior is supported by sexually unique aggression circuit. Specifically, cells in the ventrolateral part of the ventromedial hypothalamus (VMHvl) that express estrogen receptor alpha/progesterone receptor (Esr1/PR) mediate male aggression but not female aggression, leaving the hypothalamic substrate for female aggression unidentified.
Methods: Optogenetic manipulation, Phamacogenetic manipulation, electrophysiological recording, fiber photometry, tracing
Results: We find a clear role of VMHvl Esr1+ cells in female aggression in various ethologically relevant conditions. Using in vivo recording, we find that this population is highly active when females attack naturally. Reversible inactivation of the cells reduces female aggression whereas their activation elicits attack. Moreover, while in males, aggression-related and mating related cells overlap substantially in the VMHvl, in females, aggression- and mating-related cells are largely distinct and concentrated in spatially segregated regions of the VMHvl.
Conclusions: The VMHvl mediates aggression in both sexes, but the spatial arrangement of the mating and fighting populations is sexually dimorphic.
Disclosure: Nothing to Disclose.

38.2 Orexin Signaling Within the Lateral Habenula Controls Aggression

Scott Russo

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

Background: Abnormal social behavior, such as maladaptive aggression or social anxiety is associated with a number of neuropsychiatric disorders including schizophrenia, mood disorders, antisocial personality disorder and autism. Such disruptions in social behavior are thought to result, in part, from inappropriate activation of brain reward systems in response to social stimuli. A series of nuclei within the ventral midbrain that control mood and emotion are known to encode certain aspects of aggressive and nonaggressive social interaction; however, little is known about the neural circuit mechanisms that directly modulate the motivational or rewarding component of social behavior.
Methods: To address this question, we established a mouse behavioral model for investigating individual differences in social behavior and social reward. We used cell type and circuit specific molecular tools along with in vivo monitoring of neural activity and optogenetics to examine the role of lateral habenula (LHb), a major hub within the brain's reward circuit, in controlling social interaction in aggressive (AGG) and non aggressive (NON) mice.
Results: We find approximately 70% of outbred CD-1 mice engage in aggressive behavior with a resident intruder and find such interaction rewarding, whereas the remaining 30% are not aggressive and find intruder interactions aversive. Interestingly, the LHb is differentially activated by intruder-based social interaction in aggressive and nonaggressive mice. We identified a novel orexin (also known as hypocretin) microcircuit within the LHb that differentially regulates LHb neural activity and social behavior in AGG and NON mice. Orexin signaling onto GABAergic neurons within the LHb controls both the initiation of aggression as well as the valence of social interaction during the resident intruder paradigm.
Conclusions: A basic understanding of the neural circuits that control social behavior is absolutely critical for developing new treatment strategies for social deficits in a range of psychiatric illnesses.
Disclosure: Nothing to Disclose.

38.3 Disruption of Brain-Derived Neurotrophic Factor (BDNF) From Promoters I and II, but Not IV and VI, Leads to Increased Aggression and Altered Sex-Specific Social Behaviors in Male and Female Mice

Keri Martinowich

Lieber Institute for Brain Development, Baltimore, Maryland, United States

Background: Brain-derived neurotrophic factor (BDNF) is an activity-dependent neurotrophin that mediates many aspects of brain plasticity. BDNF-deficiency is associated with numerous psychiatric conditions that feature altered social behavior. In rodents, BDNF reduction causes elevated aggression and is associated with impaired social function. BDNF is highly expressed in the hypothalamus, a region critical for the organization of social behaviors. Transcription of BDNF is controlled by several promoters, which drive expression of multiple transcripts encoding an identical protein. The existence of unique BDNF transcripts allows for precise control of BDNF production, but the functional consequences of multiple BDNF transcripts, and how they regulate circuits that control social behaviors are unknown.
Methods: We generated mice in which production of BDNF from promoters I, II, IV and VI is selectively disrupted (BDNF-e1, BDNF-e2, BDNF-e4, and BDNF-e6). We tested these animals on a battery of social behavior assays and used quantitative PCR, ELISA and single molecule in situ hybridization to analyze expression of BDNF transcripts and BDNF protein in cell-specific populations of the hypothalamus.
Results: Disruption of BDNF produced from promoters I and II, but not promoters IV and VI, resulted in abnormal social behaviors. BDNF-e1 and BDNF-e2 animals displayed decreased latency to attack, increased number of fights, and increased number of tail rattles compared to wild-type controls. BDNF-e1 females failed to nurse pups and BDNF-e1 and BDNF-e2 breeders showed significant mating impairments. BDNF-e1 males also attacked foreign pups and estrous females significantly more than wild-type males. BDNF-e4 and BDNF-e6 animals showed normal aggression, maternal care, and reproductive behaviors. Deficits in social behaviors were accompanied by significant decreases in BDNF in the hypothalamus, but not the prefrontal cortex, of BDNF-e1 and BDNF–e2 mice. We further identified exon-1 BDNF transcripts as highly expressed in Esr1-expressing neurons in the ventromedial hypothalamus (VMH), a cell population that is causally implicated in aggression and sex-specific social behaviors.
Conclusions: These data suggest that BDNF promoters I and II play a critical role in regulating social behaviors, likely through regulation of specific neuronal populations in the hypothalamus. Disruption of BDNF from promoter I causes decreased sociability and increased aggression towards other males, pups and females, suggesting that BDNF derived from this promoter plays an important role in regulating social interactions. These studies highlight BDNF as a key molecular player in modulating complex social behaviors such as aggression, reproduction, and parenting.
Disclosure: Nothing to Disclose.

38.4 Compromised Functional Integration and Segregation in Habenula and DMN Nodes in Human Reactive Aggression

Nelly Alia-Klein

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Background: Intermittent-explosive disorder (IED) is characterized by disproportionate reactive aggression. Previously, we have shown that a propensity for physical aggression in healthy adults is linked to heightened resting glucose metabolism in the default-mode network (DMN), which is active at rest. Abnormal resting-state functional connectivity (rsFC) in the default-mode network (DMN) has been suggested as a biomarker of neuropsychiatric disorders that are characterized by emotional instability (e.g., schizophrenia, bipolar disorder, ADHD). Until today it is unclear if rsFC is compromised in reactive aggressive individuals.
Methods: We applied graph theory to identify dysfunctional nodes in RA men ($n = 12$), reporting significant behavioral features of IED including elevated trait aggression and anger, compared to low-aggressive male controls ($n = 12$). All participants underwent a 5-minute resting-state fMRI scan. We assessed differences in complex network properties including (1) global efficiency, indicating functional integration/transfer of data information across the whole network, and (2) clustering coefficient, indicating functional segregation and network robustness of resting state networks by applying graph theory to a high-resolution anatomical template.
Results: Graph theory revealed significantly increased global efficiency in the left temporal pole, and a subcortical-prefrontal network including the right middle frontal gyrus, the right thalamus, and the right habenula in RA individuals compared to controls. Moreover, RA individuals exhibited significantly decreased clustering coefficient ($p = .01$) in the occipital cortex and DMN nodes including the precuneus, the dorsal anterior cingulate cortex, and the dorso-medial prefrontal cortex.
Conclusions: Our findings suggest that an imbalance between heightened functional integration in a subcortical-prefrontal network and diminished functional segregation in DMN nodes at rest may be a biomarker for reactive aggression. Interestingly, the habenula, which showed increased global efficiency in RA individuals, has recently been linked to the valence of aggressive behavior in mice as well as the likelihood of winning fights in zebrafish. Thus, our study provides first support for the role of the habenula in human reactive aggression. Further studies across species are needed to delineate the function of the habenula with respect to the motivation to engage in aggressive behavior.
Disclosure: Nothing to Disclose.

Study Group

39. The National Neuroscience Curriculum Initiative (NNCI) – An Update on New Ways to Communicate Neuroscience

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Harvard Medical School, Boston, Massachusetts, United States

Study Group Summary: Over the past two decades, advances in neuroscience have dramatically enhanced our understanding of the brain and of the neurobiological basis of psychiatric illness. Yet teaching neuroscience remains fraught with challenges: the field is vast and constantly evolving; many programs lack access to faculty with content

expertise; the content itself is not presented in an engaging manner and the clinical relevance is not always clear. At the same time, the way we approach teaching and learning is itself experiencing a paradigm shift. New technological advances and insights from the literature on adult learning theory are ushering in a new era in education. Lecture halls are vacant, textbooks are passé, and Google is the go-to resource. Emerging data confirm what we instinctively understand: the fundamental way in which learners engage with content is also changing and our ability to communicate key findings needs to evolve. The National Neuroscience Curriculum Initiative (NNCI) is an NIH-funded collaboration with the mission of developing and disseminating clinical neuroscience teaching resources to practicing psychiatrists and residents in training. The guiding principles for the design of NNCI resources are to: maintain an integrative, patient-centered approach; use evidence-based principles of adult learning, eschewing traditional lecture formats in favor of innovative and experiential exercises and; ensure that materials can be implemented by anyone, anywhere, regardless of content expertise.

In furthering the aims of the NNCI it has become clear that one of the key barriers to the impactful dissemination of the latest research findings is the manner in which these findings are communicated outside of the scientific community. It is clear that effective communication of scientific findings is a core professional skill. While researchers may receive training and feedback on traditional forms of scientific communication (such as giving scientific talks and writing for journals), relatively little time is devoted to honing the unique skills required to communicate to a broader audience including trainees and other healthcare providers. Making science accessible requires learning to distill complex topics down to their core concepts, to craft a narrative arc around key translational applications, to optimize the visual representation of data, and to attend to technical aspects of the performance. While challenging to learn, such skills are invaluable for disseminating key research findings and developing engaging and impactful teaching.

In this session participants will be introduced to more of the new learning approaches developed by the NNCI to engage trainees in active learning. We will review core principles of communication and then present qualitative and quantitative data illustrating the success of different approaches for engaging diverse audiences. Our goal with this study group is to foster a discussion around novel strategies for making neuroscience teaching engaging and relevant to clinical practice in psychiatry.

Disclosure: Part 1: Resilience Therapeutics, Advisory Board, Biogen, Consultant.

Panel
40. Circadian Rhythms, Sleep and the Adolescent Brain

40.1 Longer Circadian Period may Arise in Early Adolescence and lay the Foundation for Later Phase Delay

Mary Carskadon

Alpert Medical School, Brown University, Providence, Rhode Island, United States

Background: A delay in circadian phase preference (chronotype) across adolescent development is well known. Because adolescence is time when many psychiatric disorders emerge and risks are accentuated by late chronotype (e.g., substance abuse, depression), understanding the biology of the adolescent delay is important. Most striking evidence for the delay shift is seen in a 2- to 3-hour delay in the timing of midsleep on "free nights" across the second decade. Biological factors underlying this delay are not identified, though two candidates are likely: lengthening of intrinsic circadian period and modification in the phase response to light. These changes were once thought to arise around the time of midpuberty; however, recent data show them starting earlier. We previously published data showing early modulation of melatonin response to evening light. New here is evidence for lengthening of circadian period in early pubertal development.

Methods: Intrinsic period was estimated from serial determinations of melatonin onset phase from forced desynchrony (FD) protocols, where participants live in the lab on a non-24-hour schedule that permits intrinsic period to break free from 24 hours. Two different FD schedules were used in these participants: 20-h FD and 28-h FD. A total of 90 protocols were accomplished in participants ranging in age from 9 to 25 years. The 20-h protocol included only participants ages ≥ 21. The sample was divided into 3 groups based on Tanner stage (<3 and ≥ 3) and age (<21 and ≥ 21). Participants in the three groups did not differ in sex, race, and ethnicity. The influence of age on period was estimated by regressing period on participant age, which was group-mean centered within each group.

Results: Mean period length was longer in the two youngest groups (Tanner <3 ($n=21$) mean = 24.29, SE = 0.21; Tanner ≥ 3 ($n=43$) mean = 22.24, SE = .23) than in the group age ≥ 21 ($n=26$) (mean = 24.17, SE = .22). The lengthening of period as a function of age was greater for the children in the younger age group of Tanner stage <3 than in the more mature group of youngsters. The rate of change per year of age was statistically significant in the less mature (0.077 h; CI = .005-0.145) and not statistically significant in the more mature youngsters (.050 h; CI = -.015-0.118) and the older participants (-0.021 y; CI = -.012 - 0.071).

Conclusions: These data confirm that lengthening of circadian period begins before midpuberty. In combination with our finding that sensitivity to evening light is greater in pre- and early pubertal youngsters, these results align well with a hypothesis that the adolescent biobehavioral phase delay has its impetus from these relatively early biological changes. When phase is later, young people also endure challenges to obtain adequate sleep when school bells ring early in the morning. Insufficient sleep also represents a factor increasing risk for psychiatric disorders that emerge during adolescence.

Disclosure: Nothing to Disclose.

40.2 Experimental Sleep Restriction Reduces Adolescents' Striatal Response to Monetary Reward With Consequences for Depressed Mood

Erika Forbes

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: Insufficient sleep is common among adolescents in the U.S. and is associated with a variety of problematic health and mental health outcomes, including depression and substance use. Our previous work has indicated that sleep problems in childhood and early adolescence are associated with later depression and substance use problems. A putative mechanism of the effects of insufficient sleep is altered function in neural reward circuitry. We conducted an experimental sleep manipulation in the laboratory to examine the effects of sleep deprivation on reward circuitry and mood symptoms in community adolescents.
Methods: Using a within-subjects design, 35 adolescents (ages 11.5-15 years) underwent an fMRI scan following two nights of sleep extension (SE: 10 hours time in bed) and two nights of sleep restriction (SR: 4 hours time in bed) under controlled laboratory conditions (counterbalanced across participants). Participants completed visual analog scales on subjective affect (Buysse et al., 2007) 3 times/day and the Children's Depressive Inventory (Kovacs, 2004) once per lab visit. They underwent functional magnetic resonance imaging on a Siemens 3T scanner during an event-related guessing task with high- and low-magnitude monetary reward after the second night of each lab visit. Data were preprocessed and analyzed in Analysis of Functional NeuroImages (AFNI; afni.nimh.nih.gov). Analyses focused on sleep condition X reward magnitude effects on BOLD response, and association between resulting clusters and positive affect and depression. Control for multiple testing included a height threshold of $p < .005$ and a cluster-level threshold using false-discovery rate at $p < .05$ with 3dttest++.
Results: During reward outcome, adolescents exhibited lower anterior putamen response after SR than after SE (p corrected $< .05$), but this was limited to the high-magnitude reward condition. During reward anticipation and low-magnitude reward outcome, adolescents' neural response did not differ by sleep condition. The effect of SR on putamen response was stronger in younger participants ($r = -.35$, $p = 0.04$). Brain-affect associations were unmasked by SR, after which greater ventral striatal response was related to positive mood. Sleep condition moderated associations between striatal response and depression, with a relation between lower putamen response and higher depressive symptoms after SR only (61 voxels, p corrected $= .04$).
Conclusions: Acute sleep loss during early/mid adolescence disrupts neural response to reward and could influence depression via function in reward circuitry. In particular, sleep loss could lead to dampened striatal responding to high-magnitude rewards. Chronically insufficient sleep in adolescence may alter the development of neural reward systems more substantially, leading to problems such as depression and substance abuse with development. Behavioral interventions targeting sufficient sleep could be applied to prevent these outcomes, with effects potentially occurring through the mechanism of flexible function in reward circuitry.
Disclosure: Nothing to Disclose.

40.3 The Circadian Clock Regulates Neuronal Maturation and Function in the PFC

Colleen McClung

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Background: Adolescence is a time for increased risk for the development of psychiatric disorders including depression, bipolar disorder, schizophrenia and substance abuse. During adolescence, there is a biological shift in circadian rhythms leading to an evening chronotype. However, due to school start times and modern lifestyles, teenagers are in a perpetual state of circadian de-synchrony likely resulting in the disruption of normal molecular rhythms in the brain. This may have serious consequences in the neuronal development of vulnerable individuals and contribute to the development of psychiatric disorders. However, surprisingly little is known about the role of circadian genes in neuronal development through adolescence and how disruptions to this system impacts cognitive, mood and reward-related circuitry.
Methods: In these studies, we used mice which have a mutation in the Clock gene, one of the central regulators of circadian rhythms. We performed immunohistochemical measures of neuron development and measures of oxidative stress. We also performed measures of mitochondrial function respiration. In addition, we performed a number of electrophysiological (in vivo and ex vivo), molecular and behavioral analyses.
Results: We find that the loss of Clock gene function results in profound changes in the development of prefrontal cortical microcircuitry across adolescence and into adulthood. Specifically, GABAergic interneurons that express parvalbumin (PV) normally show an increase in PV expression and perineural net formation from P20-P90, indicative of neuronal maturation. In contrast, Clock mutants have a progressive decrease in PV expression ($P < 0.01$) and fail to show maturation in perineuronal nets across development ($P < 0.05$). These changes are associated with increased oxidative stress specifically in PV cells in the Clock mutant mice across adolescence ($P < 0.01$) and decreased mitochondrial function ($P < 0.05$). Associated with these changes, the mice display significantly increased exploratory drive and risk-taking behavior, as well as electrophysiological changes indicative of altered oscillatory activity in the gamma, beta and theta ranges during exploratory tasks across brain regions, and altered synaptic plasticity in striatal regions which results in decreased glutamatergic signaling.
Conclusions: We find that disruption of the circadian clock has a particularly strong impact on the function and maturation of PV expressing interneurons in the mPFC. Interestingly, human postmortem studies suggest that these cells are particularly compromised in individuals with schizophrenia and that deficits like these will alter the excitatory/inhibitory balance in the cortex. The CLOCK protein directly regulates several mitochondrial and antioxidant genes and these changes to PV cell maturation are likely due to the progressive increase we see in oxidative stress in these neurons. These changes lead to lasting effects on neuronal activity and behavior. Taken together, these studies help define the consequences of circadian clock disruption to neuronal maturation and function across adolescence into adulthood, and shed light on how clock disruption is associated with the development of psychiatric diseases.

Disclosure: Nothing to Disclose.

40.4 Modifying the Impact of Eveningness Chronotype in Adolescence on Sleep, Circadian and Risk Outcomes

Allison Harvey

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Background: Adolescence is a one of the most important developmental stages and a time of great vulnerability. There is evidence that the onset of puberty triggers a general preference for eveningness. Evening chronotype ('night-owls') adolescents follow a delayed sleep-wake schedule, increasing mental and/or physical activity later in the day, compared to morning chronotypes ('larks'). The evening preference has been identified as a contributing factor for poorer health across multiple domains (emotional, cognitive, behavioral, social, physical). A 'treatment experiment' will be described in which a psychosocial intervention (Transdiagnostic Sleep and Circadian Intervention; TranS-C-Youth) was administered to test the hypothesis that reducing eveningness will improve sleep and circadian functioning and reduce risk.
Methods: Youth aged 10 to 18 with an evening chronotype were randomized to: (a) TranS-C ($n = 89$) or (b) Psychoeducation (PE; $n = 87$). Treatments were 6 individual, weekly 50-minute sessions during the school year. Using multiple methods (global and prospective self-report, dim light melatonin onset, ecological momentary assessment) and multiple informers (adolescents, parents), outcomes were assessed by blind assessors pre-treatment and post-treatment.
Results: Relative to PE, TranS-C was associated with less evening circadian preference, earlier endogenous circadian phase (dim light melatonin onset; DLMO), less weeknight-weekend discrepancy in Total Sleep Time (TST) and wakeup time, less daytime sleepiness, and better self-reported sleep via youth and parent report. In terms of risk outcomes, relative to PE, TranS-C was not associated with greater pre-post change on the primary outcome. However, there was no group difference for total sleep time or bedtime on weeknights. There were significant interactions favoring TranS-C on the Parent-Reported Composite Risk Scores for cognitive health and selected other risk outcomes.
Conclusions: Relative to PE, TranS-C was associated with improvement on selected sleep, circadian and risk outcomes.
Disclosure: Nothing to Disclose.

Panel
41. Fatty Acid Amide Hydrolase (FAAH) as a Therapeutic Target in Psychiatric and Addictive Disorders

41.1 Fatty Acid Amide Hydrolase: Past, Present and Future

Daniele Piomelli

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

Background: Fatty acid amide hydrolase (FAAH) is an intracellular serine hydrolase that catalyzes the deactivation of the endocannabinoid neurotransmitter, anandamide. Anandamide is released by neurons and other cells of the brain and activates CB1-type cannabinoid receptors on presynaptic nerve terminals to regulate social behavior and the emotional and somatic response to stress. Pharmacological agents that selectively inhibit FAAH activity exert profound pro-social, anxiolytic-like and antidepressant-like effects in animal models. By acting on CB1 receptors of peripheral nociceptive neurons, these agents can also gate the entry of pain stimuli into the central nervous system.
Methods: Our laboratory has used a combination of medicinal chemistry, molecular pharmacology, and behavioral approaches to discover novel FAAH inhibitors, characterize their pharmacological properties, and identify pathological conditions to which they might potentially be applied.
Results: The first systemically active FAAH inhibitor, URB597, was discovered by our laboratory in 2002. This compound, which acts by covalently binding to FAAH's catalytic serine, has been widely used in animal models to probe the physiological functions of anandamide and to validate FAAH as a therapeutic target. Preclinical studies have documented its pro-social, anxiolytic-like, antidepressant-like and analgesic properties, as well as its ability to alleviate nicotine addiction. Since the discovery of URB597, a large number of chemically distinct FAAH inhibitors have been disclosed in the scientific and patent literature, and several such inhibitors have been advanced to clinical development for the treatment of anxiety, depression, pain, and addiction. The armamentarium of FAAH inhibitors has been recently enriched by novel compounds that selectively inhibit FAAH in the periphery of the body (URB937), or simultaneously inhibit FAAH and other functionally-related targets (e.g., cyclooxygenase, ARN2508). URB937 and ARN2508 have demonstrated marked analgesic and anti-inflammatory properties, respectively.
Conclusions: Selective FAAH inhibitors may offer a new approach to the treatment of pathological states in which social reward and the response to stress are impaired, including anxiety, depression, and autism spectrum disorders. They may also find application in the treatment of addiction disorders, pain and inflammation.
Disclosure: Part 1: NeoKera Pharmaceuticals, Stock / Equity, Therapix, Advisory Board,
Part 2: NeoKera Pharmaceuticals, Consultant.

41.2 PF-04457845: From Design to Clinic, an Interesting Journey

Zoe Hughes

Pfizer, Inc., Cambridge, Massachusetts, United States

Background: The fatty acid amide hydrolase (FAAH) enzyme plays a critical role in regulating one of the predominant endogenous ligands for cannabinoid type 1 receptors (CB1R). CB1R are one of the most abundant receptors in the CNS and are known to be involved in the regulation of a number of fundamental biological processes, from anxiety to food intake and pain. While targeting CB1 receptors directly has been associated with serious challenges, activating CB1 through increasing levels of anandamide, an endogenous CB1 ligand offers an attractive alternative. Numerous FAAH inhibitors have been under

investigation for a range of different indications and have reached various stages of development. Here the careful translation of findings from preclinical species to the clinic performed in order to determine an appropriate human dosing paradigm for PF-04457845 will be described.
Methods: The potency (kinact/Ki value in the glutamate dehydrogenase-coupled FAAH assay) and selectivity of PF-04457845 against a broad range of targets were established using a traditional 68 target screening panel plus an extensive competitive activity-based protein profiling screen of serine hydrolases. Modulation of biomarkers in rodents and humans were measured using an ex vivo FAAH enzyme activity assay using isolated leucocytes or brain homogenate (rat only) to measure labeled ethanolamine produced from labeled AEA. Brain (rat only) and plasma compound and fatty acid amide levels were also measured using LC/MS/MS. [18F]PF-9811 PET imaging was used to demonstrate CNS binding of PF-04457845 to FAAH in the brain. Finally, electroencephalography (EEG) and electromyography (EMG) signals were recorded from rats, and polysomnography recordings were made from healthy human subjects, following administration of PF-04457845.
Results: PF-04457845 was demonstrated to have high in vitro potency (kinact/Ki value: $40{,}300 \pm 11{,}000$ $M-1\,s-1$) for inhibition of FAAH and an exquisite selectivity profile (~14,000 fold) against mouse and human proteomes. Through measurement of exposure levels, increases in anandamide and inhibition of FAAH activity or binding, PF-04457845 was shown to have dose dependent effects associated with central and peripheral FAAH inhibition. Through measurement of these key biomarkers in humans the relationships between central and peripheral compartments as well as between rodents and humans were established and used to determine the optimal clinical dosing paradigm. Demonstrating that PF-04457845 selectively suppressed REM sleep in rats and humans, and caused a significant increase in total sleep time and reduction in Stage N1 sleep in humans allowed us to confirm that inhibition of FAAH in the brain had a functional effect in humans.
Conclusions: The data presented demonstrate that PF-04457845 is a highly potent and selective FAAH inhibitor. The non-clinical and clinical programs for PF-04457845 provide a thorough understanding of the PK/PD relationship for the molecule. Through the course of an extensive preclinical toxicology evaluation and 7 Pfizer sponsored clinical studies PF-04457845 has been found to be safe and well tolerated.
Disclosure: Part 1: Pfizer Inc., Employee, Self, Biogen, Employee (Spouse), **Part 5:** Pfizer Inc., Employee, Biogen, Employee (Spouse).

41.3 Fatty Acid Amide Hydrolase FAAH C385A Functional Gene Variation Validates FAAH Inhibition as a Therapeutic Mechanism to Promote Fear Extinction

Leah Mayo

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Background: Dysregulation of stress- and threat-responding can lead to psychopathologies such as anxiety disorders and PTSD. Components of the networks responsible for stress- and threat-responding overlap with the endocannabinoid (eCB) system in anatomical location and physiological activation, suggesting a role of the eCB system as a stress buffer. In fact, most people who take THC report doing so for its stress-dampening effects. However, the indiscriminate activity of cannabinoid receptor (CBR) ligands produces unwanted side effects and limits their therapeutic potential. Preclinical studies have suggested that fatty acid amide hydrolase (FAAH), the main catabolic enzyme for the endocannabinoid anandamide (AEA) may offer an attractive therapeutic target, since FAAH inhibition amplifies the physiologically-relevant, endogenous AEA signal without the unwanted side effects of indiscriminate CBR activation. In preclinical models, FAAH inhibition facilitates fear extinction and protects against the anxiogenic effects of stress. Behavioral genetics studies offer an opportunity to validate this mechanism in humans. Approximately 38% of people of European descent carry a loss-of-function mutation in the FAAH gene (FAAH C385A), which renders the gene product more vulnerable to degradation. These A-allele carriers have higher levels of AEA than non-carriers and evidence suggests that they demonstrate attenuated threat responding. However, due to the rarity of A/A homozygotes, studies to date have only assessed A/C heterozygotes, and effect sizes have been limited. Here, we used a prospective genotyping approach to enrich for the rare A/A genotype, allowing us to examine whether there are gene-dose effects of this functional variant on stress- and threat-responding in humans. This would provide compelling evidence in support of FAAH inhibition as a therapeutic.
Methods: We screened several hundred individuals to recruit 25 of each FAAH C385A genotype (C/C, A/C, A/A) and exposed them to a 2- day behavioral and psychophysiological paradigm. Participants completed a fear conditioning and extinction task assessed via potentiation of the eyeblink component of the startle reflex. We also assessed affective responses to emotional images using facial electromyography of the corrugator ("frown") and zygomatic ("smile") muscles. Participants then completed a control or stress task followed by another affective images task to assess stress-induced changes in affective responding. Blood samples were taken regularly to assess changes in cortisol and eCB levels.
Results: The functional role of FAAH C385 A allelic variation was confirmed by analysis of circulating AEA levels. The variant produced a gene-dose-dependent effect on the extinction of conditioned fear ($A/A > A/C > C/C$) with a large effect size (A/A vs C/C: Cohen's $d = 1.2$). Differences between genotypes were specific to the extinction of fear, producing no effect on fear acquisition or responding. We found no genotype effect on the response to emotional images, but did find a protective effect against increased baseline or "resting" corrugator activity following stress. Thus, the FAAH 385A variant is also associated with attenuated stress-induced negative affect in a gene-dose-dependent manner.
Conclusions: FAAH modulation appears to play a pivotal role in stress- and fear-related behaviors in healthy humans and is thus a prime candidate for interventions aimed at clinical populations with dysregulation of such behaviors. In light of these encouraging findings, a proof-of-concept study

is underway to determine the effect of pharmacological manipulation of FAAH levels in healthy humans.
Disclosure: Nothing to Disclose.

41.4 FAAH Inhibitor PF-04457845 in the Treatment of Cannabis Dependence

Deepak D'Souza

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Background: Cannabis is the most widely used illicit substance in the United States. Cannabis use disorder (CUD) is a well-recognized syndrome characterized by tolerance and withdrawal. There are no FDA -approved or clinically-proven treatments for CUD. In attempting to quit, many experience a cannabis withdrawal syndrome (CWS) which is characterized by craving for cannabis, irritability, anger, depression, sleep disturbance, and decreased appetite and weight that begins soon after abstinence lasts for up to weeks. Importantly sleep disturbance, a prominent feature of CWS, is accompanied by alterations in sleep architecture evident on polysomnography (PSG), and this disturbance is associated with relapse. CWS makes it more difficult to quit and maintain abstinence. Furthermore, CUD individuals may use cannabis to relieve or avoid CWS.
Individuals with CUD have 1) lower brain cannabinoid receptors (CB1R) and 2) lower levels of the eCB-metabolizing enzyme fatty acid amide hydrolase (FAAH) which is responsible for degrading anandamide (AEA). Lower brain FAAH levels may be a consequence of cannabis exposure and may contribute to cannabis withdrawal. Potentiating eCB tone may be one approach to treating CUD. FAAH inhibitors which increase AEA levels can reset eCB tone and reduce CWS in THC-dependent animals. Compared to THC or cannabis, FAAH-inhibitors do not have psychoactive effects, are not rewarding, do not increase the abuse liability of other addictive drugs, and are not associated with tolerance. PF-04457845 is an orally active, long-acting, potent, and selective FAAH inhibitor.
Hypothesis: PF-04457845 was hypothesized to reduce CWS, sleep architectural abnormalities and cannabis use, in individuals with a cannabis use disorder (CUD).
Methods: In a double-blind, parallel group study, DSM-IV cannabis dependent subjects were randomized to receive either PF-04457845 (4 mg) or placebo in a 2:1 ratio. Subjects were hospitalized on a locked, smoke-free, inpatient research unit for ~ 1 week to force abstinence before being discharged to complete the remainder of the 4-week study as outpatients. Cannabis withdrawal, visual analog measures of mood states, self-reported and laboratory measures of cannabis use, self-reported and polysomnographic measures of sleep, drug and endocannabinoid levels, and safety data were collected. Adherence to once daily PF-04457845 (4 mg QD) or placebo was witnessed in person during the inpatient phase, and daily during weekdays during the outpatient phase using videocalling.
Results: Complete data was obtained in 58 male subjects with DSM-IV cannabis dependence. Adherence to study medication was confirmed visually to be at least 95% and was corroborated by plasma drug levels and weekly pill counts. Target engagement was demonstrated by increased plasma AEA levels in the group randomized to PF-04457845. Relative to placebo, the FAAH inhibitor PF-04457845 (4 mg QD) administered for 4 weeks reduced 1) cannabis withdrawal measured by the Withdrawal Discomfort Score of the cannabis withdrawal scale, 2) mood disturbances measured by a Visual Analog Scale, 3) cannabis use measured by the Time Line Follow Back approach and urinary THC-COOH levels, and 4) in stage N3 sleep measured by polysomnography (PSG). PF-04457845 was very well-tolerated and did not have any effects that were rewarding/reinforcing.
Conclusions: FAAH Inhibition reduced cannabis withdrawal, use and sleep disturbances. The promising results of this study warrant further study of FAAH-Is in the treatment of CUD.
Disclosure: Nothing to Disclose.

Mini Panel
42. NMDA Regulation of GABA Interneurons: From Circuits to New Therapies

42.1 NMDA Receptor Positive Allosteric Modulation Regulates Network Oscillations and Ameliorates Aberrant Brain Activity in Dravet Syndrome and Alzheimer's Disease Models

Jesse Hanson

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Background: NMDA receptors (NMDARs) play critical roles in synaptic and circuit functions and are implicated in neuropsychiatric and neurodegenerative disorders. Currently, the in vivo effects and therapeutic potential of direct pharmacological enhancement of NMDAR function are largely unknown. We have recently developed a positive allosteric modulator (PAM) of GluN2A subunit-containing NMDARs, GNE-0723, which has properties allowing for in vivo NMDAR enhancement using oral administration.
Methods: Synaptic NMDAR current potentiation was examined ex vivo using whole-cell recordings from excitatory and inhibitory neurons in brain slices. Effects of in vivo treatment with the NMDAR PAM, GNE-0723, were compared to effects of the psychotomimetic NMDAR antagonist, (+)-MK-801, using locomotor assays and EEG recordings. EEG was also used to measure the effects of GNE-0273 treatment on epileptiform activity in the Scn1a-KI loss-of-function model of Dravet syndrome epileptic encephalopathy and in the J20 transgenic APP model of Alzheimer's disease.
Results: GNE-0723 enhanced synaptic NMDAR currents in neurons including parvalbumin interneurons, suggesting that the in vivo effects of NMDAR PAM treatment could involve interneuron activation. In contrast to the increased locomotion caused by NMDAR antagonist treatment, GNE-0723 caused a reduction in locomotion. GNE-0723 treatment also had opposite effects as NMDAR antagonist treatment on brain oscillations, and caused a dose-dependent decrease in broadband EEG oscillatory power. Notably,

GNE-0723 reduced low frequency oscillation power even at doses that did not affect locomotion. Consequently, we tested GNE-0723 treatment in mouse models of Dravet syndrome and Alzheimer's disease, which exhibit hyper-synchronous oscillatory activity driven by interneuron dysfunction. GNE-0723 treatment reversed elevated low-frequency oscillatory power and reduced epileptiform discharges in these mouse models.
Conclusions: These results suggest that NMDAR PAMs could have therapeutic potential not only in disorders like schizophrenia where the pathophysiology is thought to involve NMDAR hypofunction, but more broadly in diseases involving interneuron dysfunction and network hyperactivation, including Dravet syndrome and Alzheimer's disease.
Disclosure: Part 5: Genentech, Inc., Employee.

42.2 Enhancing PV Interneuron Function Through Modulation of Kv3 Ion Channels

Charles Large

Autifony Therapeutics, Stevenage, United Kingdom

Background: Accumulating evidence supports a central role for fast spiking GABAergic interneurons in the pathophysiology of schizophrenia. Dysfunction of these interneurons, associated with reduced parvalbumin (PV) expression, leads to disinhibition of cortical circuitry, dysregulation of gamma oscillations, and contributes to cognitive deficits in patients with schizophrenia (Lewis et al. TiNS 2012; 35: 57–67). Reduced function of PV interneurons can be induced by acute or sub-chronic treatment with NMDA receptor antagonists. These models present an opportunity to evaluate targets that might enhance PV interneuron function as potential treatments for schizophrenia. Voltage gated Kv3.1 potassium channels are selectively expressed on PV interneurons in cortical circuits, where they permit rapid and accurate firing required to synchronise neural networks at gamma frequencies. Kv3.1 channel expression has been shown to be reduced in un-medicated patients with schizophrenia (Yanagi et al. Mol Psychiatry 2014; 19: 573-579). Modulation of Kv3.1 may provide a means to restore PV interneuron function in schizophrenia patients.
Methods: Adult female Lister Hooded rats received phencyclidine (scPCP, 2 mg/kg) for 7 days, followed by 6 weeks' washout. This procedure produces enduring cognitive and social behaviour deficits associated with reduced PV in prefrontal cortex and hippocampus (Neill et al. Pharmacol & Ther. 2010; 128(3): 419-432). Rats were tested in novel object recognition (NOR) and attentional set shifting for cognition. Social behaviour was also evaluated. Prefrontal cortical slices of the prelimbic and infralimbic regions were prepared from scPCP-treated rats to explore drug effects on kainate/carbachol-induced fast (20-80 Hz) network oscillations. Human frontal or temporal neocortex slices were also obtained from patients undergoing elective brain surgery and exposed to kainate (KA) or KA+PCP. Finally, adult drug naïve rats were acutely administered ketamine and BOLD signals recorded using functional MRI, following pre-treatment with either a Kv3.1 modulator or vehicle.
Results: The selective Kv3.1 positive modulator, AUT00206 significantly enhanced the power of fast network oscillations by $26.1 \pm 8.1\%$ ($n = 11$, $p < 0.01$) in prelimbic cortex and by $19.7 \pm 8.4\%$ ($n = 19$, $p < 0.05$) in infralimbic cortex from scPCP treated rats. The drug also enhanced gamma oscillations by $40.4 \pm 12.5\%$ ($n = 3$) in human neocortical slices treated with KA+PCP. Acute treatment with AUT00206 dose-dependently restored NOR and social behaviour deficits induced by scPCP. A significant and selective deficit in the extra dimensional shift phase of the ASST ($P < 0.001$) in scPCP rats was significantly attenuated by 14- day treatment with AUT00206 alone ($P < 0.001$) or when added on to haloperidol ($P < 0.001$). 21- day treatment with AUT00206 also restored levels of PV in hippocampus and frontal cortex. Finally, acute AUT00206 significantly prevented the BOLD response induced by acute ketamine across cortical and subcortical brain areas.
Conclusions: Selective activation of Kv3.1 channels can restore cognitive, social and BOLD signal changes induced by exposure to NMDA antagonists. The effects of AUT00206 ex vivo? on gamma synchrony and on PV levels after chronic dosing are consistent with effects of the drug to enhance PV neuron activity. These results suggest potential for a novel approach for the treatment of schizophrenia.
Disclosure: Part 1: Autifony Therapeutics Limited, Board Member, Autifony Therapeutics Limited, Stock / Equity, **Part 2:** Autifony Therapeutics Limited, Employee, **Part 3:** Autfifony Therapeutics Limited, Board Member, **Part 5:** Autifony Therapeutics Limited, Employee.

42.3 Excitatory-Inhibitory Balance in Schizophrenia: In Vivo DREADD Pre-Clinical and Combined PET-MR Clinical Imaging Findings

Michelle Kokkinou

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Background: NMDA hypofunction on parvalbumin positive GABAergic interneurons is thought to dysregulate the glutamatergic control of midbrain dopamine neurons to lead to psychosis. However, it is unknown if NMDA hypofunction results in the same dopaminergic changes seen in schizophrenia, if this is mediated by parvalbumin positive GABAergic interneurons, or if dopamine and glutamate function are linked in patients.
Methods: Clinical experiment: 30 first episode drug naïve/free patients and 20 controls received 18F-DOPA and [1H]-MRS imaging to measure striatal dopamine synthesis capacity and cortical glutamate levels respectively prior to receiving clinical follow-up to determine response to treatment.
Preclinical Experiments:
Experiment 1: Twenty-eight mice received a sub-anaesthetic dose of ketamine or saline for five consecutive days and locomotor activity was assessed in the open field test and striatal dopamine synthesis capacity (indexed as the influx rate constant Kimod) via 3,4-dihydroxy-6-[(18)F]-fluoro-l-phenylalanine positron emission tomography (PET) was measured in-vivo.
Experiment 2: Midbrain dopamine neurons were transduced with adeno-associated virus vector expressing Gi-coupled

(hM4Di) DREADDs (designer receptor exclusively activated by designed drug) under control of a dopamine transporter (DAT) promoter in forty-three DATCre positive mice. Two weeks following the stereotaxic injection of the viral construct, mice were pre-treated with clozapine N-oxide (CNO) to selectively study the effect of neuronal firing on locomotor activity and striatal dopamine synthesis capacity in the sub-chronic ketamine model.

Results: Clinical experiment: In patients, but not controls, frontal glutamate levels were negatively correlated with striatal dopamine synthesis capacity ($r = -0.42$, $p = 0.03$), which predicted subsequent response to treatment ($r = 0.6$, $p < 0.05$).

Preclinical Experiments:

Sub-chronic ketamine treatment significantly increased locomotor activity ($p < 0.01$) and increased striatal dopamine synthesis capacity ($p < 0.05$, effect size > 1.1) compared to saline treated controls. Ketamine's effects on locomotor activity and dopamine synthesis capacity were reversed by DREADD-mediated inhibition of midbrain dopamine neuron firing prior to ketamine administration (control comparison: $p < 0.01$).

Conclusions: Our data indicate that the glutamatergic regulation of striatal dopamine is altered in psychosis, and show that sub-chronic ketamine treatment results in the same striatal dopamine synthesis capacity alterations seen in patients via a mechanism that requires midbrain dopamine neuronal activity.

Disclosure: Nothing to Disclose.

Panel
43. Optimizing the Actions of Opioids for Pain and Depression

43.1 Tianeptine: A Treatment for Emotional Pain

Rene Hen

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Background: Our recent work demonstrates that the antidepressant tianeptine (Stablon®, Coaxil®, Tatinol®) is a full agonist at the Mu opiate receptor (MOR) (Gassaway et al., 2014) (Samuels et al., 2017).

Methods: To localize the site of expression of MOR that is responsible for the antidepressant effect of tianeptine, we took advantage of mice containing a floxed MOR locus. These mice were bred with VGAT-cre mice that express the cre recombinase specifically in GABAergic inhibitory neurons.

Results: The resulting double transgenic mice no longer displayed a response to tianeptine in the forced swim test but still responded to tianeptine in the open field, feeding test, analgesia test and conditioned place preference. These results indicate that MOR expressed on GABAergic neurons are critical for the antidepressant effects of tianeptine. We are currently assessing which brain structure is critical by using AAV viruses expressing a cre recombinase and by targeting candidate structures such as the insula and the anterior cingulate cortex. These structures have been implicated in the emotional reactions to pain, and there is some evidence that they are part of a circuit that represents both physical pain, and emotional pain, which has been proposed to be one of the symptoms of depression (Hsu et al., 2015).

Conclusions: It is interesting in that respect that tianeptine and opiates in general, may impact both physical pain and emotional pain. The fact that tianeptine acts on a different molecular target, and on different brain circuits than SSRIs suggests that tianeptine or tianeptine analogues may be an effective treatment option for patients who do not respond to SSRIs.

Disclosure: Nothing to Disclose.

43.2 Targeting RGSz1 to Optimize the Analgesic Actions of Opioids

Venetia Zachariou

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Background: Opioid analgesics have been prescribed for the treatment of many chronic pain conditions, but their long term use is highly problematic as it is accompanied by several adverse effects, the development of analgesic tolerance, dependence and addiction. As research efforts are directed towards the development of more efficient and safer morphine-like compounds it is important to comprehend the brain region and cell type-specific intracellular pathways mediating the analgesic versus rewarding/reinforcing effects of mu opioid receptor (MOPR) agonists. We focus on the MOPR signal transduction modulator RGSz1 (Regulator of G protein signaling z1), which is expressed in the periaqueductal gray (PAG) as well as in the basal ganglia. RGSs are potent modulators of GPCR function that control the activation of Galpha and/or Gbetagamma effectors, and therefore the amplitude and direction of signal transduction. Since MOPR function is regulated by distinct RGS complexes in various brain regions, we aim to target RGSz1 in order to dissociate the analgesic from the rewarding actions of opioids.

Methods: We used conditional knockout models, behavioral paradigms of addiction and analgesia, to study the actions of RGSz1 in the PAG in pain-free and chronic pain states. We also applied RNA Sequencing and IPA pathway analysis to gain insight on the gene expression adaptations in the PAG under states of morphine tolerance. Information from these studies was used along with western blot, nucleo-cytoplasmic fractionation and co-immunoprecipitation assays to delineate the mechanism by which RGSz1 complexes modulate morphine tolerance.

Results: Results: we show that prevention of RGSz1 action in the periaqueductal gray enhances the analgesic actions of opioids and delays the development of morphine tolerance. While RGSz1 enhances the analgesic effects of opioids, it does not promote addiction-related behaviors. RGSz1KO mice are less sensitive to the rewarding and locomotor activating effects of morphine. Furthemore, deletion of the RGSz1 gene does not affect the development of morphine dependence. Using RNA sequencing we identified several signal transduction pathways affected by chronic morphine treatment, including the Wnt/beta-catenin pathway. Our biochemical assays show that RGSz1 controls the stability of Axin2/beta-catenin in the nucleus. In the periaqueductal

gray, RGSz1 completes with Axin2 for binding to Galphaz, an effect that impacts beta-catenin-mediated transcription. This mechanism occurs only in analgesia related networks, whereas in the VTA and NAc, RGSz1 acts as a positive modulator of MOPR function.
Conclusions: Targeting RGSz1 complexes in the brain may enhance the analgesic actions of opioids and prevent the development of tolerance and addiction-related behaviors. Our genomic and biochemical findings provide novel information on molecules and pathways that can be targeted to for more efficient treatment of chronic pain states.
Disclosure: Nothing to Disclose.

43.3 Dissecting the Functional Organization of Opioid Receptors in Pain Neural Circuits to Optimize Opioid Analgesic Therapies

Gregory Scherrer

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Background: Opioids represent the mainstay treatment for the management of severe acute, perioperative, and chronic pain. However, opioids also generate numerous detrimental side effects that can significantly complicate their use, including analgesic tolerance, opioid-induced hyperalgesia (OIH), and transition to addiction. Additionally, opioid analgesics used in the clinic are limited to mu opioid receptor agonists, and show limited utility against certain types of pain (e.g. neuropathic tactile allodynia). Strikingly, despite opioids having been used in human medicine for millenaries, the mechanisms underlying their analgesic versus detrimental properties remain largely unclear. Elucidating the functional organization of opioid receptors in neural circuits and opioids mechanisms of action is urgently needed to develop innovative analgesic therapies with limited side effects.
Methods: We use a combination of mouse genetics, molecular biology, neuroanatomy, electrophysiology, behavior, opto- and chemogenetics. With these approaches, we identify the neural circuits and cells that express opioid peptides and the delta, kappa, mu opioid receptors (DOR, MOR, KOR), and nociceptin/orphanin FQ (NOP) receptor and determine their function and signaling mechanisms.
Results: We found that MOR expressed by primary afferent nociceptors initiate analgesic tolerance and OIH development. RNA-sequencing and histological analysis revealed that MOR is expressed by nociceptors, but not by spinal microglia. Deletion of MOR specifically in nociceptors eliminated morphine tolerance, OIH, and pronociceptive synaptic long-term potentiation, without altering antinociception. Furthermore, we found that co-administration of methylnaltrexone bromide, a peripherally restricted MOR antagonist, is sufficient to abrogate morphine tolerance and OIH without diminishing antinociception in perioperative and chronic pain models. We also identified the spinal neurons expressing MOR, DOR, and both receptors. In the dorsal horn, DOR and MOR are present in separate neural populations, with only limited co-expression in subclasses of excitatory lamina II interneurons and NK1R+ lamina I projection neurons. Similarly, DOR and MOR are predominantly segregated in brain pain affect circuits. Conditional knockout experiments revealed that DOR agonists act on DORs expressed by somatostatin+ interneurons to reduce mechanical pain, but can also non-selectively activate MOR to diminish heat pain. Unexpectedly, we found that the majority of spinal neurons co-expressing DOR and MOR correspond to ventral horn V1 inhibitory interneurons that regulate the activity of motoneurons.
Conclusions: Our data on MOR pronociceptive function in nociceptors support combining opioid agonists with peripheral MOR antagonists to limit analgesic tolerance and OIH. These results also establish the divergent function of DOR and MOR in pain circuits, and identify key neuronal populations for dissecting DOR-MOR functional interactions in vivo and their contribution to opioid analgesia.
Disclosure: Nothing to Disclose.

43.4 Pain-Induced Negative Affect is Mediated via Recruitment of the Nucleus Accumbens Dynorphin-Kappa Opioid System

Jose Moron-Concepcion

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Background: The mesolimbic pathway undergoes long term changes in the presence of inflammatory pain. The kappa opioid receptor (KOR) is expressed on dopaminergic terminals in the nucleus accumbens (NAc) where it locally controls dopamine release. The KOR system can also be altered by pain and involved in pain-induced alterations in motivation for reinforcers. Indeed, KOR stimulation in the so-called NAc Shell "cold spot" decreases the "liking" of sucrose tasting. Furthermore, activation of the KOR system by dynorphin, its endogenous agonist, mediates aversive behaviors. In this work, we focused on pain-induced neurobiological modifications in the KOR system in the NAc shell, and the consequences of these alterations on both reinforcing and aversive behaviors in the presence of pain.
Methods: We used an inflammatory pain model (CFA in the hind paw of rats) to assess pain-induced changes in KOR expression and function with GTPgammaS binding and western blot analysis. We also monitored the excitability of dynorphin-containing neurons using ex vivo patch clamp recording. In addition, we conducted in vivo experiments to further dissect the involvement of KOR system in conditions of pain. We determined whether confirmed endogenous dynorphin was necessary for decreased motivation for sucrose by using an chemogenetic silencing approach. An HSV-dynorphin-Gi-DREADD construct was used to silence dynorphinergic neurons in the NAc. We then determined whether CFA treatment was sufficient to potentiate dynorphinergic neuron-mediated place aversion. We expressed a cre-dependent channelrhodopsin (ChR2) virus in dynorphin-IRES-cre mice to induce dynorphin release in the real-time place aversion assay. All experimental protocols in animal studies were approved by the Institutional Animal Care and Use Committee at Columbia University and Washington University in St. Louis.

Results: Our western blot and GTPgammaS results show that 48 hours after the induction of pain, both the expression and function of KORs in the NAc are enhanced. In addition to the increase in receptor function, whole-cell recordings from dynorphin-cre mice confirmed that pain also potentiates the excitability of dynorphin-containing neurons in the NAc. In order to examine whether this upregulation of the dynorphin-KOR system underlies the pain-induced changes in motivation, we microinjected the long-acting KOR antagonist NorBNI into the NAc. We were able to reverse the previously observed pain-induced decrease in motivation by blocking KOR, highlighting the critical role of this receptor in this pain effect in motivation. In a complementary study, we found that activation of KOR in the NAc by U50,488 is sufficient to decrease motivation for sucrose in a way that mimics an inflammatory pain state. We followed up on these findings by silencing dynorphinergic neurons, using a HSV-dynorphin-Gi-DREADD, and this experiment revealed the necessity of endogenous dynorphin release to mediate the deleterious effects of pain on motivation. Finally, we found that photo-stimulation of dynorphinergic neurons in the ventral NAc shell induces a real time place aversion that is potentiated in the presence of pain (CFA exposure). The activation of these neurons in both control and painful conditions did not alter locomotor activity, which suggests that our effects are specific to motivation, and not a generalized inhibition of locomotion.
Conclusions: Here we show that accumbal dynorphin-KOR system is recruited in the presence of pain. Furthermore, we report that pain decreases motivation for rewards and potentiates dynorphin-KOR mediated aversion via activity in the ventral NAc shell. Characterization of pain-induced changes in neural circuits that regulate motivation and reward is critical for the development of strategies to overcome opioid misuse, affective disorders, and their comorbidity with pain.
Disclosure: Nothing to Disclose.

Panel
44. Estrogen and Schizophrenia: What Happens in the Brain?

44.1 The Effect of Estrogen in Treatment Resistant Schizophrenia: Results From a Randomized Controlled Trial

Mark Weiser

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Background: Previous studies showed that estrogen has a positive effect on symptoms in schizophrenia, and a recent study published by Kulkarni in 2015 found that transdermal estradiol is effective as an adjunctive treatment for female, pre-menopausal, treatment-resistant schizophrenia patients. This study aims to replicate these findings in a large randomized controlled trial.
Methods: This was an 8-week, double-blind, randomized, placebo-controlled study on 200 pre-menopausal women meeting DSM-V criteria for schizophrenia or schizoaffective disorder, who were resistant to antipsychotic treatment. Patients were recruited from December 2015 to July 2016 in Moldova, and were randomized to receive either 200 μg estradiol patches or placebo in addition to antipsychotic treatment.
Results: At the end of this 8 week trial, patients whose antipsychotic medication was augmented with estradiol patch improved more than placebo, with 68% of participants receiving estrogen patch having a 20% or more reduction in total PANSS in contrast to 48% to placebo (chi squared = 7.9, df = 1,$p = 0.005$). A median split by age revealed that the estrogen patch only produced improvement in the women over 38 years or older, in whom 76% responded to the patch, versus 38% to placebo (chi squared = 14.7, df = 1, $p = 0.00001$). There was essentially no difference in response in participants 38 years old or younger, estrogen patch 58% vs placebo 56%). ANOVAs showed that the estradiol group showed statistically significant improvement in PANSS total score (difference in PANSS total points = 4.08, 95% CI 1.36-6.79, $p = 0.003$, effect size = 0.4), and in the PANSS subscales for positive symptoms (difference in PANSS points = 0.93, 95% CI 0.20-1.67, $p = 0.013$), negative symptoms (difference in PANSS points = 1.01, 95% CI 0.26-1.76, $p = 0.008$), and general psychopathology (difference in PANSS points = 2.22, 95% CI 0.67-3.78, $p = 0.005$) compared to the placebo group. This effect was not driven by large improvements on a few symptoms, rather, estrogen was associated with improvements on a relatively large number of symptoms, the differences being particularly pronounced in delusions (difference = -0.44, 95% CI -0.72, -0.16) and suspiciousness (difference = -0.49, 95% CI -0.79, -0.19). There was a significant difference between groups in the CGI Severity scale (difference = 0.17, 95% CI 0.00-0.34, $p = 0.049$, effect size = 0.28), and in the total MADRS score (difference = 1.48, 95% CI 0.59-2.37, $p = 0.001$, effect size = 0.57). There was no significant difference in the composite BACS score ($p > 0.1$). Side effects were minimal and included breast tenderness and/or weight gain in 14% of the patients receiving estrogen.
Conclusions: This large RCT, performed in a different country by different investigators replicates the finding that transdermal estradiol is an effective adjunctive treatment for female, pre-menopausal, treatment-resistant schizophrenia patients. Further studies with estrogen in schizophrenia are warranted, and short-term add-on treatment with estradiol patches might be considered in these patients.
Disclosure: Nothing to Disclose.

44.2 Aromatase (Estrogen Synthase) in the Human Brain: Where and What for?

Anat Biegon

Stony Brook University School of Medicine, Stony Brook, New York, United States

Background: Aromatase, the last and obligatory enzyme catalyzing estrogen biosynthesis
from androgenic precursors, can be visualized and measured in the living human brain using a radiolabeled aromatase inhibitor ([11C]vorozole) and positron emission tomography (PET).
Methods: Forty-eight healthy men and women (age range, 23–67 y; BMI 18-38) were given an IV injection of

the radio-tracer and their brain or body scanned for 90 minutes using PET. Young women were scanned at two distinct phases of the menstrual cycle. A subgroup of subjects also filled a personality questionnaire (MPQ) and were tested for verbal learning and memory and non-verbal reasoning.
Results: We found that the human brain is a major site of aromatase expression, whereby several brain regions, including the thalamus, amygdala, and preoptic area; contained higher levels of aromatase than any other organ besides the ovulating ovary (i.e. high levels found unilaterally in young women around midcycle). Aromatase levels in the male brain were higher than levels in female brain, which did not change significantly across the menstrual cycle. Cigarette smoking and aging reduced 11C-vorozole uptake in both sexes. Aromatase expression in the brain was inversely correlated with body mass index; such that overweight individuals had lower levels relative to normal-weight individuals and the lowest levels were found in obese individuals. This finding could be explained at least in part by a significant association we found between aromatase in amygdala and trait constraint (inhibitory control and harm avoidance) in both men and women. Finally, brain aromatase availability predicted individual differences in verbal and non-verbal cognitive performance in men but not in women. Men with lower amygdala levels of aromatase had better recall for a word list and performed better on non-verbal spatial reasoning test.
Conclusions: These findings support an important role for extragonadal estrogen production in the control of non-reproductive brain function in men and women.
Disclosure: Nothing to Disclose.

44.3 Sex-Dependent Mechanisms of Acute Estrogen Signaling in the Brain

Catherine Woolley

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Background: In addition to their roles as hormones, estrogens are also synthesized in the brain, of both sexes, as neurosteroids that can modulate neurophysiology on a time scale of minutes. We are investigating neurosteroid estrogen synthesis and actions in the hippocampus, a brain region important in cognition and affect.
Methods: We have used a combination of in vitro whole-cell voltage clamp recording in hippocampal slices from gonadectomized adult male and female rats and behavioral analyses following acute infusion of an estrogen receptor β (ERβ) agonist into the hippocampus of gonadectomized adult male and female rats.
Results: We find that the key estrogen, 17β-estradiol (E2), acutely potentiates a subset of excitatory synapses in the hippocampus of both sexes, but that this occurs through a distinct combination of estrogen receptors (ERs) and downstream signaling in each sex. For example, agonists selective for ERβ potentiate excitatory synapses in females by increasing presynaptic glutamate release probability, whereas in males, ERβ agonists potentiate postsynaptic sensitivity to glutamate. Furthermore, E2-induced synaptic potentiation requires cAMP-regulated protein kinase (PKA) and Ca++ release from internal stores in females, but not in males. Parallel behavioral testing shows that ERβ agonists delivered to the hippocampus in vivo are acutely anxiolytic in females but anxiogenic in males.
Conclusions: Our results show that although the hippocampus of both male and female rats is capable of synthesizing neurosteroid estrogens, the downstream signaling that mediates acute estrogen actions in the hippocampus differs between the sexes and can result in divergent functional consequences. Thinking more broadly, beyond estrogens, these studies reveal that molecular mechanisms of synaptic modulation can differ between males and females, which highlights the value of considering sex in the development of therapeutics that target neuromodulatory systems.
Disclosure: Nothing to Disclose.

44.4 Estradiol Mediates a Sensitive Period in Cerebellum Development

Margaret McCarthy

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

Background: Traditional views of estrogen action in the developing brain focus on sexual differentiation and the organization of enduring sex differences in reproductive physiology and behavior. However, we have found that endogenous estradiol synthesis within the developing cerebellum both defines and determines the outcome of a sensitive period in Purkinje cell maturation. The cerebellum is one of the earliest brain regions to develop and last to mature, making it particularly vulnerable to developmental perturbation. Pathologies of the cerebellum are strongly associated with neuropsychiatric disorders with origins in development.
Methods: Postnatal rat pups were injected into the cerebellum with either the prostaglandin PGE2, the inflammatory agent LPS or the COX-inhibitor, indomethacin for 2 days during either the 1st, 2nd or 3rd week of life. The cerebellar vermis was collected at varying times after injection depending upon the endpoints measured with included estradiol and endogenous PGE2 production and Purkinje neuronal morphology. A separate group of animals received the same treatment paradigm and were raised until the juvenile period at which time they were assessed for social play, object recognition and perseveration and open field behavior. Lastly, a detailed developmental time course of gene expression of the components of a signaling pathway that begins with prostaglandin production and ends with estradiol synthesis locally within the cerebellum was conducted by qPCR from birth to the end of the 3rd week of life.
Results: Injection of PGE2 into the cerebellum was found to stimulate aromatase activity, the estradiol synthesizing enzyme, and estradiol content was found to increase, but only if the treatment was given during the 2nd postnatal week, and not the 1st or the 3rd. Similarly, treatment with LPS, either peripherally or directly into the cerebellum increased PGE2 and estradiol synthesis, but only if administered during the 2nd week. Indomethacin reduced PGE2 production below baseline at all developmental time

points and in response to LPS treatment during the 2nd postnatal week. This and other observations led to the conclusion that PGE2 stimulates aromatase to regulate endogenous estradiol synthesis within the cerebellum and that this increases further if inflammation occurs during the 2nd postnatal week. Elevated estradiol resulted in a stunting of Purkinje neuron dendritic trees, whereas reduced estradiol had the opposite effect, Purkinje dendrites that were longer and more branched than controls. Behavioral assessment revealed enduring consequences in that social play was reduced and animals tended to perseverate on objects if Purkinje cell development was impaired. However, these behavioral effects were only observed in males. Analyses of gene expression profiles revealed that the 2nd week is characterized by a peak in aromatase and estrogen receptor-alpha gene expression and this confers the unique sensitivity of this time period.
Conclusions: Sensitive periods in development are essential for understanding when and how external insults can derail normal developmental processes. We have identified a heretofore unknown sensitive period of cerebellar development in the rat during the 2nd postnatal week, a time period roughly analogous to full-term birth in the human. Peripartum inflammation and/or treatment with NSAIDS or other COX inhibitors may be contributing factors to risk for later development of neuropsychiatric disorders.
Disclosure: Nothing to Disclose.

Mini Panel
45. Transgenic and Targeted Gene Editing Research Using Non-Human Primates: Progress, Prospects and Ethical Issues (Sponsored by ACNP Ethics Committee)

45.1 Primate Genetic Manipulation in the United States: Present Aspects and Future Prospects

Karen Bales

University of California, Davis, California, United States

Background: Non-human primate (NHP) models have long provided a crucial translational step in biomedical research. Laboratories in the United States have made significant progress in genetic manipulation of non-human primates, primarily common marmosets (Callithrix jacchus), rhesus macaques (Macaca mulatta), cynomolgous macaques (Macaca fascicularis), and vervet monkeys (Chlorocebus sabaeus). Manipulations include both gene editing with new tools such as CRISPR-cas9, as well as introduction of human stem cells into non-human primates. In this talk, we summarize the current state of the art in NHP genetic manipulation in the US.
Methods: We conducted a national survey of the current use of gene editing and human stem cell introduction in NHPs. This included contacting scientists at National Primate Research Centers as well as universities known to be working with these techniques. We summarize the genes of interest, disorders being studied, and advantages and disadvantages of different NHP species. We reexamine ethical issues involved in transgenic NHP research.
Results: NHP models of Parkinson's disease, Alzheimer's, Huntington's, and other neurological disorders are utilizing genetic approaches. These trials are being conducted in both public and private facilities, including several national primate centers, and in several different primate species, each of which has specific advantages and disadvantages. The unique aspects of neurobiology that make primates the best models for humans also increase our ethical responsibilities to their appropriate care. The applicability of the American Animal Welfare Act and other regulations, in particular, the Guide for the Care and Use of Laboratory Animals, to transgenic NPH research is reviewed.
Conclusions: Genetic manipulation of NHP models promises future advances and is making current strides in the United States. A detailed understanding of each species that may be used as preclinical models will advance our understanding of human neurological and neuropsychiatric diseases.
Disclosure: Nothing to Disclose.

45.2 Genetically Modified Non-Human Primates: Applications in Neurological and Psychiatric Disorders

Erika Sasaki

Central Institute for Experimental Animals, Kawasaki, Japan

Background: Nonhuman primates (NHPs) offer excellent models for preclinical research to develop novel therapeutic approaches because of their genetic, metabolic and physiological similarities to humans. Recent progresses in genetic modification (GM) technologies, including genome editing, have enabled the development of GM human disease NHP models. The common marmoset (Callithrix jacchus) is a suitable NHP experimental animal for producing GM models because of their unique reproductive characteristics such as multiple ovulation, short sexual maturity period and relatively short gestation period. Using this marmoset model, GM human disease models have been developed in Japan following strict ethical animal care guidelines.
Methods: For production of GM human disease model marmosets, currently, transgenic models and gene-targeted knock out models are available in the marmoset. For the transgenic models, an objective exogenous gene is introduced into the marmoset zygotes using lentiviral vectors. For the gene-targeted knock out (KO) models, genome editing tools, the artificial nucleases, ZFNs and TALENs are used. To avoid mosaicism of a modified gene in the organism, screening of the artificial nucleases are performed prior to generation of KO marmosets.
Results: Several lines of transgenic marmosets have been produced by lentiviral vectors and these transgenic marmosets show objective phenotypes. This lentiviral vector based transgenic approach is a stable method for not only production of transgenic marmosets but also obtaining the next generation offspring. Use of selected highly efficient ZFNs or TALENs pairs has resulted in reduction of mosaicism of the targeted gene KO in the marmoset and enabled it to exhibit altered phenotypes. Specifically, the IL2RG gene KO marmoset shows immunodeficient phenotypes.
Conclusions: GM marmoset models are becoming available for understanding human brain and neuronal disorders.

However, to fully utilize GM marmoset models in biomedical science, including neuroscience, there are further methodological challenges. Especially important is the establishment of highly efficient homologous-gene knock in models and new technics to accelerate reproduction to obtain the next generation.
Disclosure: Nothing to Disclose.

45.3 Bioethical Considerations in Genetic Modification of Nonhuman Primates

Jeffrey Kahn

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Background: The development and structure of the central nervous systems in non-human primates (NHP's) makes them more useful than other mammals in modeling human brain diseases which we seek to understand and find therapeutic interventions. Yet ethical issues arise in their use. The director of the UC-Davis MIND institute was quoted: "If you develop an animal model of something like autism, where repetitive behavior and self-injurious behaviors are actually part of the diagnostic features, how do we deal with animal-welfare constraints on maintaining those animals?".
Methods: Review and conceptual analysis of recent policy statements and the bioethics literature regarding research with nonhuman primates with an emphasis on the moral standing of NHPs and their cognitive and emotional capacities.
Results: The cognitive and emotional attributes that render NHPs vulnerable to harm led to the closure of some primate centers and the dismantling of the NIH programs that use chimpanzees. However, research is permissible when alternatives such as in vitro testing or computer modeling will not suffice, when the animals will be maintained in as "humane" a setting as possible, and when the benefits to science and human health are sufficient to outweigh the distress caused to the animal subjects. Still these factors have become the subject of debate. While we permit research on non-consenting human subjects, such as children and the profoundly intellectual disabled, it is done with the expectation that the work will rebound to the benefit of the subjects or, at least, to members of their own species and, by extension, their own moral community. When research using NHPs is aimed not at veterinary illness but at human illness, this reciprocity may be lost.
Conclusions: The value of research using NHPs is clear. Their use increasingly demands we answer the question: is there a defensible basis for preferencing our own species? If there is, what are limits on the range of defensible experiments that can be done, particularly in the area of neurological and psychiatric diseases? And if those experiments include introduction of traits typical of our own species, at what point have we created a new kind of animal, one that has a nearly human interior life but is still regarded as sub-human and unworthy of membership in our own moral community.
Disclosure: Nothing to Disclose.

Mini Panel
46. Genetic, Epigenetic and Neural Correlates of Allostatic Load in Neuropsychiatry

46.1 Role of the Epigenetic Agent Acetyl-L-Carnitine as Gating Biomarker in Depression and Influences of Early Life Experiences: From Animals to Humans and Back to Animals

Carla Nasca

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Background: Converging evidence has demonstrated that epigenetic drugs, including histone deacetylase inhibitors and the acetylating agent acetyl-L-carnitine (LAC), promote rapid antidepressant responses. In particular, findings from our group and others have shown that LAC rapid actions were seen after 3 days of administration in animals with an endogenous reduction in LAC in plasma and mood regulatory brain regions, whereby responses to standard antidepressant medications required repeated weeks of administration. LAC effects occur via a mechanism of histone acetylation to regulate glutamatergic functions, trophic environment and brain structural plasticity. Taken together with our recent reported preclinical association of reduced LAC with insulin resistance (IR), the ability of the epigenetic agent LAC to impact several biological systems suggests the importance of investigating LAC as gating biomarker for depressive disorder and the origins of its aberrant levels.
Methods: 71 patients with major depressive disorder (MDD) and 45 age and sex matched healthy controls (HC) 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), the two psychiatric scales, HDRS-17 and MADRS. All patients with MDD were in an acute episode during study participation. 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. Plasma LAC and free-carnitine concentrations were determined using ultra-performance liquid chromatography-electrospray-tandem mass spectrometry (UPLC-MS/MS).
Results: Supported by validation and replication, human data will be presented showing that plasma LAC (and not free-carnitine) was significantly reduced in patients in acute episode of MDD compared to age and sex matched 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 ($p = 0.04$, $r = 0.35$) and earlier age of onset of MDD ($p = 0.04$, $r = 0.32$), independently of psychotropic drug treatment. Consistent with lower LAC levels in a more severe clinical phenotype of MDD (i.e., greater severity, earlier age of onset), we report that patients with treatment resistant depression (TRD) showed significant reductions in LAC and were more likely to have been abused in childhood than HC. This is suggestive of an allostatic overload over the life course that contributes to LAC concentrations in patients with TRD. Moreover, in

patients with TRD, history of emotional neglect moderated LAC levels ($p = 0.04$, $r = 0.66$). Preliminary data from reverse translational studies will be presented showing the contribution of maternal care to adulthood LAC levels with associated IR and trajectory of depressive-like disorder.
Conclusions: These new findings suggest a role for LAC, an epigenetic modulator of glutamatergic function, as biomarker to define a severe clinical phenotype of depression, providing a potential biological target for a precision medicine approach. Our findings also support the importance of considering the developmental origins of severe types of depression, including TRD. Future studies will explore our recent preclinical finding linking reduced LAC to IR in MDD. Thus, LAC may constitute a useful biomarker not only as gating factor part of common neurobiological platform of depression, but also as a tool for investigating the developmental trajectory of depression.
Disclosure: Nothing to Disclose.

46.2 Central Insulin Resistance and Synaptic Plasticity Deficits: Towards an Integral Model of Hypothalamic and Hippocampal Effects of Insulin

Lawrence Reagan

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Background: In addition to peripheral complications, it is becoming increasingly clear that the complications of type 2 diabetes (T2DM) and insulin resistance (IR) extend to the central nervous system (CNS) and include deficits in cognitive function. A major obstacle in the identification of the mechanistic basis for neuroplasticity deficits in T2DM and IR is that metabolic disorders are characterized by a variety of endocrine and metabolic changes, all of which may act independently, as well as in additive or synergistic ways, to promote cognitive dysfunction.
Methods: In an attempt to disentangle the relative contributions of peripheral IR versus CNS IR in the development of neuroplasticity deficits, we developed and characterized a lentiviral vector packaged with an antisense sequence selective for the insulin receptor (IRAS). Following administration of this viral vector to either the hypothalamus or hippocampus, measures of hippocampal synaptic plasticity were evaluated.
Results: Hypothalamic-specific IR elicited the expected metabolic and endocrine changes including increases in body weight and body adiposity, IR, leptin resistance and increases in pro-inflammatory cytokines. Additionally, rats with a hypothalamic-specific IR exhibited structural and functional deficits in hippocampal neuroplasticity. These results indicate that hypothalamic IR appears to be a keystone mechanism through which peripheral endocrine changes lead to deficits in the structural and functional integrity of brain regions like the hippocampus. However, these results cannot differentiate between the peripheral versus CNS changes that are responsible for these deficits in hippocampal synaptic plasticity. To address this question, rats received bilateral injections of the IRAS construct to induce a hippocampal-specific IR in order to more selectively examine the potential role of hippocampal IR in the neuroplasticity deficits associated with metabolic disorders and aging. Compared to control rats, rats with hippocampal-specific IR did not exhibit changes in metabolic or endocrine parameters such as body weight, body adiposity or glucose tolerance. However, hippocampal-specific IR was associated with deficits in spatial learning, as well as impairments in stimulus-evoked long-term potentiation that likely resulted from alterations in the expression and phosphorylation state of glutamate receptor subunits.
Conclusions: These data support the concept that CNS insulin activity acts independently of peripheral insulin to facilitate hippocampal synaptic plasticity. Additionally, these findings illustrate that hippocampal IR, alone and in combination with peripheral IR, are responsible for the neurological complications observed in patients with metabolic disorders, including impairments in cognitive function.
Disclosure: Nothing to Disclose.

46.3 Brain Correlates of Allostatic Load in Population-Based and Clinical Samples

Sophia Frangou

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Background: Large genomics studies and consortia have identified genetic factors that increase the risk for metabolic dysregulation and cellular integrity as well as the risk for adverse mental health outcomes. The biological pathways leading from risk to outcomes are yet to be fully elucidated but are likely to involve alterations in brain structure and function. Here we present results from three lines of research investigating this link.
Methods: We used three different approaches. First, we used mendelian randomisation to test the association between brain structure and a composite measure of genetic load for insulin resistance based on the findings by Lotta et al 2017 in the ENIGMA cohort of 30717 individuals. Second, we estimated the population covariation-between brain imaging metrics relating to working memory and proxy indices of metabolic and glucose control in 823 healthy individuals from the Human Connectome Project (HCP). Third, we tested the association between telomere length and hippocampal volume as an exemplar of the link between a marker of allostatic load and mood disorders.
Results: In the ENIGMA cohort, increased genetic load for insulin resistance was associated with smaller intracranial volume (inverse variance weight p-value = 0.002). In the HCP cohort, increased body mass index and increased HB1Ac were associated with reduced activation and connectivity within the working memory, even after accounting for multiple other behavioral and lifestyle risk factors. Finally, telomere length was negatively associated with hippocampal volume bilaterally but this effect was mitigated by long-term lithium treatment.
Conclusions: Multiple lines of evidence suggest the measures relating to poor glucose and metabolic control and greater allostatic load impact directly brain structure and function even in healthy individuals and may increase the vulnerability for a range of adverse mental health outcomes.
Disclosure: Nothing to Disclose.

Panel
47. Hippocampal Dysfunction in the Pathophysiology of Schizophrenia

47.1 Using MEG to Probe Hippocampal-Cortical Networks Across Illness-Stages in Schizophrenia

Peter Uhlhaas

University of Glasgow, Glasgow, United Kingdom

Background: A considerable body of work over the last 10 years combining non-invasive electrophysiology (electroencephalography/magnetoencephalography; EEG/MEG) in patient populations with preclinical research has contributed to the conceptualization of schizophrenia as a disorder associated with aberrant neural dynamics and disturbances in excitation/inhibition (E/I) balance parameters. Specifically, I will propose that recent technological and analytic advances in MEG-techniques provide novel opportunities to address the role of neural oscillations in hippocampal and cortical networks and to establish important links with translational research.
Methods: We recruited participants meeting clinical high-risk criteria ($n = 90$), 19 antipsychotic-naïve, first episode patients (FEP) and 21 chronic schizophrenia patients as well as healthy control participants ($n = 49$). Participants underwent measurements with MEG and Magnetic Resonance Imaging. Magnetoencephalographic resting-state and task-related activity was examined in the 3-90 Hz frequency range in combination with source reconstruction. In addition, we tested for network interactions between 80 nodes of the AAL-atlas with a coherence measure. MEG-data from clinical groups was compared to the effects of Ketamine on hippocampal-cortical oscillations in a group of 15 healthy volunteers to test similarities with the pattern of oscillatory activity across illness-stages in schizophrenia.
Results: Clinical high-risk and FEP participants were characterized by increased resting-state, broad-band gamma-power and functional connectivity compared to chronic schizophrenia-patients in hippocampal-cortical networks, which were characterized by reduced 30-90 Hz activity. Differences emerged also between FEP and clinical high-risk groups with elevated connectivity in the first-episode patients involving subcortical-cortical interactions. Moreover, impairments in task-related gamma-band oscillations were observed in all clinical groups. In healthy controls, Ketamine led to an upregulation of both resting-state and task-related high-frequency oscillations in hippocampus and cortical networks that overlapped with the effects observed in clinical high-risk and FEP-groups.
Conclusions: The current study suggests that early psychosis involves specific changes of hippocampal-cortical oscillations that resemble effects induced by NMDA-antagonists. Accordingly, these data support the view that aberrant glutamatergic neurotransmission at illness-onset could be a driver of both cognitive deficits and psychopathology.
Disclosure: Part 1: Lundbeck, Grant, **Part 2:** Lilly, Grant.

47.2 A Temporal Model of Hippocampal Pathophysiology in Schizophrenia

Scott Small

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Background: Studies have three suggested three hippocampal abnormalities in schizophrenia—atrophy, hypermetabolism, and glutamate elevations. Based in a previous published mouse model of disease, we hypothesized that elevations in hippocampal glutamate is the driver of the two other defects, first leading to hypermetabolism and then leading to atrophy.
Methods: To test this hypothesis we recently completed a longitudinal study in which we imaged 76 patients who fulfilled criteria for 'clinical high risk' and 20 age-matched controls. We use MRS to assess glutamate, CBV-fMRI to assess metabolism, and structural MRI to assess volume.
Results: Confirming our hypothesis, our results suggest that prodromal psychosis is characterized primarily by elevation in hippocampal glutamate and secondary by hippocampal hypermetabolism. The results suggest that loss of volume occurs last. We observed that men were more likely to progress than woman, and that measures of hippocampal volume was a strong predictor of progression to psychosis.
Conclusions: Together with other results, we believe it is possible to propose a temporal model of pathophysiological progression in the hippocampus, from a prodromal to psychosis stage. This model has clear therapeutic implications, suggesting that glutamate reducing agents might best work in the earliest pathophysiological stage of the illness.
Disclosure: Part 1: Denali, Advisory Board, Janssen, Advisory Board, **Part 2:** Janssen, Advisory Board.

47.3 Hippocampal and Relational Memory Function in Schizophrenia

Stephan Heckers

Vanderbilt University, Nashville, Tennessee, United States

Background: It is well known that the hippocampus is abnormal in schizophrenia. But there is considerable debate about three questions: Is hippocampal pathology in schizophrenia 1) localized or diffuse, 2) related to abnormalities of memory, 3) present before, at the onset, or in later stages of the illness? Several neuroscientific methods (neuropathology, neuroimaging, cognitive neuroscience) and several study designs (cross-sectional and longitudinal, observational and experimental) have been employed to answer these questions. However, in contrast to other hippocampal models of disease (e.g., dementia and epilepsy), schizophrenia has not been adequately explained by hippocampal pathology. Here I will review emerging data from our laboratory and articulate crucial experiments that are needed to advance a hippocampal model of schizophrenia.
Methods: We used 3T and 7T MR to map hippocampal structure and function in schizophrenia in first episode and chronic patients with schizophrenia. We tested the hypothesis that anterior, but not posterior, changes of hippocampal blood volume, blood flow and volume are present in the early stages of schizophrenia and can explain memory deficits.
Results: Changes of hippocampal volume and activity preferentially affected the anterior hippocampus. Abnormalities of relational memory were present already in a subgroup of patients during the early stage of schizophrenia and were related to abnormal hippocampal-cortical connectivity. We will present the initial analysis of a 2-year

longitudinal study of first episode schizophrenia to test the hypothesis of progressive changes of hippocampus and relational memory.
Conclusions: The anterior hippocampus is more affected than the posterior hippocampus in schizophrenia. Abnormal hippocampal-cortical connectivity can explain relational memory impairment in schizophrenia. Findings are more heterogeneous in the early stages of schizophrenia. Longitudinal and interventional studies of schizophrenia patients in the early stage of their illness provide several opportunities to test predictions of a hippocampal model of schizophrenia.
Disclosure: Nothing to Disclose.

47.4 Hippocampal Glutamate and Functional Connectivity as Biomarkers of Treatment Response to Antipsychotic Medication

Adrienne Lahti

University of Alabama at Birmingham, Birmingham, Alabama, United States

Background: We previously reported elevated hippocampal regional cerebral blood flow (rCBF) in unmedicated patients with schizophrenia (SZ) and rCBF reduction associated with good treatment response to antipsychotic drug (APD). To characterize the pathophysiology of this pattern, we conducted a prospective study where hippocampal neurochemistry and function were measured before and after APD treatment.
Methods: Unmedicated SZ were scanned before and after 6 weeks of treatment with risperidone and categorized as good (TR) ($n=17$) or poor (TPR) ($n=17$) responders based on a median split of treatment response [percent change on the Brief Psychiatric Rating Scale psychosis score]. Matched healthy controls (HC) ($n=23$) were scanned twice, 6 weeks apart. Resting state fMRI and magnetic resonance imaging (MRS) (PRESS sequence) of the left (L) hippocampus were obtained at both time points. fMRI preprocessing and analyses were conducted using the CONN toolbox in SPM8. The first eigenvariate of the blood oxygen level-dependent time series from a L hippocampal seed region was extracted and correlated with the time series from each of fourteen individual network masks. MRS data were quantified in the time domain using jMRui. N-acetyl-aspartate, choline, and glutamate+glutamine (Glx) were quantified. Generalized linear mixed effects models were employed to estimate the association between hippocampal metabolites, hippocampal connectivity to each of the fourteen networks, hippocampal connectivity to networks covaried by Glx levels, and Group and Time.
Results: For hippocampal metabolites, there was a significant group by time interaction, but only for Glx levels. Post hoc contrasts reveal that, at baseline, Glx levels were significantly higher in TR compared to HC ($p<0.01$). After treatment, in TR, but not in TPR, there was a significant decrease in Glx levels ($p<0.02$). For hippocampal functional connectivity, there was a significant group effect, but not time or group by time interaction; post hoc contrasts reveal there was a significant difference between HC and TPR in the functional connectivity to the executive control network. For hippocampal functional connectivity modulated by Glx levels, there was a significant effect of group, but not time or group by time interaction; this effect was driven by a significant difference between both the HC and TR and the TPR in the hippocampal functional connectivity modulated by Glx levels to both the salience and basal ganglia networks.
Conclusions: These results suggest both state and trait hippocampal glutamatergic dysfunctions that are related to treatment response. In TR, there is an elevation of glutamate levels at baseline that disappears with treatment, and in TPR, there is an alteration of the relationship between hippocampal functional connectivity to salience and basal ganglia networks and Glx; this alteration is present across time.
Disclosure: Part 1: Janssen, Grant, **Part 2:** Janssen, Grant.

Panel
48. Sex Steroids and Adolescent Brain Development: A Translational Approach to Understanding Vulnerabilities to Psychopathology in Adolescence

48.1 Pubertal Maturation: Intrinsic Functional Connectivity Increased in Boys, but Decreased in Girls, a Risk for Depression?

Tiffany Lago

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Background: Adolescence, the transition from childhood to adulthood, is associated with a marked increase in psychiatric disorders, together with a sex-related imbalance in psychopathology. Girls exhibit a preponderance for internalizing disorders (e.g. mood and anxiety), while boys are more likely to be affected by externalizing disorders (e.g. attention-deficit hyperactivity and conduct). The neurological underpinnings of this variance is unknown. Presumably, ontogenic changes in adolescence can depend on solely age, puberty, or a combination of both. The present study examines the effects of sex and puberty on the intrinsic functional connectivity (iFC) of structures implicated in subthreshold mood disorders in adolescence: the left medial prefrontal cortex (lmPFC), pregenual anterior cingulate cortex (lpgACC), and posterior cingulate cortex (lPCC). Hypotheses were three-fold: (1) puberty affects iFC of these structures, (2) pubertal effects would differ between boys and girls, (3) mood symptomotology would be associated with iFC, regardless of sex.
Methods: Resting state fMRI scans from a subset ($n=304$; male 51%; age: 14.46 (0.43); Puberty Development Scale: 2.86 (0.57)) of the large longitudinal cohort of the European IMAGEN consortium were analyzed. Data processing and analyses were conducted with Analysis of Functional NeuroImages (AFNI). Multiple regression analyses were used to examine the effects and interactions of puberty and sex on lmPFC-, lpgACC- and lPCC-iFC. Individual variability in internalizing symptoms were indexed using the Development and Well-Being Assessment (DAWBA) at time of scan, and 2-year follow-up.
Results: Higher puberty level was associated with stronger lmPFC- and lPCC-iFC in boys, and weaker iFC in girls (sex X puberty interaction). Specific clusters of this interaction

include lmPFC-iFC: dorsomedial prefrontal cortex (dmPFC), posterior middle temporal gyrus (pMTG), and dorsolateral prefrontal cortex (dlPFC); lPCC-iFC: inferior parietal lobule, precentral gyrus, mPFC, and pMTG. Sex did not interact with pubertal effects on lpgACC-iFC. A sex main effect on lPCC and lpgACC revealed stronger iFC in boys than girls. The main effect of puberty on iFC was not significant. Behaviorally, exploratory analyses indicated negative associations of lpgACC-iFC and lmPFC-iFC with internalizing symptoms at 2-year follow-up. In other words, the weaker the iFC of lpgACC and lmPFC was at age 14 years, the more likely participants were to have mood or anxiety (internalizing) symptoms at age 16 years.
Conclusions: This study revealed that puberty influences resting state connectivity of regions that have been identified as conferring risk for mood problems in adolescents. As hypothesized, puberty affected lmPFC and lPCC iFC differently in boys and girls. Additionally, weaker iFC of the lpgACC and lmPFC at age 14 predicted internalizing symptoms at age 16. Results failed to support an interaction of sex and puberty on lPCC iFC, or a main effect of puberty. Importantly, main sex effects and sex interactions found in these results could underlie the sexually-dimorphic development of mood disorders during adolescence.
Disclosure: Nothing to Disclose.

48.2 Sex Steroid Influences on Striatal Activation to Reward Cues in Adolescence

Cecile Ladouceur

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Background: Affective neuroscience research suggests that changes in the reward circuitry during adolescence could contribute to increased risk for psychiatric disorders such as depression and substance abuse. Although emerging evidence from animal and human neuroimaging research has begun to document the specific effects of puberty and sex steroids on reward circuitry, few studies have employed a methodological design that can disentangle the effects of puberty and age. Here, we used such a design with a novel fMRI reward cue processing task to examine how neural activation to reward cues was associated with pubertal maturation, including basal levels of sex steroids. We hypothesized that striatal activation to reward cues would be positively pubertal status and levels of sex steroids.
Methods: Sixty-four adolescents (39 girls; mean age = 11.63; SD = 1.01) (girls, aged 10-12; 21 boys, aged 11-13) varying in pubertal status underwent a fMRI scan while performing a reward cue processing task. This task consisted of 60 pseudo-randomized trials (30 reward cue, 30 no reward cue trials). Participants had to respond quickly based on to the spatial location of a gopher with (Reward cue) or without (No Reward cue) a bag of money. Participants earned $0.10 at 75% chance for correctly responding to reward cue. Statistical analyses focused on correct response trials. Pubertal maturation was assessed using Tanner staging and the Pubertal Development Scale (PDS) (Petersen et al. 1988), a self-report measure containing questions about physical development. We implemented a scoring algorithm that optimally parallels Tanner staging, and provides a more sensitive way to capture adrenal and gonadal hormonal signals of physical development (Shirtcliff et al. 2009). Three scores were computed: an overall PDS score (PDSS), an adrenal axis score (PDSA) and gonadal axis score (PDSG). Sex steroids (DHEA, estradiol and testosterone) were measured using salivary assays that were obtained on 3 separate days over a one-month period.
Results: Reward cue activation was found in the bilateral striatum (caudate and putamen) and activation was negatively correlated with PDSG scores and levels of estradiol in girls, such that girls more advanced in gonadarche and with higher levels of estradiol showed reduced striatal activation to reward cues. There were no significant associations in boys.
Conclusions: Our results provide evidence for sex-specific puberty effects on neural activity during reward anticipation. Given evidence of reduced striatal response to reward in depression, these findings pertaining to pubertal maturation and functioning of the reward circuitry could represent possible markers of vulnerability to depression in girls. Future studies will confirm the effects of pubertal maturation on the development of reward systems and behavior through employment of a longitudinal design in at-risk youth.
Disclosure: Nothing to Disclose.

48.3 Developmental Origins of Sex Differences in Adult Mood

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Background: Major depressive disorder (MDD) is a debilitating disorder of altered mood regulation. Despite the substantial financial and emotional burden of MDD, understanding the pathological and molecular features of this disorder remains a considerable challenge in neuropsychiatry research. One major risk factor for depression is sex: one in four women, but only one in ten men, will experience a debilitating episode of MDD in the course of a lifetime. Studies in human subjects and in rodent models suggest a developmental origin for mood disorders. Interestingly, a developmental process also establishes sex differences in the brain, as developmental exposure to testosterone around the time of birth and through adolescence permanently masculinizes the structure of several brain regions (termed organizational effects of hormones). Notably, adolescence is also a sensitive developmental time-period in which there is extensive neuroanatomical, functional, and chemical brain maturation. Events during adolescence that interact with these developmental processes can increase risk for adult psychopathology. Thus, we hypothesize that sex differences in exposure to gonadal hormones during these critical periods of development cause sex differences in adult mood-related behaviors.
Methods: Although researchers are attempting to determine the mechanisms underlying these sex differences, it is impossible to distinguish between the roles of genetic sex and gonadal sex in humans, since genetic sex determines

gonadal sex. Therefore, we have been using the genetically-engineered Four Core Genotypes (FCG) mice, in which genetic and gonadal sex are decoupled, to examine the mechanisms underlying observed sex differences in humans. This study used adult FCG mice of each genotype [XX gonadal females ($n=21$), XY gonadal females ($n=21$), XX gonadal males, ($n=16$), and XY gonadal males ($n=19$)]. In adulthood, all mice were gonadectomized to remove endogenous sources of gonadal hormones. This step is necessary to isolate potential effects of developmental hormone exposure from activational effects of circulating hormones in adulthood. We assessed mood-related behavior using the novelty suppressed feeding (NSF) test and the social conditioned place preference (CPP) test (a test for social reward). All experimental protocols in 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: We found that gonadal males, regardless of genetic sex, had reduced latency to eat in the NSF test ($p<0.01$). There was no effect of genetic sex ($p>0.5$) and no interaction of genetic and gonadal sex on latency to eat ($p>0.3$). In the social CPP test, gonadal males showed a stronger preference for the social condition compared to gonadal females ($p<10\text{-}4$). Again, there was no effect of genetic sex ($p>0.9$) and no interaction of genetic and gonadal sex on social CPP scores ($p>0.1$).
Conclusions: Notably, since all mice were gonadectomized in adulthood, our findings indicate that sex differences in gonadal hormone exposure during critical periods of development permanently program mood-related behaviors in adulthood. Together, these results suggest that the elevated incidence of MDD in women partially results from early life hormonal processes interacting with development of adult mood.
Disclosure: Nothing to Disclose.

48.4 Pubertal Testosterone Programs Experience-Dependent Expression of ΔFosB in the Infralimbic Medial Prefrontal Cortex to Facilitate Social Competence in Adulthood

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Background: Adolescence is associated with increased prevalence of psychiatric disorders that involve dysfunction in social cognition and disproportionately affect males and females. An important component of social cognition is social competence–the ability to make behavioral adaptations as a function of social experience. Acquisition of social competence during adolescence is particularly advantageous for successful social interactions in adulthood. Here we investigated the contributions of pubertal testosterone and ΔFosB, a transcription factor linked to experience-dependent neural plasticity, to the adolescent maturation of social competence in male-female social interactions.
Methods: To determine whether pubertal testosterone organizes neural circuits underlying social competence, Experiment 1 compared behavioral adaptations to sexual experience in male Syrian hamsters that were deprived of testosterone during puberty (prepubertal castration; NoT@P) to those of males deprived of testosterone for an equivalent period of time in adulthood (postpubertal castration; T@P). All males received testosterone replacement for two weeks before sexual behavior testing in adulthood, when they were allowed to interact with a receptive female once per week for five weeks. Sexual competence, defined as a decrease in ectopic (mis-directed) mounts with sexual experience, and ΔFosB immunoreactivity in medial prefrontal cortex (mPFC) were quantified. To determine whether ΔFosB expression facilitates social competence, Experiment 2 assessed the effects of ΔFosB viral vector-mediated over-expression in mPFC on competence in NoT@P males. To identify a potential neurobiological mechanism by which social experience and ΔFosB modify social competence, Experiment 3 examined effects of experience and over-expression of ΔFosB on dendritic spine density in mPFC of gonad intact adult males.
Results: Experiment 1: T@P males showed the expected decrease in ectopic mounts with sexual experience (i.e., they became socially competent), whereas NoT@P males continued to show high rates of ectopic mounts even with experience (i.e., they did not become socially competent). Sexual experience resulted in increased ΔFosB expression in infralimbic (IL) mPFC in T@P, but not NoT@P, males, indicating that in the absence of pubertal testosterone, induction of ΔFosB expression by social experience is dysregulated in adulthood. Experiment 2: Over-expressing ΔFosB in the IL of NoT@P males prior to sexual experience was sufficient to reverse high rates of ectopic mounting and resulted in a T@P behavioral phenotype. Experiment 3: Both sexual experience and over-expression of ΔFosB in IL in adult gonad intact males increased the density of immature spines on IL dendrites.
Conclusions: This study shows that pubertal testosterone programs the ability to make behavioral adaptations as a function of social experience, thereby facilitating social competence in adulthood. Findings further propose a neurobiological mechanism for the organization of social competence: pubertal testosterone permits upregulation of ΔFosB in the ventromedial mPFC by adult experiences, leading to heightened synaptic lability that may be necessary for behavioral adaptations to social experience.
Disclosure: Nothing to Disclose.

Panel
49. Novel Neuroimaging Biomarkers of Treatment Response From the EMBARC Study

49.1 Examination of Reward-Related Ventral Striatal Activity in Relation to Treatment Response in Major Depressive Disorder: Findings From the EMBARC Study

Tsafrir Greenberg

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Background: Abnormalities in reward neural circuitry centered on the ventral striatum (VS) and medial prefrontal

cortex have been repeatedly reported in major depressive disorder (MDD). However, few studies have examined the extent to which reward-related neuroimaging measures predict antidepressant treatment response. We examined the relationship between reward expectancy (RE) and prediction error (PE) related right VS activity and treatment response in individuals with MDD recruited for the EMBARC study, a large multi-site placebo-controlled clinical trial of sertraline (SERT).

Methods: Participants were 134 individuals with MDD who completed two functional magnetic resonance imaging (fMRI) sessions, at baseline and one week after treatment onset, while performing a reward task. We examined reward expectancy (RE) and prediction error (PE) related right VS activity in relation to treatment response (at week 8) to sertraline and placebo. Treatment response was assessed with the Clinical Global Improvement scale.

Results: A 2 (condition: RE, PE) × 2 (session: 1, 2) × 4 (group: SERT responders, SERT non-responders, placebo responders, placebo non-responders) ANOVA showed a significant session by group interaction ($F = 2.75$, $p = .045$), and a trend for a condition × session by group interaction ($F = 2.32$, $p = .078$). SERT responders and placebo non-responders showed the normal pattern of VS activity reduction across scanning sessions that we previously reported. By contrast, SERT non-responders and placebo responders, showed an increase in VS activity from session 1 to session 2. These differential VS activity patterns in SERT responders and SERT non-responders were also observed when controlling for age, sex, education, anxiety symptoms, and depression severity.

Conclusions: These findings suggest that abnormal functioning of dopaminergically-modulated reward circuitry may characterize sertraline non-responders but not sertraline responders.

Disclosure: Nothing to Disclose.

49.2 Cerebral Blood Perfusion as a Biomarker to Predict Treatment Outcomes in Major Depressive Disorder

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Background: Arterial spin labeling (ASL) is a noninvasive, functional, resting-state neuroimaging technique used to measure cerebral blood flow (i.e., perfusion). To date, cerebral blood perfusion has been used as a biomarker to understand resting state abnormalities in patient populations such as major depressive disorder (MDD). Neuroimaging research in MDD using ASL-derived perfusion has identified abnormalities in the default mode network, salience, higher order sensory perceptual processing, emotion and reward processing (Cooper et al., R&R JAMA Psychiatry; Orosz et al., 2012). Furthermore, perfusion in the anterior cingulate cortex has also been observed to accurately classify unipolar from bipolar depression (Almeida et al., 2013). Perfusion has the prospect of being used as a biomarker of treatment outcomes in MDD. The EMBARC (Establishing Moderators and Biosignatures of Antidepressant Response for Clinical Care) study is a nation-wide randomized control trial investigating such biomarkers in MDD, particularly early biomarkers of treatment outcomes (i.e., to drug versus placebo). Cerebral blood perfusion is being investigated for prediction, moderation and mediation of treatment outcomes.

Methods: Patients in the EMBARC study were scanned at baseline, prior to starting treatment, and at 1 week after starting treatment at one of four sites. Pseudo-continuous arterial spin labeling (PCASL) was performed at rest for time-points. The sequence lasted approximately 6 minutes. Relative cerebral blood perfusion was extracted for regions of interest involved in the salience, reward, emotion, and default mode networks. A mixed effect model was conducted for placebo versus antidepressant care (i.e., sertraline) on Hamilton Depression Rating Scale (Hamilton, 1960) scores across the duration of the study (e.g., 8-weeks).

Results: Using PCASL, relative cerebral blood perfusion in bilateral amygdala (left $p = .002$; right $p = .02$), left posterior cingulate cortex ($p = .03$; right was moderate at $p = .06$), right insula ($p = .02$; left was moderate at $p = .06$), and bilateral inferior parietal lobule (left $p = .002$; right $p = .01$) predicted treatment outcomes independent of treatment type, sertraline versus placebo. Additionally, perfusion in the left anterior cingulate cortex moderated outcomes to treatment ($p = .03$) with a differential change in treatment outcome by treatment type.

Conclusions: While ASL has been more widely used as a research tool to characterize populations, it has the prospects of being used as a tool for clinical diagnostics and informing treatment decisions. Our present work provides evidence that perfusion can predict treatment outcomes for MDD. Moreover, the regions that predict and moderate treatment outcomes are those implicated to be important in the phenotype of MDD. This data present resting-derived measures of brain function as successful in predicting treatment outcome. This work may have implications for future studies aimed at further developing CBF as a biomarker in other clinical populations, particular mood and anxiety disorders.

Disclosure: Nothing to Disclose.

49.3 Resting-State Functional Connectivity Separates Patients With Major Depressive Disorder Into Clinically Distinct Subgroups

Gagan Wig

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Background: There is significant variability in the clinical presentations of Major Depressive Disorder (MDD). Any two individuals diagnosed with MDD may only share a few symptoms among many. It is hypothesized that some of this clinical variability is due to distinctions in brain network dysfunction across patients. However, few studies have found evidence that brain network-based descriptions can distinguish patients into clinically meaningful subgroups based on traditional analytic approaches. To explore this possibility, we examine a subset of functional MRI brain measures collected from a large cohort of MDD patients, use

a data-driven clustering approach to separate patients into subgroups, and evaluate their clinical profiles and medication outcomes.
Methods: Data were analyzed from a large multi-site placebo-controlled trial of un-medicated patients ($n = 187$ after data processing and quality control, 18-65 yrs) with early-onset MDD (first episode < 30 years of age) from the Establishing Moderators and Biosignatures of Antidepressant Response for Clinical Care (EMBARC) study. For each participant, a resting-state functional connectivity (RSFC) matrix was built by measuring the pair-wise correlations in the blood oxygen level-dependent (BOLD) timeseries of putative neocortical areas and anatomically-defined subcortical regions. We focused on the patterns of RSFC in individuals' emotion regulation and reward circuits. A clustering algorithm (k-means clustering) was applied to patient-to-patient RSFC similarity matrices to identify subgroups of patients who exhibited similar patterns of RSFC. These subgroups were then evaluated to determine whether they exhibited similar clinical profiles, and medication outcome.
Results: Ten patient clusters were identified using RSFC clustering. The identified subgroups exhibited multivariate clinical patterns that distinguished them from one-another. The RSFC-derived MDD subgroups also exhibited unique patterns of RSFC in comparison to one another and compared to non-MDD healthy controls. One of the identified MDD subgroups (i.e., patients with increased ventral anterior cingulate cortex RSFC, and symptoms of mania and insomnia) exhibited significantly elevated treatment response to sertraline (75% remission vs. sample-average 38%). Critically, the presence and characteristics of the RSFC-defined subgroups in this initial sample—including the subgroup with an elevated sertraline remission rate—were validated in a left-out test sample of subjects ($n = 80$).
Conclusions: These findings suggest that RSFC network patterns may be used to differentiate patients with MDD into clinically meaningful subgroups, including some with treatment-moderating features, and may assist with clinic-based diagnoses.
Disclosure: Nothing to Disclose.

49.4 Comorbid Anxiety, Anger, and Anhedonia as Moderators for Recruitment of Emotion Regulation Neurocircuitry in Major Depression

Kristen Ellard

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Background: Major depressive disorder (MDD) is associated with abnormalities in the processing and regulation of emotion. Recent studies suggest heterogeneity in patterns of neural activation during emotion processing corresponding to specific subtype presentations of MDD. To date, investigations of specific MDD subtypes have occurred using different paradigms, methodologies, and single subtype classifications. The purpose of the present study is to directly compare MDD subtype presentations using fMRI and a single, well-validated emotion regulation task. Drawing from a large multi-site trial of MDD, the current study examines differential patterns of neural activation during emotion processing between MDD with anger attacks (MDD+Ang), anhedonia (MDD+Anh), and anxiety (MDD+Anx). We examine the effect of MDD subtype on recruitment of neurocircuitry previously implicated in successful emotion regulation [dorsolateral prefrontal cortex (DLPFC), ventrolateral prefrontal cortex (VLPFC) anterior cingulate cortex (ACC), insula and amygdala].
Methods: Participants include 188 unmedicated patients with MDD. Patients performed the Emotion Conflict Resolution (ECR) task, a well-validated task assessing implicit emotion regulation, while undergoing BOLD fMRI. Participants are presented images portraying either happy or fearful facial expressions overlaid with the word "happy" or "fear" and are asked to indicate the expression on the face (happy or fear) while ignoring the word. Trials included both "congruent" (same facial expression/word) and "incongruent" (different facial expression/word) presentations. Participants must regulate the affective response to incongruence in order to answer correctly, serving as a proxy for implicit emotion regulation. Preprocessing and analysis of all fMRI data were run using SPM8. First-level linear contrasts between congruent and incongruent trials (incongruent > congruent) were computed for all participants. Within-group regressions between Incongruent-Congruent contrasts and psychometric scales assessing anger attacks (Anger Attacks Questionnaire), anhedonia (Snaith-Hamilton Pleasure Scale) and anxiety (Mood and Anxiety Symptom Questionnaire) were computed in specific a priori ROIs (DLPFC, VLPFC, ACC, insula and amygdala).
Results: All three depression subtypes demonstrated significant negative correlations with DLPFC, VLPFC, ACC and anterior insula activation during incongruent trials relative to congruent trials (all $p < .001$). Elevations in anger attacks (MDD+Ang) differentiated from other subtypes by a significant positive relationship to amygdala activation during incongruent > congruent trials ($t = 2.57$, $p = .005$). Elevations in anhedonia (MDD+Anh) differentiated from other subtypes by a significant negative relationship to amygdala ($t = 3.17$, $p = .001$), posterior insula ($t = 5.11$, $p < .001$) and posterior cingulate ($t = 3.81$, $p < .001$) activation during incongruent > congruent trials.
Conclusions: Results of the current study suggest differential recruitment of the broader neurocircuitry supporting emotion regulation differentiates depression subtypes. Elevations across all three subtypes were associated with less recruitment of core regulatory structures during emotion regulation, suggesting a core deficit across domains of depression. Anhedonia was further differentiated by less amygdala and posterior insula recruitment, and less posterior cingulate recruitment during emotion regulation, suggesting decreased processing of salience. By contrast, anger attacks were associated with greater amygdala activation during emotion regulation, suggesting increased processing of salience. The implications of differential engagement of the broader neurocircuitry supporting emotion regulation for the development of dimensional assessment and targeted interventions will be discussed.
Disclosure: Nothing to Disclose.

Panel
50. Tracking and Manipulating Drug-Cue Reactivity: An Interventional Target for Relapse Prevention in Substance Use Disorders

50.1 Fos-Expressing Neuronal Ensembles Encode Drug-Cue Interactions in Animal Models of Drug Addiction

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Background: Learned associations between environmental stimuli and drug rewards drive goal-directed learning and motivated behavior in human addicts and animal models of addiction. These learned associations are thought to be encoded within specific patterns of sparsely distributed neurons called neuronal ensembles that are selectively activated during learning and recall of drug-associated memories. The most strongly activated neurons induce the neural activity marker Fos that allows us to identify and manipulate these neuronal ensembles and identify unique molecular and cellular alterations within these ensembles.
Methods: Fos-positive neurons were identified using immunohistochemistry. We manipulated specific Fos-expressing neuronal ensembles using the Daun02 inactivation method in FosLacZ transgenic rats. We used fluorescence-activated cell sorting (FACS) followed by qPCR to identify altered gene expression within behaviorally activated Fos-immunolabeled neurons in rats. We used slice electrophysiology to identify intrinsic excitability and synaptic alterations in Fos/GFP-labeled neurons in FosGFP transgenic rats and mice.
Results: We will present recent and unpublished findings using different animal models of drug and cue associations in rats and mice. In behavioral experiments, Daun02 inactivates different patterns of Fos-expressing ensembles that were strongly activated by different cues or training conditions. Inactivation of drug-paired Fos-expressing ensembles attenuated drug-paired behaviors, while inactivation of non-drug-paired Fos-expressing ensembles had no effect on the drug-paired behaviors. In molecular experiments, gene expression (e.g. Fos, FosB, Arc, GluN2A) was increased only in the Fos-positive neurons, and not in the Fos-negative majority of neurons, when placed in either drug-paired or non-drug-paired environments. In electrophysiology experiments, silent synapses (in nucleus accumbens) and intrinsic excitability (in prelimbic cortex) were increased only in Fos-positive neurons, and intrinsic excitability was decreased in the Fos-negative majority of neuron, when placed in the drug-paired, but not when placed in the non-drug-paired environment.
Conclusions: Using different animal models of operant learning, acquisition versus extinction, and cue- and context-induced reinstatement, we have consistently found that Daun02 inactivation of Fos-expressing neurons (that were previously activated by drug-related cues) in different brain areas attenuated subsequent drug-associated behavior. Molecular and cellular alterations induced only in these drug-associated Fos-expressing neuronal ensembles may be targets for selective manipulation of drug-related memories in human addicts.
Disclosure: Nothing to Disclose.

50.2 Modulation of Drug-Cue Reactivity During Abstinence and With Cognitive Training can be Quantified Using Translational Psychophysiological Markers

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Background: Drug addiction is a chronically relapsing disorder. Relapse can be precipitated by re-exposure to cues previously associated with drug use that evoke craving. An underlying mechanism of drug-cue reactivity and subsequently induced craving encompasses enhanced spontaneous attention to drug-related cues, which in turn is associated with (re-)initiation of drug-seeking behavior in addicted individuals. For relapse prevention, it is therefore imperative that the attention afforded to drug-cues is reduced and that such reduction can objectively be measured. This presentation will present results of two recent studies showing that the Event-Related Potentials (ERPs) can robustly quantify motivated attention to drug cues as well as detect meaningful change therein.
Methods: In the first study, 76 individuals with cocaine use disorder (iCUD) with varying durations of abstinence (i.e., 2 days, 1 week, 1 month, 6 months, and 1 year) were presented with cocaine-related pictures (drug cues) during which ERP data was acquired. Amplitude of the late positive potential (LPP) component of the ERP, a temporally precise measure of motivated attention, was used as the marker of drug-cue reactivity. In the second study, LPP data was acquired from 37 iCUD and 23 healthy controls (HC) while they completed a cognitive reappraisal task, in which they viewed drug-related (e.g., pipe) pictures either normally or down-regulated their reactivity using cognitive reappraisal strategies. Eye-tracking was used to quantify the ensuing spontaneous attention to drug cues.
Results: The first study demonstrated that ERP markers can track the dynamics of cue-reactivity with increasing abstinence duration. The LPP amplitudes showed an inverted U-shaped trajectory, such that it increased from 2 days to 1- and 6-months before declining at 1 year. In contrast, the subjective assessment of baseline craving and cue-induced craving showed a linear decline from 2 days to 1 year of abstinence. The second study suggested that these LPP-based markers can quantify self-regulation of cue-reactivity (i.e., reduced LPPs during down-regulation compared to normal viewing of drug cues) and that eye-tracking can be used to assess the immediate after-effects of self-regulation on spontaneous attention to drug-related cues, such that gaze duration during viewing of drug-related pictures after the self-regulation compared to normal viewing of the drug cues was significantly reduced in iCUD.
Conclusions: In the first study, we showed that cue-reactivity follows an inverted-U shaped trajectory, similar to pre-clinical findings of incubation of cue-reactivity during early periods of abstinence. These results highlight periods of high vulnerability to relapse – a clinically crucial finding. In the second study, again using LPP, we show that iCUD can self-regulate their reactivity to drug-cues, which in turn has a

direct impact on reducing spontaneous attention to drug-related cues – a desirable clinical outcome for relapse prevention. Together, these results underscore the effectiveness of drug cue-reactivity as a potential target for intervention and the utility of psychophysiological biomarkers, especially those with high translational potential, to track the performance and outcomes of these interventions.
Disclosure: Nothing to Disclose.

50.3 Attenuating Cue-Reactivity With Human Continuous Theta Burst Stimulation to the Ventromedial Prefrontal Cortex: Results From the First Two Clinical Trials in Cocaine and Alcohol Users

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Background: The ventromedial prefrontal cortex (vmPFC) is a critical node in the neural circuitry of drug craving. Preclinical research has established that optogenetic manipulation of the infralimbic cortex {the rodent homologue of the vmPFC} can decrease cocaine and alcohol self-administration. This presentation will describe the results of two recent brain imaging and stimulation studies which evaluated continuous theta burst stimulation (cTBS, a new form of TMS) as a tool to change brain reactivity to drug cues.
Methods: The primary dependent measure of both studies was a change in the brain response to drug cues immediately before and after the cTBS treatment (FDR-corrected clusters, $p < 0.05$). In study 1 (SINGLE DAY, $n = 49$) participants received 1 session of real cTBS (3600 pulses to the left frontal pole) and one session of sham cTBS (1-week intervisit interval, within subject control). In study 2 (MULTIDAY, $n = 41$) participants are receiving 10 sessions of Real of Sham cTBS (between group control).
Results: In the first study, compared to sham, real cTBS induced a significant decrease in the vmPFC, anterior cingulate cortex, and insula BOLD activity within the cocaine users. In alcohol users, relative to sham, real cTBS induced a significant decrease in the left vmPFC and insula BOLD signal. The brain response to real versus sham cTBS was not significantly different between cocaine and alcohol users. In the second study, the results remain blinded but thus far 82% of eligible participants have completed all 10 days of treatment, and 100% of them believe that they are receiving real cTBS (rather than sham). Preliminary analysis demonstrates that the cue-associated brain reactivity is significantly lower in the anterior cingulate and insula in one group than it was before commencing cTBS treatment. Neuroimaging data and clinical outcome data will be completed in the Fall 2017 and presented at ACNP.
Conclusions: Both the single day and multiday studies highlight the emerging value of vmPFC theta burst stimulation as a tool to attenuate brain reactivity to drug cues, specifically in the cingulate and insula - brain regions critically involved in craving.
Disclosure: Nothing to Disclose.

50.4 Paced Breathing Changes Neural Reactivity to Alcohol Cues

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Background: The baroreflex is a neurocardiac mechanism that buffers blood pressure and is cardioprotective. Recent research suggests a further role in affecting automatic, visceral reactivity to environment by regulating bidirectional communication between the heart and the brain. This mechanism can be enhanced by breathing paced at 0.1 Hz (six breaths-per-minute; 6 P). 6 P increases baroreflex sensitivity and heart rate variability, improves neurocardiac functioning, and may improve behavioral flexibility. Through momentary manipulation of the baroreflex via 6 P, it may be possible to regulate neurophysiological processes that affect neural reactivity to substance use triggers. This study examined the effects of 6 P on neural cue reactivity to visual alcohol cues and subjective craving response in the fMRI environment.
Methods: Men and women ages 18-25 years ($N = 48$), who met criteria for either DSM-IV-TR alcohol dependence (AUD) or NIAAA "low-risk" alcohol consumption (LR), were randomized to one of two intervention conditions: 6 P (active intervention) or control. fMRI BOLD data were collected during the following cue exposure paradigm: neutral (nature) images (Set 1), personalized alcohol images (Set 2), randomized condition (6 P or control), personalized alcohol images (Set 3). Acute alcohol craving was assessed following each image set with a single visual analogue scale (VAS). Change in BOLD response to alcohol images from pre- to post-intervention was assessed separately in the 6 P and vanilla groups using FSL's FEAT. The pre > post contrast assessed decreased activation, and the post > pre contrast assessed increased activation. Activation was thresholded at $z > 1.65$ and alpha was $p < .05$. The effects of cue type (neutral versus alcohol) and diagnosis (AUD versus LR) on craving were analyzed using mixed model ANOVA at $p < .01$.
Results: There was a main effect of cue type on craving; participants endorsed higher craving to alcohol compared to neutral cues ($F(1, 41) = 15.93$; $p < .001$). There also was a main effect of diagnosis; the AUD group endorsed higher craving compared to the LR group ($F(1, 41) = 10.70$; $p < .01$). In the 6 P group, but not the control group, the pre > post contrast indicated decreased activation in the left lateral occipital cortex, left inferior temporal gyrus, and left temporal fusiform cortex during the second alcohol cue exposure. As well, the pre < post contrast in the 6 P group, but not the control group, showed increased activation in the brain stem, bilateral precuneus cortex, bilateral posterior cingulate cortex, bilateral frontal medial cortex, bilateral paracingulate cortex, and left subcallosal cortex during the second alcohol cue exposure. Analyses are ongoing, including subjective craving relations to neural response.
Conclusions: Participants' experience of craving in the fMRI environment was dynamic; it changed depending on cue type. This emphasizes the importance of momentary context to subjective craving states and suggests that craving can be

estimated in-the-moment with a single item VAS within an fMRI environment following a brief presentation of visual alcohol cues. The paced breathing intervention changed neural response to alcohol-related images. Paced breathing is a potentially promising relapse prevention tool that could be deployed in the face of, or in anticipation of, high risk triggers for substance use outside the treatment context.
Disclosure: Nothing to Disclose.

Panel
51. New Drugs for PTSD Prevention and Treatment: Are Molecular Targets of PTSD Prevention and PTSD Treatment Similar?

51.1 A Roadmap to Medication Assisted Augmentation of Psychotherapy for Posttraumatic Stress Disorder

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Background: Several psychotherapeutic procedures have been identified for the treatment of PTSD: patients are confronted to increasingly difficult feared situations while applying cognitive restructuring techniques and forms of avoidance strategies. Memory is a good endophenotype of PTSD. Fear learning and extinction is the prevailing model. Compelling evidence exists that fearful memory becomes labile upon re-exposure to cues that are related to that particular fearful experience.
Targeting reconsolidation - as is proposed in exposure-based psychotherapy - offers a potentially effective tool to manipulate fear memories, and subsequently treat PTSD. Recently several compounds have been identified to be used in conjunction with this procedure. Medication provided before or after exposure therapy can enhance outcome by: strengthening learning and memory of fear extinction; manipulating reconsolidation, thereby reducing the expression of fear; altering prediction error towards new learning; facilitating engagement in psychotherapy by reducing fear and enhancing openness to experience.
Medication assisted augmentation of psychotherapy offers a unique opportunity to target emotional memories and the expression of fear. Some validation from clinical studies exist. There is a need for a roadmap that will assist in identification and evaluation of compounds in their efficacy in treatment for PTSD.
Methods: Trials are evaluating modulation of glucocorticoids neurosteroids, glutamate, GABA, endocannabi-noids, oxytocin, neurokinin/Substance P, and dopamine. It is not exactly known what the optimal dosing is, as well as which parameters need to be addressed to assess their clinical utility for the treatment of symptoms of PTSD.
Results: Data are available for the following compounds, and receptor targets: Propranolol (b-adrenergic receptor), Hydrocortisone (corticosteroid receptor), Mifepristone (glucocorticoid receptor antagonist), D-Cycloserine (partial agonist at glycine site of NMDA receptor), Ketamine (NMDA receptor antagonist), Oxytocin (oxytocin receptors), mGLUR2/3 modulators (mGLUr2/3 receptors), MDMA (serotonin and norepinephrine transporters, oxytocine release), LSD (5-HT2A, D2 receptors).
Parameters that are needed to assess the effect of the intervention: preclinical/clinical; Study population, incl. sex (veterans, MVA, other); memory phase (consolidation /retrieval/extinction/reconsolidation); time of administration (hours/mins; before, during, after exposure); mode of administration (oral, intranasal, other); number of sessions (single, multiple); dosing (mg, ml); therapeutic strategy/ reactivation procedure (VRET, imaginary, EMDR, other); identification of SUDS/emotional involvement during therapy; assessment of outcome (PTSD, fear, intrusions, sleep); follow-up (days, weeks, months).
Conclusions: Targeting retrieval, extinction or reconsolidation is a potentially promising avenue targeting fearful memories in PTSD. Several compounds exist that are or will be investigated in near future for medication assisted augmentation of psychotherapy for PTSD, as well as compounds that facilitate the engagement in therapy by reducing fear may be targeted for a specific population. Success of the manipulation depends on subtle differences in the reactivation procedure. These interventions require careful planning, and collaboration with prescribers. This roadmap will assist in moving the field forward in terms of design, dosage as well as effectivity as augmentation strategies for treatment of PTSD.
Disclosure: Nothing to Disclose.

51.2 Long-Term Effects of High-Dose Hydrocortisone Immediately After Trauma: A Double Blind, Placebo-Controlled Prospective Study

Joseph Zohar

Research Foundation by the Sheba Medical Center, Israel, Ramat Gan, Israel

Background: Blunted and/or hypoplasticity of HPA axis was found to be associated with increase susceptibility to post-traumatic stress response in rats (Cohen, Zohar et. Al, 2006). An increasing body of indirect evidence suggests that adequate cortisol response, in the first few hours after exposure to traumatic events has a protective effect against developing post-traumatic stress syndrome (McFarlane et. Al, 2011). However, administration of hydrocortisone in the aftermath of potentially traumatic event was not studied yet.
Methods: Individuals who were exposed to traumatic events that arrived at the ED at Sheba Medical Center (Tel Aviv, Israel) within 6 hours were screened. Those who experienced clinical significant response, were not physically injured and signed an informed consent were recruited to the study. The participants were randomized by a predetermined program, and entered in a D/B, placebo-controlled design. Hydrocortisone or placebo was given within the first 6 hours after traumatic events.
Patients were followed up at 6 time points: before the intervention, at two weeks, one month, three months, eight months and a final visit at 13 months. Blood samples for Cortisol, ACTH, DHEA, DHEA-S, NPY and methylations were taken prior the intervention and at 3 months and at the final visit. The dexamethasone suppression test (DST) was used to assess negative feedback inhibition of the HPA axis at 3 and 13 month. The patients were rated with CAPS, CGI and MADRAS at 1, 3, 8 and 13 months.

Results: 118 patients were recruited to the study after signing an informed consent. 20 patients were withdrawn at follow up and 3 were excluded from the study. 45 received placebo and 50 received hydrocortisone. Out of all the participants (who completed the study (95) 7 had PTSD at the 13 months follow up.
When comparing the hydrocortisone group versus the placebo group there was no difference of PTSD rate at the end of the study. However, taking into account (a) the diurnal variation of cortisol secretion (higher in the day and lower in the night) and (b) dividing the group into those who were exposed to the traumatic event from 8:00 to 18:00 (N = 44) and those who were exposed to the traumatic event from 18:00 up midnight (N = 6) (we didn't have staff at the ED from midnight to 8:00) – a different picture had emerged. None of those who received hydrocortisone from 18:00 to midnight developed PTSD at 13 months.
However, 3 of 39 who received placebo during the day developed PTSD and 3 out of 44 who received hydrocortisone during the day developed PTSD at 13 months follow up.
Moreover, individuals who had low plasma level of cortisol and NPY at the ED, before the intervention, tend to have more PTSD symptoms at the 13 months follow up. The other biological measures are currently being analyzed and will be presented in the session.
Conclusions: The time of day of the traumatic exposure markedly affected the pattern of
the behavioral stress response. Single dose injection of hydrocortisone (100-140 mg) within 6 hours after exposure to traumatic events has a long-term effect (13 months later) if given when cortisol levels are low (during the night). The small number of individuals who received hydrocortisone shot after 18:00 (N = 6) has hampered the interpretation of the result. Nevertheless, looking at NNT analysis, it suggests NNT = 7 for significant favorable alternation in the trajectory for those who received hydrocortisone after 18:00.
Some of the biological data (e.g. cortisol levels at ED) are in line with the hypothesis that (hypo)-plasticity of HPA axis might be implicated in the response to exposure to traumatic events.
Disclosure: Part 1: Brainsway, Grant, Lundbeck, Grant, Servier, Grant, Pfizer, Grant, Servier, Advisory Board, Pfizer, Advisory Board, Abbott, Advisory Board, Roche, Advisory Board, Lundbeck, Honoraria, Roche, Honoraria, Lilly, Honoraria, Servier, Honoraria, Pfizer, Honoraria, Abbott, Honoraria.

51.3 The Effect of Hydrocortisone Augmentation on Prolonged Exposure Psychotherapy Outcomes

Rachel Yehuda

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Background: There is great diversity in responses to cognitive behavioral therapy in PTSD prompting an examination of augmentation strategies. Since glucocorticoids are altered in PTSD, the current study examined whether the administration of glucocorticoids could enhance the effects of an exposure based psychotherapy in combat veterans with PTSD.
Methods: 96 combat veterans were enrolled, and 60 were randomized in a clinical trial examining the effects of a 30 mg dose of oral hydrocortisone (Hcort) or placebo (administered prior to imaginal exposure sessions 3-12) in reducing PTSD symptoms in response to prolonged exposure (PE) therapy. Response status was determined by presence or absence of PTSD at treatment end. Participants were also evaluated at a 3-month follow-up. A complete psychological evaluation and blood draw was obtained at pre-treatment, post-treatment, and at follow-up.
Results: PTSD severity, as assessed by total scores on the Clinician Administered PTSD Scale (CAPS) for DSM-IV, declined from a mean of 85.9 ± 14.1 to 51.5 ± 22.0 from pre- to post treatment with 36 percent no longer meeting criteria for PTSD at post-treatment. Hcort augmentation was not associated with clinical improvement in comparison to placebo in the sample as a whole. However, there was a significant advantage of Hcort in the subgroup of combat veterans with mild traumatic brain injury (mTBI). Effects of hydrocortisone in responders appeared to be mediated by cognitive and biological variables. Machine learning approaches were employed to identify predictors of treatment outcome in general, and in response to Hcort.
Conclusions: There may be a role for hydrocortisone augmentation in some patients with PTSD. Knowledge of a series of biological and cognitive factors may help in the prediction of treatment response, and in the tracking of psychotherapeutic outcome.
Disclosure: Nothing to Disclose.

51.4 Novel Therapeutics in PTSD: A Randomized Clinical Trial of Mifepristone

Julia Golier

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Background: PTSD is an often debilitating illness for which few effective pharmacological treatments exist. Given evidence of dysregulation of the HPA axis, including of enhanced glucocorticoid receptor sensitivity, we sought to assess the safety and efficacy of mifepristone, a type II glucocorticoid receptor antagonist, in the treatment of PTSD.
Methods: This was a phase IIa double-blind RCT of mifepristone. Eighty male veterans were randomized to treatment with mifepristone (600 mg/day) or placebo for seven days followed by periodic assessment over three months. The primary analysis was based on responder status (> = 30% reduction in CAPS score) at 4 weeks.
Results: The drug was well-tolerated with no group differences in adverse events. At four weeks, 38% of mifepristone vs. 31% of PBO treated veterans showed a clinical response, a difference that did not reach the threshold for an efficacy signal. Interesting treatment-responsive subgroups emerged in relation to severity of PTSD symptoms, comorbid depressive disorder, and TBI status.
Conclusions: The primary data do not support efficacy for mifepristone in a general population of male veterans with PTSD. Exploratory analyses suggest there may be more responsive subgroups for consideration for future

exploration. Information on biological predictors and the effect of drug levels will also be provided.
Disclosure: Nothing to Disclose.

Panel
52. Markers for Course of Illness and Treatment Outcome in Schizophrenia

52.1 Glutamate as a Predictor of Treatment Outcome in Schizophrenia

Alice Egerton

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Background: There is considerable interest in identifying biomarkers of antipsychotic response in schizophrenia, and brain glutamate is one key candidate. Our previous cross-sectional magnetic resonance spectroscopy (MRS) studies have shown that elevated anterior cingulate cortex (ACC) glutamate may be associated with poor treatment response in both early psychosis and established schizophrenia (Egerton et al. Neuropsychopharmacology. 2012;37:2515–2521; Demjaha et al. Biol Psychiatry. 2012;75:e11–e13; Mouchliantis et al. Schizophr Bull. 2016;42:744–752). However longitudinal studies are required to determine whether glutamate levels are predictive of antipsychotic response.
Methods: As part of the OPTiMiSE consortium, we examined glutamate levels in the ACC and left thalamus in minimally-treated patients with first episode psychosis (FEP; n = 71; < 2 weeks' antipsychotic medication). Participants were recruited and scanned across three centers (London, Copenhagen, Utrecht). After MRS acquisition, participants entered a clinical trial whereby they received oral amisulpride for 4 weeks, after which MRS was repeated. Psychopathology was assessed at baseline and at 4 weeks, and the primary outcome measure was whether or not patients reached remission criteria after amisulpride. Glutamate is reported as scaled to creatine (Glu/Cr).
Results: Baseline ACC Glu/Cr was significantly elevated in the non-remission compared to remission group ($F_{1,64} = 5.995$; $P = 0.017$; partial eta2 = 0.09). There was no significant difference between groups in baseline Glu/Cr in the left thalamus. Baseline Glu/Cr in the ACC and thalamus glutamate correlated with symptom severity after amisulpride ($P < 0.05$).
Conclusions: These results suggest that glutamate may be predictive of symptomatic response to antipsychotic administration in first episode psychosis, with those with lower glutamate levels being more likely to respond well.
Disclosure: Nothing to Disclose.

52.2 Glutamate and GABA in Antipsychotic-Naïve Schizophrenia: Association to Treatment Outcome and Cognitive Deficits

Kirsten Bojesen

Center for Neuropsychiatric Schizophrenia Research, Glostrup, Denmark

Background: Recent cross-sectional studies suggest that elevated levels of glutamate in the anterior cingulate cortex (ACC) may serve as a marker for treatment response in schizophrenia patients (SCZ). Here, we investigated glutamate and GABA levels in the ACC and glutamate in the left thalamus during the first two years after the patient's first antipsychotic treatment. The aim was to examine whether baseline levels of glutamate and GABA were different in remitters and non-remitters and whether they changed during treatment. We further explored if ACC levels of glutamate and GABA could predict cognitive performance prior to treatment.
Methods: Longitudinal study of 36 initially antipsychotic-naïve SCZ and 31 matched healthy controls (HC) assessed at baseline, after 6 weeks (N(SCZ) = 23, N(HC) = 29), 6 months (N(SCZ) = 22, N(HC) = 26), and 24 months (N(SCZ) = 18, N(HC) = 10) of treatment. Patients were treated with aripiprazole for the first 6 weeks. Hereafter, treatment could be modified. Remission (R) and non-remission (NR) were assessed using the criteria by Nancy Andreasen. The cognitive domains included were associative learning and memory, sustained visual attention, and spatial working memory tested with CANTAB, and premorbid intelligence. Glutamate spectra in ACC and left thalamus were acquired with the PRESS sequence, and GABA spectra in ACC with the MEGA-PRESS on a 3T MR scanner.
Results: In left thalamus, a significant diagnosis*time interaction was found ($p = 0.003$) due to SCZ having higher glu/Cr at baseline ($p = 0.04$) and lower glu/Cr after 6 months ($p = 0.03$). In ACC, there was a trend toward lower glu/Cr ($p = 0.07$) and GABA/Cr ($p = 0.07$) in SCZ at all assessments compared to HC.
Looking at glutamate levels in the left thalamus in the antipsychotic-naïve state, we found higher baseline levels of glu/Cr in the NR group compared to HC after 6 weeks ($p = 0.03$) and at trend level after 24 months ($p = 0.07$). At 6 months, however, both the R ($p = 0.03$) and the NR groups ($p = 0.03$) showed higher baseline glu/Cr values compared to HC.
For ACC, no difference was found in baseline glu/Cr and GABA/Cr between the R, NR, and HC groups at any assessment.
Exploring if glutamate and GABA differed in the R and NR groups during treatment, we found that the NR group had higher glu/Cr in the left thalamus compared to the R group after 6 weeks ($p < 0.05$) and at trend level after 24 months ($p = 0.07$), but not after 6 months. GABA/Cr was lower in the ACC in the NR compared to the R ($P = 0.04$) group after 6 weeks, but not after 6 and 24 months.
Finally, we investigated if glutamate and GABA in ACC could predict neurocognitive performance in antipsychotic-naïve SCZ and HC at baseline. Glu/Cr significantly predicted performance of sustained visual attention ($p = 0.004$) and premorbid intelligence ($p = 0.03$) in both SCZ and HC. For associative learning and memory, significant Glu/Cr *diagnose ($p = 0.004$) and GABA *diagnose ($p = 0.02$) interactions were found due to worse performance in SCZ with low levels of these metabolites.
Conclusions: The data represent work in progress. They indicate that high glu/Cr in the left thalamus of antipsychotic-naïve SCZ is a marker of a more severe illness trajectory, although not unambiguously associated with non-remission.

On the contrary, low glu/Cr and GABA/Cr in ACC were associated with poor cognitive performance. The degree of glutamatergic and GABAergic disturbances might reflect the severity of the illness, irrespective of metabolite levels being too high or low.
Disclosure: Nothing to Disclose.

52.3 The Ubiquitin Proteasome System is Dysregulated in the Blood and Brain of Individuals With Schizophrenia

Chad Bousman

University of Calgary, Calgary, Canada

Background: Over the past two decades, evidence from genome-wide association, microarray, and protein studies have indicated dysregulation of genes and proteins within the ubiquitin proteasome system (UPS) in the blood and brain of individuals with schizophrenia. However, it is not clear which components of the UPS, if any, are dysregulated in both blood and brain in schizophrenia. As such, the current study measured free ubiquitin, ubiquitinated proteins, ubiquitination activity, and proteasome activity in both erythrocytes and postmortem orbitofrontal cortex tissue from individuals with schizophrenia and controls.
Methods: Frozen postmortem orbitofrontal cortex (OFC) tissue from 76 (38 schizophrenia, 38 control) individuals was obtained from the New South Wales Brain Tissue Resource Centre (Sydney, Australia). In addition, we collected erythrocytes from 181 participants, consisting of 63 individuals with treatment-resistant schizophrenia (mean illness duration = 17 years), 30 recent onset schizophrenia (mean illness duration = 1 year), and 88 healthy control participants recruited from multiple clinical services and the community in Melbourne, Australia. Levels of free ubiquitin and ubiquitinated proteins as well as activity of endogenous ubiquitination ligases (E1–activating, E2-conjugating, and E3), and proteases (capsase-like, chymotrypsin-like and trypsin-like) were quantified in erythrocytes and postmortem OFC blind to diagnosis.
Results: Individuals with treatment-resistant schizophrenia had higher levels of ubiquitinated proteins compared to those with recent onset schizophrenia ($pFDR < 0.001$, Hedges' $g = 0.81$) and healthy controls ($pFDR < 0.001$, $g = 0.79$). Likewise, in postmortem orbitofrontal cortex we detected elevated ubiquitinated protein levels in those with schizophrenia compared to controls ($pFDR = 0.002$, $g = 0.73$). We also detected significantly lower ubiquitination activity in erythrocytes among individuals with treatment-resistant compared to recent onset schizophrenia ($pFDR < 0.001$, Hedges $g = 1.13$) and healthy controls ($pFDR = < 0.001$, $g = 0.79$) but we did not detect this difference in the OFC of those with schizophrenia ($pFDR = 0.151$, $g = 0.33$). Levels of free ubiquitin and proteasome activity (chymotrypsin-like, trypsin-like and caspase-like) did not differ between healthy controls and those with schizophrenia in either the erythrocytes or OFC tissue.
Conclusions: The results suggest that protein homeostasis may be abnormal in both the blood and brain of those with schizophrenia, particularly in the later stages and/or subgroups (i.e. treatment-resistant schizophrenia) of the illness. Given the concordance in blood and brain, elevated ubiquitinated proteins should be investigated further as a marker for treatment-resistant schizophrenia and determine whether it has prognostic utility.
Disclosure: Nothing to Disclose.

52.4 Stratification of First Episode Schizophrenia Patients Based on Multimodal Disturbances Before Initiation of Antipsychotic Treatment: Towards Precision Medicine?

Bjørn Ebdrup

Center for Neuropsychiatric Schizophrenia Research, Glostrup, Denmark

Background: A wealth of clinical studies have identified para-clinical biomarkers, which on a group level separate schizophrenia patients from healthy controls, but inconsistent findings have hampered clinical application.
Recently, we used measures of electrophysiology and cognition in a Gaussian Mixture Model (GMM) and identified two distinct subgroups of antipsychotic-naïve, first-episode schizophrenia patients. Before treatment, these two subgroups did not differ in psychopathology, but the response to six weeks of amisulpride treatment in the groups was significantly predicted (Bak et al. Transl Psychiatry).
Aiming for further clinical translation, we here applied multiple multivariate algorithms on multimodal data from this extensively examined cohort of antipsychotic-naïve schizophrenia patients and matched healthy controls.
Methods: Forty-six patients and 58 controls subjects underwent an extensive neurocognitive test battery, an electrophysiological test battery, structural magnetic resonance imaging and diffusion tensor imaging at 3 Tesla. Patients were assessed on the positive and negative syndrome scale (PANSS) before and after six weeks of antipsychotic monotherapy with amisulpride. On generalizable measures of cognition, electrophysiology, structural magnetic resonance imaging and diffusion tensor imaging, we applied nine different supervised multivariate algorithms to: i) estimate the diagnostic accuracy; ii) explore the potentially added diagnostic value of application of multimodal data; iii) predict symptom remission after six weeks.
Results: With accuracies ranging between 57% and 69%, cognitive data significantly classified patients and controls (p-values between $p = .028$ and $p = .0001$). Data on electrophysiology, gray matter and white matter did not reach accuracies above chance level. Multimodal analyses with cognition plus any combination of one or more of the remaining three modalities did not reveal higher accuracies than cognition alone. No modality significantly predicted treatment outcome after six weeks.
Conclusions: Although multivariate analyses can successfully classify distinct subgroups of schizophrenia, which are predictive of treatment response, our present analyses indicate that only cognitive data significantly discriminated patients from controls. Adding electrophysiological or neuroanatomical data did not improve the diagnostic accuracy.
To facilitate progress from the current symptom-based psychiatry towards precision medicine, these advanced

algorithms may require a careful a priori variable selection and refined data processing procedures.
Disclosure: Part 1: Janssen-Cilag A/S, Advisory Board, Eli Lilly AB, Honoraria, Otsuka Pharma Scandinavia AB, Honoraria.

Panel
53. Leveraging Memory Traces (Engrams) in Rodents to Gain Insights Into Psychiatric Disorders

53.1 Artificially Manipulating Positive and Negative Memories to Alleviate Psychiatric Disease-Like States

Steve Ramirez

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Background: Memories thread and unify our overall sense of being. With the accumulation of our knowledge about how memories are formed, consolidated, retrieved, and updated, neuroscience has reached a point where brain cells active during these discrete mnemonic processes can be identified and manipulated at rapid timescales in both healthy and pathological states.
Methods: We utilize and activity dependent and inducible strategy to tag active cells with rhodopsin effectors. Our system involves a virus cocktail in which the activity of the c-Fos promoter drives a downstream gene of interest and the process is under the control of doxycycline.
Results: Our studies' conclusions are twofold: a defined subset of hippocampus cells are sufficient to elicit the neuronal and behavioral expression of positive and negative memory recall, which are also sufficient to modify social and hedonic behaviors; and, artificially activated positive memories, as well as artificially inhibited negative memories, can be leveraged to acutely and chronically suppress psychiatric disease-related states.
Conclusions: We propose that hippocampus cells that show activity-dependent changes during learning construct a cellular basis for contextual memory engrams and that directly activating these endogenous neuronal processes may be an effective means to correct maladaptive behaviors.
Disclosure: Nothing to Disclose.

53.2 The Influence of Memory on Drug Tolerance

Brian Wiltgen

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Background: The utility of opiates for treating pain is limited by the development of tolerance; a phenomenon that can lead to dose-escalation and an increased liability for dangerous side effects like dependence and overdose. A major goal in neuroscience, therefore, is to understand the mechanisms that give rise to tolerance so that interventions can be designed to prevent it. Interestingly, environmental cues that are associated with opiate drugs exert a dramatic influence over tolerance. Animals that receive morphine in a particular context, for example, exhibit analgesic tolerance that can be eliminated by simply administering the drug in a new environment. The mechanisms underlying this 'associative tolerance' have not been well characterized. However, they are highly relevant clinically as many opiate overdoses occur in novel environments. The goal of our work is to dissect the circuitry underlying associate tolerance using newly developed genetic tools in mice.
Methods: Our central hypothesis is that context-specific representations in the hippocampus predict that opiates are imminent and elicit associative tolerance by activating monosynaptic targets in the amygdala and ventral striatum. To manipulate hippocampal representations, my lab combines optogenetic techniques with transgenic reporter mice to identify the specific neurons and circuits that mediate associative tolerance. This system allows us to tag active neurons during learning and silence or stimulate these same cells during memory testing.
Results: Our data indicate that: 1) Mice exhibit associative opiate tolerance that develops across days and is specific to the context where drugs were encountered. If opiates are administered in a new context, tolerance is eliminated. 2) Context-specific opiate tolerance is significantly reduced when the hippocampus is silenced during training and testing.
Conclusions: Our data suggests that opiate tolerance is regulated by contextual representations that are processed in the hippocampus. These cues elicit a compensatory (or modulatory) response that gradually reduces the analgesic effects of opiate drugs. Future experiments in our lab will identify the circuits that are essential for this effect by stimulating or silencing monosynaptic projections from the hippocampus to targets in the amygdala and ventral striatum. We will also examine the impact of spatial/contextual cues on opiate analgesia in humans and design interventions to reduce the impact of associative tolerance.
Disclosure: Nothing to Disclose.

53.3 Developing Prophylactics Against Stress-Induced Depression: Ketamine Modifies Memory Traces in the Ventral Hippocampus

Christine Ann Denny

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Background: Stress exposure is a major risk factor for mood disorders, such as major depressive disorder (MDD) and post-traumatic stress disorder (PTSD). However, some individuals can successfully adapt to stress and do not develop mood disorders. This ability is known as stress resilience. We previously reported that a single injection of ketamine, an NMDA receptor antagonist, prior to stress protects against the development of depressive-like behavior and attenuates learned fear in mice. However, the cellular and molecular pathways underlying ketamine-induced stress resilience are still largely unknown.
Methods: Here, we will discuss ongoing work to identify the mechanisms mediating prophylactic ketamine-induced stress resilience. We utilize a combination of behavioral paradigms, drug development, viral strategies, and the ArcCreERT2 mice, a line that allows for the indelible labeling of neural ensembles representing a single behavioral experience.

Results: Prophylactic ketamine protected against the development of stress-induced depressive-like behavior and attenuated fear responses. Prophylactic ketamine administration increased deltaFosB expression in a number of brain regions to include the ventral hippocampus (HPC). In a second set of experiments, mice were stereotaxically injected into ventral CA3 (vCA3) with viral vectors in order to upregulate or downregulate deltaFosB expression before prophylactic ketamine administration. Inhibition of deltaFosB only in vCA3 prevented ketamine's prophylactic effect on fear expression. Current studies are focused on identifying and optogentically manipulating memory traces following prophylactic ketamine administration.
Conclusions: Overall, these data indicate that prophylactic ketamine may induce protective effects by altering aversive memories, specifically in the ventral HPC. Understanding how prophylactic ketamine may prevent stress-induced depressive-like behavior can elucidate both the pathophysiology of depression and provide insights into potential new treatment targets.
Disclosure: Nothing to Disclose.

53.4 Mis-Linking Memories: Implications for Psychiatric Disorders

Sheena Josselyn

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Background: Memories are thought to be sparsely encoded in ensembles of neurons (an 'engram'). Although computational theories help explain the sparseness of an engram, why one neuron (rather than a neighbor) is recruited (or allocated) to an engram was largely unknown until my lab and others examined mechanisms underlying neuronal allocation to an engram. Using a variety of techniques, we found that in the lateral nucleus of amygdala (LA) in mice, eligible principle (excitatory) neurons compete against one another for allocation to an engram supporting a conditioned fear memory and that the "winning" neurons are those with increased excitability.
What is the overall function of neuronal competition for allocation to an engram? We recently found that allocated neurons remain more excitable for a time (~6 h) after the event. If a related event occurs within this time window, then an overlapping population of neurons is allocated to the first and second memory. This co-allocation to overlapping neuronal populations links the two related memories. After the 6 h time window, the allocated neurons are no longer more excitable than their neighbors, and a novel population "wins" the competition for allocation to a second memory. Therefore, two events occurring > 6 h apart engage non-overlapping neurons and the memories are kept separate. Here we examined the mechanism mediating memory linking or separating, specifically the role of "communication from winning neurons".
Methods: We used a viral vector that allowed us to both "pick the winners" of neuronal competition by increasing their excitability and silence these same neurons subsequently. We developed a herpes simplex viral vector expressing both an excitatory (ChR2) and inhibitory (eNpHR3.0) opsin (HSV-NpACY). When injected in LA HSV-NpACY infects a random population of ~10% of principle neurons. Activating ChR2 before auditory fear conditioning allowed us to allocate these neurons to the engram supporting that fear memory (as evidenced by a decrease in freezing when the inhibitory opsin was activated during the memory test). To prevent neurotransmitter release from "winning" neurons, we constructed a vector that also expressed light chain of tetanus toxin (TeTx), which cleaves synaptobrevin and prevents neurotransmitter release. We examined the effects of blocking neurotransmission from "winning" neurons on subsequent memory expression and co-allocation of two memories.
Results: As before, optogentically activating principal neurons expressing NpACY (without Tx) before training allocated these neurons to the engram. In contrast, optogenetically activating neurons expressing NpACY-Tx before fear conditioning was unable to induce normal allocation, as these mice froze even when infected neurons were optogentically silenced. These findings suggest that inhibiting the neurotransmitter release from "winning" neurons disrupted normal neuronal competition and allowed another ensemble of neurons to "win" the competition and be allocated to the memory trace. Results using nuclear-localized mRNA levels of the neuronal activity marker Arc support this. Moreover, preventing neurotransmission of "winning" neurons also prevented co-allocation of two events that occur closely together in time. These memories were kept separate as if unrelated.
Conclusions: Here we examined the neurobiological mechanism underlying neuronal allocation and memory linking. Our findings suggest that co-allocation is an important process to link related memories and that this process is impaired when the competitive process is disrupted. Linking related memories may allow us to acquire knowledge of the world, rather than remembering a string of particular instances. Our findings are important as disruption of allocation processes, including mis-allocation, may underlie several brain disorders, including PTSD, depression and schizophrenia.
Disclosure: Nothing to Disclose.

Mini Panel
54. Complex Effects of Alcohol on Brain Immune Function

54.1 Primed for Activation: Complex Effects of Binge-Like Alcohol Exposure on Microglia in a Rat Model of an Alcohol Use Disorder

Kimberly Nixon

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Background: Innate immune system activation may contribute to the development of alcohol use disorders (AUDs). A hallmark of innate immune activation is microglia activation, the process by which microglia alter their morphology, expression of intracellular and surface antigens and therefore function. Microglia can have different

phenotypes that coincide with their varying roles in cytotoxicity (pro-inflammatory) to neuroprotection (anti-inflammatory). Our recent work has shown complex effects of alcohol: 1) microglia show a dystrophic morphology concurrent with a reduced number of microglia with 4- day binge exposure and 2) that a low-level and/or anti-inflammatory activation of microglia persists for months following this damaging 4- day binge alcohol exposure. Such long-term, low-levels of activation predict that microglia may become primed, i.e. hyper-sensitive to subsequent insult. Data from a single, 4- day binge shows subtle microglia morphology changes (ramified, some dystrophic), surface marker upregulation (e.g. CD11b), a decrease in microglia number, then proliferation and an absence of pro-inflammatory cytokine expression – all suggestive of microglia priming – while a double binge exposure produces evidence of a stronger pro-inflammatory activation. However, whether microglia are truly over-responsive to insult has never been tested. Therefore, we examined whether microglia are primed after merely 2-days of binge ethanol exposure.

Methods: Young, male rats were gavaged every 8 h for 2 days with ethanol or isocaloric control diet (Majchrowicz model) for a mean dose of ethanol at 12.7 g/kg/day. Rats were sacrificed 2 days after alcohol exposure and microglia were isolated from entorhinal cortices and hippocampus by Percoll density gradient centrifugation. Cells were labeled with microglia surface antigens and analyzed by flow cytometry. Some isolated cells were cultured in DMEM with 10% fetal bovine serum and exposed to LPS (0, 1, 10 ng/ml) for 24 h. Cell culture supernatant were collected for TNFa ELISA.

Results: Two days after the last dose of alcohol, a highly-enriched population of brain macrophages/microglia ($>95\%$ pure) were isolated, evidenced by expression of the macrophage/microglia antigen, CD11b. Ethanol treated animals showed increased expression of CD11b in both the hippocampus ($p<0.05$) and entorhinal cortex ($p=0.07$) and increased MHC-II in entorhinal cortex ($p<0.05$) only. Cell culture supernatant for isolated microglia showed that LPS resulted a dramatic concentration-dependent increase of TNFa production in microglia isolated from alcohol exposed rats as compared to microglia isolated from control rats.

Conclusions: These data support that merely 2-days of binge-like alcohol exposure primes microglia as demonstrated by the striking increase in proinflammatory cytokine, TNFa, expression in microglia isolated from ethanol-exposed rats as compared to controls. These data directly support that the initial hit of damaging, high peak blood ethanol concentrations prime microglia to result in the enhanced pro-inflammatory response upon subsequent insult, as reported in the double 4- day binge. Furthermore, HMGB1, a factor identified as the priming agent in other models, is also increased in binge-like alcohol exposure. Thus, the expression of various factors associated with immune system activation support that microglia are primed to over-respond with subsequent insults. Intriguingly, priming occurs simultaneously to the observation that some microglia demonstrate a dystrophic morphology and microglia number is decreased. In sum, the effect of alcohol on microglia is complex, characterized by loss though activation of the remaining cells.

Disclosure: Nothing to Disclose.

54.2 Imaging the 18-KDa Translocator Protein, a Marker of Neuroinflammation, in Alcohol Dependence: Findings and Interpretations

Ansel Hillmer

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Background: Chronic alcohol use is thought to disturb brain homeostasis by influencing microglial function. To examine microglial activation in alcohol dependence, positron emission tomography (PET) imaging of the 18-kDa translocator protein (TSPO), a PET imaging marker of neuroinflammation, was used to compare TSPO levels in 15 healthy controls and 15 alcohol-dependent subjects with the radioligand [11C]PBR28. In a subset of subjects, the response of peripheral monocytes to an immune challenge was assessed. Finally, preliminary TSPO imaging studies validating a paradigm of microglia depletion are presented to aid interpretation of the clinical imaging findings.

Methods: Alcohol-dependent subjects were imaged with [11C]PBR28 PET after 1-4 days ($n=14$) or 24 days ($n=1$) of alcohol abstinence. TSPO levels were quantified by estimating total distribution volumes (VT) using multilinear analysis with arterial blood sampling to measure the parent [11C]PBR28 input function. Peripheral immune response was assessed by culturing monocytes extracted from venous blood samples both with and without lipopolysaccharide (LPS). Monocyte response was quantified by measuring the fold-change of cytokine levels in LPS-stimulated cultures relative to saline cultures. Finally, a nonhuman primate was imaged with [11C]PBR28 PET at baseline and after 8 days of exposure to an inhibitor of colony stimulating factor 1 (CSF1), a drug class known to deplete brain microglia levels.

Results: Alcohol dependent subjects exhibited significantly lower [11C]PBR28 levels than healthy controls ($p=0.034$). On average, TSPO levels were 10% lower in alcohol dependent subjects compared to healthy controls. Exploratory analyses suggested a negative association of TSPO levels in hippocampus and striatum with alcohol dependence severity ($p<0.035$). In alcohol dependent subjects, peripheral monocyte response to immune stimulus was lower for the pro-inflammatory cytokines interleukin-6 and interleukin-8 compared to healthy controls. Finally, in a preclinical model of microglia depletion, chronic CSF1 inhibition reduced [11C]PBR28 VT levels throughout the brain by $46\pm3\%$ from baseline levels.

Conclusions: Our imaging data indicate lower TSPO levels throughout the brain and a blunted peripheral pro-inflammatory response in alcohol dependent subjects compared to healthy controls. A preliminary imaging paradigm of microglial depletion showed significant reductions of [11C]PBR28 VT throughout the brain, suggesting that reductions in microglia number could contribute to lower TSPO levels observed with [11C]PBR28 PET. Taken together, this work suggests altered microglia homeostasis in alcohol dependence, implying a potential role for pharmaceuticals tuning the neuroimmune system as therapeutics for alcohol dependence.

Disclosure: Nothing to Disclose.

54.3 Alcohol and the Brain Neuroimmune Transcriptome

Robert Adron Harris

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Background: There is emerging evidence that chronic, excessive, alcohol consumption activates neuroimmune signaling in brain but many key points remain to be defined. We used transcriptome profiling to provide an unbiased measure of changes in gene expression found in human alcoholics and several mouse models of alcohol exposure to determine commonalities and differences about the species. We also asked if the transcriptome changes in astrocytes and microglia, key cells for neuroimmune responses, are missed in the total tissue measures used in all published transcriptome studies of alcohol action on brain in vivo.
Methods: We used RNAseq to profile brain gene expression brain regions from mouse models of alcohol consumption (voluntary drinking) and dependence (repeated vapor) and from human alcoholics. In the mouse models, we profiled isolated astrocytes and microglia as well as total tissue from prefrontal cortex. For human alcoholics, we profiled central nucleus of amygdala (CNA), basolateral amygdala (BLA), the nucleus accumbens (NAC) and superior prefrontal cortex (SFC).
Results: In humans, we identified genes that were differentially expressed between alcoholic and control samples and these included genes related to innate immune response such proinflammatory cytokines (IL18BP, ILF3, TNFAIP2). Functional units enriched among these novel differentially expressed isoforms included the immune pathways such as the toll-like receptor pathway and inflammation related terms such as MAPKK binding and negative regulation of interferon. The mouse models also showed changes in expression of neuroimmune genes. Of particular interest is the increased expression of brain TSPO in human alcoholics and after vapor exposure in mice (astrocytes and microglia). Note that this target was measured in PET studies that will be presented by other speakers.
Conclusions: Chronic alcohol intake, ranging from human alcohol is voluntary drinking in mice, increases expression of proinflammatory genes in brain. The specific gene changes are both partially overlapping and partially distinct for the different species and models. The use of isolated astrocytes and microglia revealed many changes in gene expression in response to alcohol treatment that are not detected in total tissue, demonstrating the importance of cellular analyses.
Disclosure: Nothing to Disclose.

Panel
55. Probing the Uncertain Brain

55.1 Network Composition of the BNST and CeA in Monkeys

Julie Fudge

University of Rochester Medical Center, Rochester, New York, United States

Background: A dominant hypothesis in rodent work is that the two poles of the extended amygdala (EA) are involved in related, but different aspects of fear learning and expression. The BNST is associated with anxiety-like responses to unconditioned stimuli and contextual cues, whereas the CeA is more associated with consolidation and expression of classic Pavlovian fear learning. This dissociation is now the basis of many translational studies of the expression of fear and anxiety in the human. However, the lack of nonhuman primate studies makes interpretation of human neuroimaging studies difficult. Additionally, while the functional anatomy of EA circuits in autonomic and 'flight/fight' responses is well-established, there is little understanding of how the EA can influence more 'habitual' behaviors through basal ganglia circuits, particularly in higher species.
Methods: We examined inputs to the EA in the monkey, using multiple injections of retrograde and bidirectional tracers into the BNST and CeA. Retrograde data was subsequently confirmed with additional anterograde tracing studies. In parallel studies, we also examined the efferent paths of the BNST-CeA, focusing on intrinsic connections, and EA communication with dopamine (DA)-striatal pathways.
Results: The amygdala, midline thalamus, entorhinal cortex, agranular insula, and area 25 of the prefrontal cortex (PFC) project to the EA. However, within this complement of inputs, PFC area 25 has relatively more input to the BNST than to CeA. Importantly, the hippocampus was found to have direct inputs only to the BNST, and not the CeA. The CeA was distinguished by its direct inputs from the sensory thalamus (medial geniculate nucleus), which were not found in the BNST. Anterograde data indicate that common afferent inputs to the EA are broad, with labeled overlap the entire BNST-sublenticular extended amygdala (SLEA)-CeA continuum. Intrinsic projections of the CeA-BNST are largely unidirectional with CeA sending strong inputs to BNST, which are not directly returned. The entire EA continuum projects to the midbrain DA neurons, targeting DA subpopulations that project to 'limbic-associative' regions of the central striatum.
Conclusions: The BNST-CeA continuum is modulated by a suite of common inputs, however, within this network, there are several 'discontinuities'. Compared to the CeA, the BNST is uniquely influenced by the hippocampus, and has relatively more input from area 25 of the PFC (which itself receives hippocampal inputs). In contrast, the CeA is uniquely modulated by direct input from the auditory thalamus. Thus, BNST receives more 'hippocampally weighted' information, while the CeA has more direct sensory inputs, lending support to the concept of the BNST's role in responses to uncertain contexts (contextual input from hippocampus) and the CeA's role in immediate fear responses (auditory input from thalamus) in primates. Intrinsic connectivity suggests that immediate fear experience mediated by CeA is transmitted directly to the BNST. The entire EA is positioned to influence DA subpopulations outside the classic 'mesolimbic' path; DA subpopulations receiving EA input project to more 'cognitive' domains of striatum. Thus, uncertain stimuli–including unpredicted contexts and immediate sensory inputs– may shift habitual cognitive/attentional responses through EA-DA-striatal circuits.
Disclosure: Nothing to Disclose.

55.2 Heritable and Environmental Contributions to Extended Amygdala Functional Connectivity in Young Non-Human Primates

Andrew Fox

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

Background: An extreme anxious temperament (AT) early in life is a risk factor for the later development of anxiety and depressive disorders. Inherited and environmental factors each contribute to this early life risk. Our group has extensively validated a nonhuman primate (NHP) model of AT (Kalin and Shelton, 1989), and use it to investigate the neural circuity underlying primate in anxiety. In a series of FDG-PET imaging studies we demonstrated metabolism within the extended amygdala circuit to be positively associated with individual differences in AT (Fox et al., 2015). In separate studies employing functional MRI, we found that intrinsic fluctuations of the blood oxygenation level dependent (BOLD) signal in the Ce is temporally correlated with BOLD fluctuations in the BST of both rhesus monkeys and humans (Oler et al., 2012). Complementary tract-tracing studies demonstrating the relative strength of Ce to BST projections, compared to the reciprocal pathway, suggest Ce-BST rsfMRI connectivity is likely to result specifically from Ce to BST projections (Oler*, Tromp* et al., 2017). Here, we present two studies aimed at better understanding the origins and functional significance of Ce-BST connectivity.
Methods: In 378 young rhesus monkeys (age: mean = 1.84) who were part of a large multi-generational pedigree, we examined the relationship between Ce-BST functional connectivity and AT, as well as the extent to which it is heritable. In a separate study, we examined the consequences of early life adversity on AT and Ce-BST functional connectivity, by examining 25 young rhesus monkeys that were separated from moms and nursery/peer reared due to rejection (age: mean = 1.85; 7F) and 25 matched controls (age: mean = 1.80 years; 7F). Each animal was exposed to the no-eye-contact (NEC) context, in which a human intruder presents his/her facial profile to the animal, ensuring no eye contact. AT was defined as the mean of z-scored increased freezing, decreased 'coo' vocalizations, and increased cortisol levels in response to the NEC context (Fox et al., 2008). On a separate occasion, each animal received a MRI scan to assess "resting" connectivity under anesthesia. All analyses were performed using a chemoarchitectonically defined Ce-seed region (see: Oler et al. 2012).
Results: Results demonstrate robust correlations between the intrinsic fluctuations of Ce and BST ($p < .05$, Bonferroni corrected), and a significant positive correlation between Ce-BST connectivity and AT ($p < .005$, uncorrected). Interestingly, analyses demonstrated modest, but significant ($p < .005$, uncorrected), heritability of Ce-BST functional connectivity.
In our second study, we found no significant differences in AT ($p = 0.22$; though a trend toward increased freezing behavior, $p = 0.075$), but found decreased Ce-BST functional connectivity in animals that were separated from their mothers early in life ($p < .005$, uncorrected; region overlapped with study 1).
Conclusions: Together, these data demonstrate that AT-related Ce-BST functional connectivity is partially heritable. Moreover, we found Ce-BST functional connectivity was susceptible to environmental influences, including nursery/peer rearing. Together these results suggest the importance of coordinated function within the extended amygdala in determining individual differences in AT, and highlight a potential mechanism by which the effects of both genetic factors and early adversity influence the risk to develop psychopathology.
Disclosure: Nothing to Disclose.

55.3 Dissociating Tonic and Phasic Neural Mechanisms of Uncertain Threat in Human Children, Adolescents, and Adults

Leah Somerville

Harvard University, Cambridge, Massachusetts, United States

Background: Uncertainty is a key antecedent of anxiety and biased attunement to uncertainty is implicated in several forms of clinical psychopathology. In my talk, I will present findings aiming to distinguish tonic neural process that serve to maintain vigilance to lengthy, uncertain threat contexts from those neural responses that represent brief, clear instances of threat.
Methods: To do so, we have developed novel neuroimaging tasks, valid in adult samples, that isolate maintenance processes relevant to uncertain threat processing. More recently, our work has applied these approaches to understand the role of individual differences in trait anxiety, and normative changes in developmental stage from childhood to young adulthood (age 8-22), as key moderators to phasic and tonic threat processing.
Results: Using functional brain imaging in young adults, we have demonstrated a functional dissociation between amygdala-centered phasic threat reactivity and BNST-centered tonic threat maintenance processes. While individuals with heightened anxiety demonstrate biased activity and connectivity between amygdala, BNST, and related nodes of affective circuitry, there is a striking consistency over development in amygdala and BNST reactivity.
Conclusions: Generally, these findings suggest an early functional organization of tonic and phasic uncertainty and threat processing. These findings will be considered in relation to emerging theories of emotional development, and more broadly for their contributions to theories of developmental psychopathology.
Disclosure: Nothing to Disclose.

55.4 CeA and BNST Conversations Across the Brain During Threat

Monique Ernst

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

Background: The two main types of defensive responses, fear and anxiety, are thought to recruit distinct, although overlapping, neural networks. However, much of the research probing these neural mechanisms come from animal models. Less is known in humans, particularly regarding how these circuitries differ from one another,

and how they are uniquely modulated. Fear has typically been associated with the central amygdala (CeA) and its neural circuitry, and anxiety with the bed nucleus of the stria terminalis (BNST) and its network. Both the CeA and BNST are small sub-cortical structures that are not easily amenable to standard 3T-fMRI. With the emergence of ultra-high field 7T-fMRI, this limitation can be addressed. Here, we use 7T-fMRI to compare the iFC of the BNST to the central nucleus of the amygdala (CeA) during threat and safety.
Methods: Four 4-min blocks of "resting state" during either threat-of-shock or safety were collected in 36 healthy volunteers using the 7T Siemens Magnetom MRI with a 32-channel head coil. Subjects looked at a white fixation cross on a black screen. In addition, two colored borders indicated the safety vs. threat condition. High-resolution MPRAGE structural images (0.7mm isotropic) were acquired for hand-tracing of the BNST masks. The CeA mask was from (Tyszka & Pauli, 2016). Briefly, AFNI, FreeSurfer, non-linear normalization into MNI space and noise cleaning with physiological recordings and ANATICOR were employed. Timecourses from ROIs were extracted before smoothing. Two-tailed, paired t-tests (for CeA and BNST, respectively) were conducted. Differences in resting connectivity between threat and safe conditions were analyzed using multiple comparison correction at $p < 0.005$, $k = 54$, determined with 3dClustSim.
Results: Both structures showed generally greater iFC during the safe than threat condition. One exception was the posterolateral thalamus, which was more strongly coupled with the CeA in threat than safe. In contrast, comparison seed-regions such as the OFC, showed generally stronger iFC coupling to a majority of targets during threat than safety. Regionally, threat on BNST-iFC impacted both cortical (PFC, PCC) and subcortical (ventral striatum) regions. Threat on CeA-iFC impacted the vmPFC and thalamus. Notably, vmPFC was a common target to both CeA and BNST.
Conclusions: Weaker connectivity during threat vs. safe could reflect more flexibility and diversity within the BNST circuit, as a way to provide optimal preparedness for a potential threat. Notably, the specific modulation of BNST-NAc with sustained threat suggests a role of the ventral striatum in anxiety-related motivated behavior. The strengthening of CeA-thalamus coupling might reflect the maintenance of salient information originating from the CeA, helping to keep sustained focus on potential threat. Finally, vmPFC is an important overlapping area linked to BNST and CeA activity that may regulate both fear and anxiety.
Disclosure: Nothing to Disclose.

Panel
56. Translation of Clinical Biomarkers of Circuit Hyperexcitability From Mouse Models to Patients With Neurodevelopmental Disorders

56.1 An Optogenetic Assay to Study the Microcircuit Basis of Abnormal Evoked and Resting State EEG in Fragile X Syndrome in Acute Neocortical Brain Slices From Mice

Kimberly Huber

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

Background: Humans with Fragile X Syndrome (FXS) experience sensory hypersensitivity and evidence suggests this is mediated by hyperexcitability of sensory circuits. In particular, EEG and MEG studies in individuals with FXS find an increased magnitude of auditory event-related potentials (ERPs) to repeated stimuli as well as enhanced resting state gamma band power compared to healthy controls (Ethridge et al., 2016; Wang et al., 2017). Remarkably, enhanced auditory ERPs and resting state gamma band power are also observed in the mouse model of FXS – the Fmr1 knock-out (KO) (Lovelace et al., 2016). The conservation of sensory circuit dysfunction across species offers an opportunity to determine the circuit basis of cortical circuit dysfunction in FXS and develop therapeutics to correct it.
Methods: With this goal in mind, we developed an optogenetic assay to mimic sensory-evoked and resting state activity in acute slices of auditory neocortex in Fmr1 KO mice and probe the excitability of local microcircuits.
Results: Extracellular recordings revealed increased firing of local circuits within layer (L) 2/3 in response to a brief, localized (10ms; 350 μm) blue light stimulation. In contrast, responses in L5 were unaffected in Fmr1 KO. In an attempt to model resting state EEG alterations in Fmr1 KO slices, we measured local L2/3 and L5 circuits in response to a low intensity and sustained (0.5 sec) blue light. Circuit activity was more sustained in L2/3 Fmr1 KO local circuits, and displayed enhanced power at higher frequencies (12-45 Hz); likely relevant to the gamma band power in the EEG. Like we observed with brief stimulation, L5 local circuits were not hyperexcitable in response to sustained blue light and did not exhibit enhanced gamma band power.
Conclusions: These results suggest hyperexcitability specifically in L2/3 contributes to the enhanced auditory ERP and resting state gamma power in FXS. Future experiments are aimed at testing the pharmacology and circuit basis of L2/3 hyperexcitability.
Disclosure: Nothing to Disclose.

56.2 Toward Translational EEG Biomarkers in Fragile X Syndrome

Devin Binder

University of California, Riverside, California, United States

Background: Sensory processing deficits are common in autism spectrum disorders, but the underlying mechanisms are unclear. Fragile X Syndrome (FXS) is a leading genetic cause of
intellectual disability and autism. Electrophysiological responses in humans with FXS show reduced habituation with sound repetition and this deficit may underlie auditory hypersensitivity in FXS.
Methods: Our previous studies in Fmr1 knockout (KO) mice revealed an unusually long state of increased sound-driven excitability in auditory cortical neurons suggesting that cortical responses to repeated sounds may exhibit abnormal habituation as in humans with FXS. We tested this prediction by comparing cortical event related potentials (ERP) recorded from wildtype (WT) and Fmr1 KO mice. We

found a repetition-rate dependent reduction in habituation of N1 amplitude in Fmr1 KO mice, suggesting ERP habituation as a translationally-relevant preclinical EEG biomarker of auditory processing deficits in FXS.
Results: In subsequent studies, we have characterized EEG parameters in WT and Fmr1 KO mice in greater detail. First, we have compared baseline EEG in WT and Fmr1 KO mice and have found a striking increase in baseline gamma band power in Fmr1 KO mice. This parallels recently published data from humans with FXS (Ethridge et al., 2017, in press). Second, we have further endeavored to characterize event-related EEG differences between WT and Fmr1 KO mice using various auditory stimuli, including broadband noise and chirp stimuli. These results have revealed consistent deficits in gamma-band phase locking to the chirp stimulus in Fmr1 KO mice, as seen recently in FXS humans (Ethridge et al., 2017, in press). Third, we are in the process of developing and characterizing a novel multielectrode array (MEA) for use in freely behaving mice in vivo in order to derive baseline, stimulus-evoked EEG amplitudes and phase information, and perform phase-locking and coherence analyses more analogous to human EEG studies.
Conclusions: These studies aim to develop and validate new and robust EEG-based biomarkers for pharmacological testing in mouse models of FXS with translational relevance to humans.
Disclosure: Nothing to Disclose.

56.3 Electrophysiological Evidence for Neocortical Hyperexcitability and Functional Connectivity Alterations in Patients With Fragile X Syndrome

John Sweeney

University of Cincinnati, Cincinnati, Ohio, United States

Background: Fragile X Syndrome (FXS) is among the most common single gene causes of autism and intellectual disability. Preclinical evidence demonstrates hyperexcitability of neocortical circuits. Limited clinical evidence demonstrates parallel abnormalities in FXS patients or their clinical relevance.
Methods: We recruited over 40 FXS patients and matched healthy controls. Resting EEG and both novel and traditional auditory ERP paradigms were administered. Social, cognitive and sensory hyperexcitability were evaluated by parents.
Results: Resting EEG studies revealed enhanced resting gamma frequency power that spread coherently several centimeters across neocortex. Synchronized gamma band power to gamma frequency sensory input was reduced. Alpha power was reduced and theta power increased at rest, and alpha-gamma amplitude coupling was reduced while theta-gamma amplitude coupling was increased. Reduced sensory habituation was observed, also indicating altered excitatory/inhibitory balance in neocortex. EEG/ERP alterations were related to sensory and social problems. Marked reductions of coherent interactions between speech and auditory cortices were seen during speech production.
Conclusions: Our findings indicate robust and clinically relevant neocortical hyperexcitability in FXS patients. These findings are consistent with preclinical observations, and are informative about functional brain alterations in FXS. Our experimental approaches may be relevant for the large subgroup of ASD patients and for other disorders demonstrating sensory and EEG hyperresponsivity, and have promise as a translational biomarker strategy for drug development targeting an altered balance of Excitatory: Inhibitory cortical physiology.
Disclosure: Part 1: Takeda, Advisory Board.

56.4 Human EEG Biomarker Use in Fragile X Syndrome and Related Disorders Clinical Trials

Craig Erickson

Cincinnati Children's Hospital Medical Center, Cincinnati, Ohio, United States

Background: Phenotypic heterogeneity has challenged translational treatment development in fragile X syndrome (FXS) and related autism spectrum disorders (ASDs). Use of novel EEG paradigms in FXS and related disorders presents an opportunity to test the pharmacodynamic impact of targeted drug treatment in neurodevelopmental disorders to parse disorder heterogeneity and begin to predict treatment response.
Methods: We are incorporating novel EEG measures of cortical excitability in FXS and related disorders including use of auditory ERP and EEG power analysis into first in disease proof of concept clinical trials. This work includes EEG biomarker use in ongoing and recently completed controlled trials of acamprosate, ketamine, and the selective GABA(A) alpha 2,3 agonist AZD7325. We are additionally utilizing the same methodology in the context of open-label clinical pharmacotherapy treatment to estimate EEG effect size with various drug therapies. In addition to our EEG read outs, we are also correlating EEG change with treatment with molecular blood assay and eye tracking/pupillometry findings from the same trials.
Results: We have characterized EEG abnormalities in persons with FXS including auditory ERP habituation and gamma band power abnormalities and employed these same paradigms in recent trials in FXS and related disorders. We will have effect size data available regarding specific targeted pharmacotherapies on these EEGs parameters in neurodevelopmental disorders. We will additionally characterize the relationship between EEG biomarker findings and molecular and eye tracking/pupillometry results generated from the same subjects in drug trial settings.
Conclusions: Novel EEG measures hold significant promise in neurodevelopmental disorder drug trials to evaluate drug pharmacodynamic effects using paradigms shown to detect import cortical abnormalities in FXS, a model single gene form of ASD. We believe this work in time will allow enable novel trials in this field to better profile which patient subgroups may best respond to a particular pharmacotherapy.
Disclosure: Part 1: Confluence Pharma, Stock / Equity.

Panel
57. Pragmatic Academic Psychiatry – Closing the Gap Between Research and Practice

57.1 Modulation of Oscillatory Synchrony as a Biomarker for Treatment Outcome in Major Depressive Disorder

Andrew Leuchter

University of California, Los Angeles Laboratory of Brain, Behavior and Pharmacology, Los Angeles, California, United States

Background: Theories of antidepressant treatment mechanism of action traditionally have focused on the level of cell-to-cell interaction and synaptic neurotransmission. Recent evidence suggests that modulation of synchronized electrical activity in neuronal networks is a common effect of antidepressant treatments, including medications as well as neuromodulatory treatments such as repetitive transcranial magnetic stimulation (rTMS). Research suggests that oscillatory synchrony may help mediate neuroplastic changes related to medication and neuromodulatory treatment for Major Depressive Disorder (MDD).
Methods: In a series of experiments, high-density quantitative electroencephalography (qEEG) was used to monitor 240 subjects with MDD undergoing several different treatments: the selective serotonin reuptake inhibitor (SSRI) antidepressant escitalopram; high-frequency (10 Hz) rTMS administered to left dorsolateral prefrontal cortex; intravenous ketamine; and placebo. Shifts in oscillatory synchrony in the delta-theta (2.5–8 Hz), alpha (8–12 Hz), and beta (12–20 Hz) bands were examined before and after treatment, and in the case of rTMS and ketamine, continuously during treatment administration. Changes in functional connectivity among brain regions were examined using phase coherence.
Results: Subjects in all active treatment conditions showed shifts in oscillatory synchrony and functional connectivity that were related to the degree of clinical improvement. In the case of escitalopram, these changes were evident within one week of the start of treatment, whereas in the case of rTMS and ketamine, these changes were seen during acute treatment administration. qEEG changes preceded change in depressive symptoms and were not seen in placebo remitters. Changes consisted of a shift in synchrony from the lower frequency delta-theta to the alpha band, as well as decreased functional connectivity. Shifts were most consistent in the frontopolar region. Biomarker-guided treatment changes were prospectively tested in subgroups of subjects treated with escitalopram or rTMS, and were found to lead to improved outcome.
Conclusions: These findings indicate that shifts in oscillatory synchrony may constitute a specific biomarker for outcome that are common across several active treatment modalities for MDD, but not placebo. The prospective testing of biomarkers suggest that they may be used to guide treatment decisions and enhance outcomes of antidepressant treatment.
Disclosure: Part 1: Ionis Pharmaceuticals, Consultant, NeoSync, Inc., Consultant, Brain Biomarker Analytics, LLC, Board Member, **Part 2:** NeoSync, Inc., Consultant, Brain Biomarker Analytics, LLC, Board Member, **Part 3:** NeoSync, Inc., Consultant, **Part 4:** CHDI Foundation, Grant, Neuronetics, Inc., Grant.

57.2 New Methods for Predictive Research in Psychiatry

Claire Gillan

Trinity College Dublin, Dublin, Ireland

Background: Pharmacological treatment response in psychiatry is highly heterogeneous. Current symptom-based categorical diagnostics do not predict who will respond to treatment; instead clinicians have no choice but to rely on trial-and-error. Finding robust predictors of treatment response in small case-control studies has proven difficult, likely due to profound genetic and environmental complexity, coupled with major questions about taxonomic validity and the use of medications that are themselves heterogeneous in receptor potency and selectivity. To improve the precision of treatment allocation in psychiatry, the field is beginning to recognise the need for large-samples that can support predictive modelling in high dimensional space.
Methods: We tested the viability of a novel Internet-based methodology for predictive treatment research in psychiatry. We conducted a series of cross-sectional studies via the Internet that demonstrate the validity of this approach. Cognitive task and clinical data were collected from ~3500 subjects via the Internet across 5 separate studies, probing aspects of reinforcement learning, executive control and meta-cognition. We compare recruitment rates across traditional studies and Internet-based alternatives.
Results: Our Internet-based approach replicates previous findings in smaller carefully diagnosed patient groups. Sample size gains allowed us to unpack co-morbidity and identify trans-diagnostic psychiatric traits that explain commonalities across disorders in terms of neurocognitive deficit profile. The most modest estimate of recruitment gain for diagnosed patients from moving to a web-based methodology was 12-fold.
Conclusions: This method provides a platform for conducting longitudinal research that is free of geographical restriction and is sufficiently powered for predictive research in mental health that incorporates internal and external model validation. We highlight several innovative approaches to treatment prediction research that are currently underway in our lab using the Internet.
Disclosure: Nothing to Disclose.

57.3 From Electronic Health Records to Treatment Recommendations for Depression

Finale Doshi-Velez

Harvard University, Cambridge, Massachusetts, United States

Background: Major depressive disorder (MDD) has an over 15% lifetime prevalence. The heterogeneity of the disease makes it challenging to match patients to treatments. Over 50% of patients fail their first therapy; 50% of those their second. When each treatment can take 4-6 weeks to evaluate,

these numbers mean that there are a lot of patients waiting a lot of time before they see benefits to their health. Electronic health records can be useful in helping characterize what treatments may be likely to succeed for different patients as they capture the variation in the population being treated.
Methods: We study a cohort of 875,080 encounters from 49,322 patients drawn from two large academic medical centers with at least one ICD9-CM for MDD. Focusing on 9 common antidepressants, each of these patients had an identified successful treatment: a treatment repeated at least 2 times in 12 months with no change. We extracted all diagnoses, labs, and meds (22,000 codewords) and reduced them to 9621 common ones. Next, we applied a novel prediction-constrained topic modeling approach that allowed us simultaneously infer disease subtypes as well as predict what drugs are most likely to be successful for what patients. Specifically, our approach resolves issues with previous approaches that ignore the asymmetry of the problem: we are interested in making treatment recommendations given subtypes, but not subtypes given treatment recommendations.
Results: We find that our approach can make treatment predictions as well as logistic regression or better: our predictions had test-set AUCs in 0.59 to 0.71 while logistic regression had AUCs in 0.54 to 0.64. Clinicians confirmed that the topics found by our approach were more interpretable than from other approaches or the weights of logistic regression (which can be challenging to interpret in the face of co-linearity in high dimensions).
Conclusions: We applied a prediction-constrained topic modeling approach to identify subtypes from electronic health record data of patients with MDD such that the subtypes correlated with treatment predictions. Such interpretability was valuable in identifying situations when the treatment recommendation was made for some face-valid reason, and situations in which the recommendation was made due to a confound; techniques that combine interpretability and predictive quality are an essential tool for gleaning scientific insights from electronic health record data.
Disclosure: Nothing to Disclose.

57.4 Can an Algorithm Select First- and Second-Line Antidepressant Treatments in Primary Care?

Adam Chekroud

Yale University, New Haven, Connecticut, United States

Background: Effectiveness studies suggest that in the first step, antidepressant treatments are approximately 11-30% likely to bring about a full remission, and differences in remission rates between treatments are generally small at the group level. The situation is remarkably similar for patients who attempt a second treatment, with many strategies effective around 25% of the time. This situation complicates the management of mild-moderate depression in primary care settings, contributes both to the financial and medical burden of depression, and often results in an overwhelming caseload in specialty care as patients are referred out by their PCPs.
Methods: This talk draws on data from multiple industry- and NIMH- funded RCTs to develop computational strategies to select medications for individual patients that will maximize their chances of remitting. It will review data suggesting that first- and second-step treatment outcomes are predictable using routine clinical data. Next, the talk will describe how these computational models were then translated from bench to bedside: (1) they were packaged into digital assessment on a mobile device that patients complete in the waiting room, and (2) the results of the assessment are securely transmitted and displayed to the physician to improve the allocation of antidepressant treatments in real-world care settings.
Results: These data suggest that antidepressant outcomes can be predicted accurately in the first and second line of antidepressant treatment, and that a platform for precision mental healthcare can be implemented in a real-world setting.
Conclusions: It is both theoretically and practically possible to use routine clinical data to match patients to antidepressant treatments that are more likely to be effective. This precision medicine approach may offer an opportunity to improve the treatment of mild-moderate depression in Primary Care: both accelerating the recovery process and reducing the rate of referrals to specialty care.
Disclosure: Part 1: Spring Health, Stock / Equity, Spring Health, Employee, **Part 2:** Spring Health, Employee, Spring Health, Stock / Equity, **Part 3:** Spring Health, Employee.

Panel
58. Re-Evaluating TSPO as a Biomarker for Neuroinflammation

58.1 Preclinical Evaluation of Translocator Protein as a Biomarker of Neuroinflammation in Neurodevelopmental and Neurodegenerative Disorders

Tina Notter

University of Zurich, Zurich, Switzerland

Background: Positron emission tomography (PET) imaging with radiotracers that target translocator protein 18 kDa (TSPO) has become a popular approach to assess putative neuroinflammatory processes and associated microglia activation in psychotic illnesses. It remains unclear, however, whether TSPO imaging can accurately capture low-grade inflammatory processes such as those present in schizophrenia and related disorders. Therefore, we evaluated the validity of TSPO as a disease-relevant biomarker of inflammation and microglia activation in preclinical mouse models of neurodevelopmental and neurodegenerative disorders. To further explore the functional involvement TSPO beyond microglia activation, we evaluated its expression upon selective activation of astrocytes and neurons.
Methods: To study TSPO and inflammatory abnormalities in a neurodevelopmental model relevant to schizophrenia, we used an established mouse model of maternal immune activation (MIA) induced by the viral mimetic poly(I:C). We further compared the MIA model with a mouse model of acute neurodegeneration and reactive gliosis, which was

induced by intrahippocampal injection of kainic acid (KA). Finally, TSPO expression in response to selective activation of astrocytes or neurons was studied using in-vivo chemogenetic approaches involving DREADD systems in mice.
Results: Using the MIA-based neurodevelopmental mouse model, we show that schizophrenia-relevant behavioral abnormalities and increased inflammatory cytokine expression are associated with reduced prefrontal TSPO levels. On the other hand, TSPO was markedly upregulated in the KA-based model of acute neurodegeneration and reactive gliosis. In both models, the changes in TSPO levels were not restricted to microglia but emerged in various cell types, including microglia, astrocytes, and vascular endothelial cells. Furthermore, we found an activity-dependent modulation of TSPO expression in astrocytes and neurons upon DREADD stimulation.
Conclusions: Our findings suggest that signs of central low-grade inflammation can occur concomitantly with reduced TSPO expression. Hence, decreased TSPO expression does not (necessarily) suggest reduced inflammation (and vice versa), implying that ongoing inflammatory processes are not always mirrored by increased TSPO signals. Moreover, altered TSPO expression does not reflect selective changes in microglia activity, neither in (infection-mediated) neurodevelopmental pathologies nor in severe neuroinflammatory conditions that are accompanied by marked microgliosis. In fact, astroglial and neuronal activation causes noticeable changes in TSPO expression as well, supporting the notion that TSPO is a relatively unspecific cellular biomarker with poorly defined functions in neuroinflammation.
Disclosure: Nothing to Disclose.

58.2 Evaluation of TSPO Responses to Pro-Inflammatory and Anti-inflammatory Stimuli

Eugenii Rabiner

Imanova Ltd., London, United Kingdom

Background: While the imaging of the 18 kDa Translocator Protein (TSPO, formerly known as the peripheral benzodiazepine receptor – PBR) using a variety of PET probes has been widespread over the last 20 years, we still have a poor understanding of the role the TSPO plays in normal physiology, or the effects of disease and treatment. The TSPO is a mitochondrial outer membrane protein, associated with the voltage dependent anion channel (VDAC), and is known to be upregulated in activated microglia and macrophages. This finding has formed the basis for the use of TSPO positron emission tomography (PET) as a method to image neuroinflammation. However, the selectivity of TSPO for microglia has been questioned, with expression found in endothelial cells and astroglia. In addition, the association of increased TSPO expression with pro-inflammatory responses has not been well established.
Methods: We evaluated the responses of human and rodent myeloid cells to IFN-γ/LPS (a classical activation pro-inflammatory stimulus) and IL4/IL13 (a stimulus of reparative activation), evaluating changes in gene and protein expression, using [3H]PBR28.
Results: Various activation challenges led to differing effects in human and rodent macrophages and microglia, with pro-inflammatory stimulus leading to unchanged or reduced TSPO expression in differing human cell types. The changes in gene and protein expression were concordant in various cell types following pro-inflammatory and reparative activation.
Conclusions: Elevations of TSPO expression seen in PET studies cannot be assumed to reflect increased inflammation, but must be interpreted in conjunction with the species and myeloid cell type examined.
Disclosure: Nothing to Disclose.

58.3 PET Studies of TSPO in Schizophrenia – Understanding Initial Findings

Simon Cervenka

Karolinska Institutet, Stockholm, Sweden

Background: Accumulating evidence suggests that the immune system may have a role in the disease mechanism in schizophrenia. Using Positron emission tomography (PET) and radioligands binding to the translocator protein (TSPO), which is expressed on glial cells, brain immune function can be assessed in vivo. Thus far, PET TSPO studies in schizophrenia have been inconclusive. However, sample sizes have been limited and TSPO were in most cases not compared to other markers of immune activation.
Methods: We recently reported reductions of brain TSPO in a cohort of neuroleptic-naïve first episode psychosis patients vs healthy control subjects. From these individuals, we will present novel data on CSF and plasma markers in a subgroup of patients and controls. Here, we focus on chemochines due to their role in immune cell recruitment and pro-inflammatory signaling. YKL-40 and MCP-1 were measured using electrochemiluminescence assay and were analyzed in relation to brain TSPO levels in this unique neuroleptic naïve cohort. Furthermore, we have conducted an individual participant data metaanalysis on TSPO in schizophrenia, including all studies that 1) used a second-generation TSPO radioligand, 2) reported brain distribution volume (VT) in patients with psychosis as compared to healthy controls, and 3) reported TSPO genotype. A fixed-effects model was selected and Bayes factors were applied to examine the relative support for higher, lower or no-change of TSPO levels in patients as compared to healthy controls.
Results: Plasma YKL-40 levels were increased in FEP patients compared to healthy controls ($p = 0.034$), after correcting for age. This difference was not found when comparing CSF YKL-40 levels. Plasma or CSF levels of MCP-1 did not differ between patients or controls. In patients, a significant positive correlation between YKL-40-levels in CSF and VT was observed ($r = 0.78$; $p = 0.021$; correcting for genotype, gender and age), whereas in controls these variables were negatively correlated ($r = -0.88$; $p = 0.009$). In the meta-analysis, strong support was found for decreased patient TSPO levels (VT) relative to no-change or increase. No support was found for an effect of anti-psychotic medication on standardized VT values.
Conclusions: These preliminary data on CSF chemochines in relation to TSPO suggest a dysregulated immune cell response in schizophrenia. The observed decrease in TSPO in the meta-analysis could indicate a compensatory

mechanism, or altered function of immune cells in psychosis patients such as abnormal energy utilization. The findings will also be discussed in relation to recent data showing reductions in brain TSPO in patients after abdominal surgery, where the PET data was mirrored by decreased response of immune cells to in vitro stimulation. Based on the data presented, the utility of TPSO PET for measuring immune activation in schizophrenia will be critically evaluated.
Disclosure: Part 1: AstraZeneca, Grant, SOBI, Employee, (Spouse) **Part 2:** AstraZeneca, Grant, SOBI, Employee, (Spouse), **Part 3:** SOBI, Employee, (Spouse), **Part 4:** AstraZeneca, Grant.

58.4 Combining TSPO Imaging and Assay of Systemic Cytokines to Probe the Pathological Neuroimmune Response After Repetitive TBI

Jennifer Coughlin

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Background: Positron emission tomography (PET)-based neuroimaging has identified elevated levels of the translocator protein 18 kDa (TSPO), a putative biomarker of inflammation, in several neuropathologies including sports-related, repetitive traumatic brain injury (TBI) among National Football League (NFL) players. However, TSPO is increased in the brain during states of brain injury or repair, and so this isolated biomarker is limited in its ability to identify the hypothesized, pathological imbalance between pro-inflammatory and reparative processes after repeated TBI. Furthermore, TSPO levels in the brain may be unchanged even in patients who have systemically elevated pro-inflammatory cytokines. Based on this background, we hypothesize that the presence of peripheral pro-inflammatory cytokine levels combined with high TSPO in the brain will best mark the immunopathology linked to neurodegeneration, discriminating it from reparative processes, following repeated TBI.
Methods: We collected [11C]DPA-713 PET imaging data from young active or recently retired NFL players and matched healthy controls. Each player provided a peripheral blood sample to test for peripheral pro-inflammatory cytokines using a multiplex immunoassay system. Since [11C]DPA-713 total distribution volume (VT), the primary binding outcome, was found to be higher in several brain regions of the players compared to the controls, we assessed the player population for those with a high versus low plasma pro-inflammatory profile (marked by levels of tumor necrosis factor α, interferon γ, interleukin 6). Among those players with a high versus low plasma pro-inflammatory profile, we chose one representative case from each group to return for repeat PET-based neuroimaging using [11C]DPA-713 at two year follow-up. Each of these two cases also underwent [18F]T807 PET to test for neuropathological tau burden at this follow-up visit.
Results: From a population of young NFL players with previously reported higher regional [11C]DPA-713 VT compared to controls, we identified eight players with high plasma pro-inflammatory profile and seven players with low/mixed plasma pro-inflammatory profile. There was $<6\%$ mean change in [11C]DPA-713 binding outcomes in subcortical and cortical brain regions between the baseline and two year follow-up scans among the two representative players. The player with high plasma pro-inflammatory profile at baseline had high tau burden (defined as [18F]T807 standardized uptake value ratio Z score >2.5) in the corpus callosum, cingulate cortex and parietal cortex at two year follow-up, while the player with low baseline plasma pro-inflammatory profile was without elevated tau in the brain.
Conclusions: High TSPO levels in the brains of NFL players may reflect pro-inflammatory or reparative processes that may be best discriminated on an individual level through complementary assay of peripheral cytokines. The longitudinal data suggesting persistently high TSPO levels among two NFL players and a link between high [11C]DPA-713 binding, high plasma pro-inflammatory profile, and later tau burden warrants further investigation.
Disclosure: Nothing to Disclose.

Panel
59. Translational, Multidisciplinary, and Integrative Research on Mechanisms That Mediate Excessive Alcohol Consumption

59.1 Novel Neuroimmune Gene Regulation in Human Alcoholics

Dayne Mayfield

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Background: The genetic liability for alcohol dependence is 50-60%, but genetic linkage is likely more complicated than the presence or absence of individual annotated genes. We generated a large-scale RNA-Seq dataset from human postmortem brain to investigate the transcriptional landscape associated with excessive alcohol consumption.
Methods: The dataset included 4 regions: central nucleus of amygdala, basolateral amygdala, nucleus accumbens and superior prefrontal cortex. We identified novel gene isoforms by assembling transcripts present in alcoholic versus matched control samples in a genome-guided fashion.
Results: In alcoholic samples, 8% of the transcripts were annotated, but a remarkable 80% were mapped to unknown/intergenic regions and 12% flanked annotated genes with different gene intervals. The transcripts associated with intergenic regions were of interest because of the potential relevance of non-coding RNAs in regulating gene expression. We identified 15% of these transcripts as candidates for non-coding RNAs. We also found differentially expressed genes between alcoholic and control samples that contained potentially novel isoforms. These included genes related to innate immune responses such as proinflammatory cytokines (IL18BP, ILF3, TNFAIP2). Functional units that were enriched among the differentially expressed isoforms included immune pathways (toll-like receptor pathway) and inflammatory-related processes (MAPK binding and negative regulation of interferon I). System biology approaches were then used to identify gene networks specific to these isoforms.

Conclusions: Overall, we identified splicing signatures that could drive the generation of novel transcript isoforms. This approach does not rely purely on annotated genes and provides an overview of all the transcriptional and regulatory elements involved in chronic alcohol abuse.
Disclosure: Nothing to Disclose.

59.2 Targeting Binge Drinking: A Translational Approach

Angela Ozburn

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Background: The FDA has approved only three drugs for treatment of alcoholism – disulfiram, naltrexone, and acamprosate. One of the impediments to discovery of new therapeutic compounds has been that existing animal models used for screening typically do not voluntarily drink to intoxication. However, recent efforts have led to animal models of drinking to intoxication (i.e. Drinking in the Dark, DID). Further work has been carried out to develop selectively bred mice for High Drinking in the Dark (HDID-1 and HDID-2), by mating individuals reaching the highest blood alcohol levels (BALs) after a second daily DID session. HDID mice achieve BALs between 170 and 150 mg% after 32 (HDID-1) and 25 (HDID-2) selected generations, respectively; this is approximately twice the 80 mg% level defined by the NIAAA as binge drinking. Binge drinking is a strong predictor of alcohol use disorder diagnosis and has deleterious health consequences, particularly in US combat veteran populations.
Methods: We used regional brain tissue from HDID-1 male mice and compared gene expression profiles between HDID mice and their non-selected HS/Npt control line with data from the Library of Integrated Cellular Signatures (LINCS), which comprises gene expression response profiles for $>19{,}000$ compounds in multiple cell lines, including responses to many FDA-approved compounds. We generated a list of genes that have increased or decreased expression in alcohol-naïve HDID mice vs HS. We then used this input to query LINCS and identify compounds with signatures that have a strong connectivity to the disease signature. We prioritized compounds and tested the effects of selected compounds on binge-like drinking in HDID-1 mice. We hypothesized that these novel drugs would normalize HDID-1 expression differences from HS and would successfully predict drug efficacy to reduce binge-like drinking.
Results: Testing has been carried out successfully in separate groups of male and female HDID-1 mice for two compounds, pergolide (dopamine and serotonin receptor agonist) and terreic acid (Bruton's tyrosine kinase inhibitor). Both drugs significantly reduced binge-like alcohol drinking and associated blood alcohol levels.
Conclusions: Our analysis and testing approach successfully identified novel compounds that drastically reduce alcohol intake and blood alcohol levels in HDID mice. Therefore, we conclude that other novel compounds suggested by our investigation could have therapeutic potential for the treatment of Alcohol Use Disorders.
Disclosure: Nothing to Disclose.

59.3 Genetic, Functional, and Behavioral Evidence for Prefrontal Cortex Fast-Spiking Interneuron Regulation of Excessive Alcohol Drinking

Patrick Mulholland

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Background: Alcohol use disorder (AUD) is a chronic relapsing disease characterized by cognitive impairments and high risk for relapse. The medial prefrontal cortex (mPFC) is a key structure involved in behavioral flexibility, and its normal role in imposing inhibitory control over reward-motivated behaviors is disrupted leading to enhanced relapse vulnerability. Despite intense investigation of the mPFC, the neural mechanisms that underpin heavy drinking remain poorly understood. Parvalbumin-positive fast-spiking interneurons (FSIs) are of interest for their role in orchestrating cognitive behaviors and decisions related to natural reward seeking. KV3 channels are encoded by Kcnc genes and are highly enriched in PV+FSIs. We previously reported changes in mPFC Kcnc expression in chronically exposed mice and correlations between Kcnc and alcohol drinking. Here, we performed additional genetic analyses coupled with functional and behavioral validation studies to elucidate the role of mPFC FSIs and KV3 channels in regulating heavy drinking.
Methods: To address the gap in our knowledge on FSI and KV3 channel control of alcohol drinking, we used a multifaceted approach to test the overarching hypothesis that alcohol dependence reduces FSIs control of drinking through neuroadaptations in KV3 channels. First, we determined if alcohol dependence alters expression of interneuron-specific genes in the mPFC using bioinformatics tools. Using whole-cell patch-clamp recordings, we then measured burst firing in mPFC FSIs from control and alcohol dependent mice. Lastly, we determined the efficacy of a novel small molecule (AUT6) that selectively activates KV3 channels to reduce alcohol intake in two mouse models of binge-like and heavy alcohol drinking.
Results: A number of genes selectively expressed in cortical interneurons (e.g., Kcnc1) were altered by alcohol dependence in BXD strains and B6 mice, and functional enrichment analyses showed that interneurons genes were over-represented when compared with all alcohol-sensitive genes, suggesting that alcohol dependence may disrupt cortical interneuron function. Consistent with the genetic findings, alcohol dependence significantly reduced the length of spontaneous burst firing and interburst intervals in mPFC FSIs. Interestingly, bath application of AUT6 restored burst firing to levels observed in alcohol-naïve mice. To validate the genetic and functional results, AUT6 was administered to mice that were drinking in two standard models of heavy alcohol drinking. In both models, AUT6 significantly reduced alcohol intake in non-dependent and dependent B6 mice.
Conclusions: The major finding from this integrative study provides evidence linking mPFC FSIs and KV3 channels with voluntary drinking and alcohol dependence. We identified cortical adaptations in interneuron-specific genes in alcohol dependent mice. At the functional level, alcohol

dependence altered burst firing properties of cortical FSIs. Pharmacological validation studies revealed that positive modulation of a novel small molecule activator of KV3 channels restored alcohol dependence-induced adaptations in mPFC FSI burst firing and reduced binge-like and heavy alcohol drinking. In summary, these multifaceted findings suggest that KV3/Kcnc channels are a possible pharmacogenetic target to restore interneuron control of goal-directed behaviors and reduce excessive drinking.
Disclosure: Nothing to Disclose.

59.4 Ethanol Self-Administration, Cognitive Flexibility and Striatal Function in Rhesus Monkeys

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Background: Attentional set-shifting ability is an executive function underlying cognitive flexibility in humans and animals. The dorsal lateral striatum (putamen) is believed to underlie habitual behavior and is implicated in impaired cognitive flexibility that accompanies excessive alcohol drinking. This presentation will integrate performance on a set-shifting task with chronic ethanol self-administration and resting state functional connectivity magnetic resonance imaging (rs fc(MRI)) of the putamen (seed) to prefrontal cortical regions.
Methods: Macaque monkeys ($n = 24$) were housed in the same room but in individual cages with drinking panels that incorporate touch screens. Baseline (pre-ethanol) corticostriatal rs fcMRI was acquired under 1% isoflurane in all monkeys. One group ($n = 12$) was also trained in a self-paced set-shifting task that encompassed 8 levels of shape/color discriminations (4 new sets, each followed by reversal). Set-shifting performance was evaluated by the number of error/trials, the level reached by the end of the session and time to finish all 8 levels. All monkeys were induced to self-administer ethanol (4% w/v) using a schedule-induced polydipsia (SIP) procedure comprised of 4 blocks of 30 consecutive sessions where water, 0.5 g/kg ethanol, 1.0 g/kg ethanol and 1.5 g/kg ethanol, respectively, was consumed. After the 30th session of 1.5 g/kg ethanol induction, the availability of ethanol was then increased to 22 hr/day, 7 days/week (open-access). Set shifting was reassessed during the 1.5 g/kg ethanol induction phase and again after 6 months of ethanol open-access. Corticostriatal fcMRI was reacquired after 6 and12 months of 22 hr/day ethanol self-administration.
Results: Ethanol and control monkeys performed similarly on set-shifting during the pre-ethanol phase. When induced to drink 1.5 g/kg/day, the average blood ethanol concentration was 86 ± 13 mg/dl, (i.e., binge intoxication) and performance on set shifting was correlated with parameters of SIP, indicating strong schedule-control of behavior. Ethanol monkeys made more errors/trial compared to control monkeys under 1.5 g/kg SIP. Set shifting performance at baseline did not predict average ethanol intake under 22 hr/day access which ranged from 1.6 -4.1 g/kg/day. Baseline rs fcMRI from putamen to premotor area F6 predicts 22/hr/day intake ($R = -0.64$, $p < 0.03$), and the change in rs fcMRI from putamen to premotor area F2 was proportional to the average ethanol intake (g/kg/day; $R = 0.59$, $p < 0.04$).
Conclusions: Rhesus monkeys show wide, but reliable, individual differences in daily ethanol consumption, cognitive flexibility, and in rs fcMRI of putamen to premotor cortical areas. By integrating these data sets, it appears that ethanol impairs set-shifting performance. Baseline putamen to premotor area connectivity is anti correlated with future ethanol daily intakes. However, the rs fcMRI of putamen to premotor areas after chronic drinking to becomes more positively correlated in the heavy drinking monkeys (intakes > 3.0 g/kg/day). These data are consistent with putamen synaptic changes following chronic ethanol drinking and provides a platform to study the emergence of habitual behavior associated with excessive alcohol intakes.
Disclosure: Nothing to Disclose.

Panel
60. Measuring LTP-Like Plasticity in Clinical Settings: New Opportunities for Understanding the Role of Neural Plasticity in Schizophrenia, and Bipolar Disorder, and the Psychosis-Risk Syndrome

60.1 Longitudinal and Cross-Sectional Investigations of LTP-Like Cortical Plasticity in Healthy Individuals and Subjects With Bipolar Disorder Type II

Torbjørn Elvsåshagen

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Background: Bipolar disorder (BD) has been conceptualized as a genetically influenced disorder of synaptic plasticity in limbic-cortical neural networks. However, there is a paucity of human evidence supporting synaptic dysfunction in BD, mainly due to a lack of methods for non-invasive measurements of synaptic plasticity. In recent years, modulation of the visual evoked potential (VEP) has emerged as a promising assay for non-invasive examination of long-term potentiation (LTP)-like synaptic processes in the human cerebral cortex. We conducted cross-sectional and longitudinal investigations of LTP-like visual cortex plasticity in individuals with BD type II and healthy controls (HCs) at baseline, as described previously (Elvsåshagen T, et al. Biol Psychiatry 2012), and on average 2.2 years later, at follow-up.
Methods: LTP-like plasticity was assessed at baseline and follow-up using the experimental paradigm described by Normann et al. (Biol Psychiatry 2007). In brief, VEPs were evoked by checkerboard reversals (check size.5°; 2 reversals/sec) in two premodulation blocks before and six blocks after a plasticity-inducing block of prolonged (10 min) visual stimulation. LTP-like plasticity was computed by subtracting premodulation VEP amplitudes from the corresponding postmodulation amplitudes. Twenty-six individuals with a DSM-IV diagnosis of BD type II and 40 HCs underwent the LTP-like plasticity paradigm at baseline and 45 of these participants were reexamined at follow-up (16 patients and

29 HCs). In addition, 13 new individuals with BD type II underwent the LTP-like plasticity paradigm at follow-up. At follow-up, saliva samples for cortisol analysis were collected using Salivette® Cortisol swabs immediately after awakening in the morning, 30 mins after the first collection, and at 12.30PM. The samples were analyzed with a Cortisol Saliva Luminescence Immunoassay according to the manufacturer ´s instructions and averaged across the three collections. All statistical analyses were conducted with SPSS version 24 for Windows and a two-tailed p value of $<.05$ was considered significant.

Results: We found 1) impaired VEP plasticity in BD type II at baseline, as described previously (Elvsåshagen T, et al. Biol Psychiatry 2012), and at follow-up (all $p<.05$), 2) that VEP plasticity was impaired in euthymic patients and was negatively correlated with depression severity at follow-up (all $p<.05$), 3) increased saliva cortisol in BD type II at follow-up and that VEP plasticity remained reduced in patients after adjusting for saliva cortisol (all $p<.05$), 4) that saliva cortisol was positively correlated with VEP plasticity in HCs (rho $=.52$, $p=.004$), and 5) that VEP plasticity exhibited moderate temporal stability when the examinations at baseline and follow-up were compared (intraclass correlation coefficients between.5 and.6).

Conclusions: The present study provides additional evidence for impaired LTP-like cortical plasticity in BD, suggests that impaired LTP-like plasticity might be a stable trait of BD which may further deteriorate during depressive episodes, and indicates that elevated cortisol does not underlie the plasticity impairment. The results also suggest a positive association between the LTP-like plasticity and saliva cortisol in HCs, possibly reflecting an inverted U-shaped relationship between cortisol and synaptic plasticity.

Disclosure: Part 1: Lundbeck, Honoraria.

60.2 A Novel ERP Paradigm to Assess Visual Neuroplasticity in Schizophrenia: Psychometric Properties

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Background: Neuroplasticity deficits may reflect the core pathophysiology of schizophrenia. Long-term potentiation (LTP), a form of neuroplasticity, can be assessed using event-related potentials (ERPs) by examining activity before and after tetanizing visual high frequency stimulation (HFS), with larger ERPs typically seen after stimulation. These changes after HFS are consistent with the signature characteristics of LTP, including persistence (minutes to hours), input specificity, and dependence on glutamatergic neurotransmission at N-methyl-D-aspartate receptors. ERP signatures of dysfunctional LTP in humans have the potential to serve as a biomarker in future treatment studies, though that biomarker must demonstrate good psychometric properties for it to be considered. However, the psychometric properties, including single session (internal consistency) and test-retest reliability, of this paradigm have not been studied in people with schizophrenia or healthy controls.

Methods: We assessed ERPs in 38 people with schizophrenia and 27 healthy controls at baseline and at a two-week follow-up. ERPs were examined in response to horizontal and vertical gratings before and 30-minutes after HFS. To assess input specificity HFS (i.e., tetanization) was administered with only one specific grating orientation, allowing us to compare tetanized vs. non-tetanized stimuli. We examined difference waves (post- minus pre-HFS) utilizing a mass-univariate permutation approach to identify time windows and electrodes exhibiting significant changes in ERPs associated with tetanization. We analyzed group differences, input specificity, internal consistency (i.e., split-half reliability) and test-retest reliability.

Results: We found evidence of greater neuroplasticity in the control group compared to patients, with a significant group by tetanization (i.e., post- minus pre-HFS) interaction. Controls showed greater negativity after HFS in parieto-occipital and occipital regions, but patients did not. We did not find evidence of input specificity, as this effect was comparable for tetanized and non-tetanized stimuli. Internal consistency and test-retest reliability of pre- and post-HFS ERPs were moderate to high in both groups; however, reliability for difference scores (i.e., post- minus pre-HFS differences) was poor in controls and moderate in patients.

Conclusions: These results demonstrate that visual neuroplasticity can be non-invasively assessed using ERPs and is impaired in schizophrenia. However, the current paradigm did not demonstrate input specificity. Regarding psychometrics, the results show that the internal consistency and test-retest reliability of the ERPs to tetanized and non-tetanized stimuli, before and after tetanization, was good to excellent in both groups. Despite this finding, the psychometrics for the difference wave (i.e., neuroplasticity effect) were poor in controls and moderate in patients. These findings show promise for using ERP-based measures of visual neuroplasticity as biomarkers in clinical trials.

Disclosure: Nothing to Disclose.

60.3 Multimodal Neuroimaging Study of Visual Plasticity in Schizophrenia

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Background: Patients with schizophrenia have learning and memory deficits that are associated with functional outcome. Alterations in the basic cellular mechanism underlying learning, long-term potentiation (LTP), may be responsible for these learning impairments. LTP is commonly assessed in rodents with high frequency electrical stimulation in the hippocampus, but LTP-like phenomena have also been observed in rodents in the visual cortex with high frequency visual stimulation. Several human studies have demonstrated LTP-like changes or "visual plasticity" as reflected by an increase in fMRI BOLD activation or visual evoked potentials following high frequency stimulation. This study used a modified fMRI visual plasticity paradigm to determine if visual plasticity was reduced in schizophrenia. The relationships between fMRI visual plasticity and occipital cortical

glutamate and glutamine levels measured with MRS, memory function, and functional capacity were examined.
Methods: Seventeen patients with DSM-IV diagnosis of schizophrenia and 18 controls participated in this study. MR scanning was conducted on a 3T Siemens Tim Trio. The fMRI visual plasticity paradigm consisted of low-frequency visual stimulation (flashing checkerboard at 0.9 Hz) for visual cortex activation, followed by high-frequency stimulation (flashing checkerboard at 9 Hz) to induce visual plasticity, and another low-frequency stimulation (flashing checkerboard at 0.9 Hz) for visual cortex activation. An increase in BOLD activation following high frequency visual stimulation is believed to reflect visual plasticity. Spectra were acquired from the occipital cortex using a STEAM sequence optimized for glutamate and glutamine. All participants also completed memory tests (HVLT and BVMT), and the UPSA-2 for functional capacity. Positive and negative symptoms were assessed with the BPRS and SANS in patients.
Results: Within group fMRI analysis indicated that visual cortex activation was significantly enhanced following high frequency stimulation in the control ($p < 0.05$, FWE), but not the schizophrenia group. Consistently, between group fMRI analysis indicated that the control group had greater visual plasticity compared to the schizophrenia group ($p < 0.05$, FWE). MRS measures of occipital glutamate and glutamine were significantly higher in the schizophrenia group compared to the control group (p's < 0.05). Occipital cortex glutamine was significantly correlated with visual plasticity in the control group but not in the schizophrenia group. There were no significant correlations between visual plasticity measures and memory, psychiatric symptom severity, or functional capacity measures.
Conclusions: These results indicate that fMRI visual plasticity was induced in the control group, but not to a significant degree the schizophrenia group. These results are consistent with previous reports of reduced visual plasticity in schizophrenia using EEG methods. Results also revealed that occipital cortex glutamine, the major metabolite of glutamate involved in neurotransmission, was related to visual plasticity in controls but not in schizophrenia. Given that glutamate is fundamentally involved in LTP, this relationship was expected. In summary, visual plasticity assessed with fMRI is reduced in schizophrenia and does not seem to be related to occipital cortical levels of glutamate or glutamine or memory function in this small sample.
Disclosure: Nothing to Disclose.

60.4 Deficits in LTP-Like Visual Cortical Plasticity Predict Transition to Psychosis in Individuals With the Psychosis Risk Syndrome

Daniel Mathalon

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Background: NMDA receptor-dependent long-term potentiation (LTP) is a mechanism of experience-dependent synaptic plasticity thought to underlie many forms of learning and memory. Consistent with the NMDA receptor hypofunction model, deficient synaptic plasticity has been increasingly implicated in the pathophysiology of schizophrenia and its associated cognitive deficits. Recently, basic neuroscience paradigms that induce LTP with high frequency electrical stimulation have been adapted for human in vivo studies by inducing LTP-like potentiation of visual evoked potentials (VEP) using "tetanizing" visual high frequency stimulation (VHFS). Using a variant of this sensory LTP paradigm, we previously demonstrated in healthy individuals that presentation of a checkerboard stimulus at a tetanizing frequency of 8.9 Hz (VHFS) for 2 minutes induced potentiation of the VEP evoked by the stimulus, evident as enhanced negativity between 125-175 ms post-stimulus onset at occipital electrode sites, that persisted for at least 20 minutes. Patients with chronic schizophrenia failed to show this potentiation, consistent with deficits in LTP-like visual cortical plasticity. Here, we extend this work by examining whether these LTP-like visual cortical plasticity deficits are present prior to the onset of psychosis in individuals exhibiting the psychosis-risk syndrome, and further, whether these deficits are predictive of increased risk of transition to full psychosis.
Methods: An interim participant sample from the ongoing 9-site North American Prodrome Longitudinal Study (NAPLS--3), comprising 246 individuals meeting criteria for the psychosis risk syndrome (PRS) and 46 healthy controls (HC), underwent baseline electroencephalographic recording during a visual sensory LTP paradigm. VEPs evoked by randomly intermixed, equiprobable, vertical and horizontal line grating stimuli were obtained pre- and 30-min post-VHFS (8.9 Hz stimulation rate, 5 sec on/off repeating pattern, for 4 minutes). One of the two line grating stimuli were presented during VHFS while the other served as a control stimulus, allowing assessment of VEP changes specific to the tetanized stimulus, analogous to the "input specific" effects that characterize classical LTP.
Results: Using a rigorous permutation-based clustering analytic approach in the entire sample, a temporal window in the VEP (128 - 204 ms post-stimulus onset) at electrode O2 showed a significant ($p = .03$, corrected) post-pre VHFS increase in negative voltage specific to the tetanized stimulus. Subsequently, this plasticity effect was examined in 20 PRS individuals who subsequently transitioned to psychosis (PRS-T), 22 PRS individuals followed for 18 months without transitioning to psychosis (PRS-NT), and 46 HC. The effect was significant in HC ($p = .05$) and PRS-NT ($p = .002$), but not in the PRS-T ($p = .59$). Moreover, the PRS-T group's plasticity effect was significantly deficient ($p = .02$) relative to the PRS-NT group.
Conclusions: VHFS induced a significant input-specific LTP-like visual cortical plasticity effect that persisted for at least 30 minutes. Significant deficits in this plasticity effect were evident at baseline only in PRS-T individuals, suggesting that LTP-like visual cortical plasticity deficits predate psychosis onset and may help predict which PRS individuals are at greatest risk for transitioning to psychosis.
Disclosure: Part 1: Boehringer-Ingelheim, Consultant, Takeda Pharmaceutical Company, Advisory Board, Alkermes, Consultant, Upsher-Smith, Consultant.

Panel
61. Duration of Untreated Psychosis: Evidence for Impact on Clinical Course and Biologic Mechanisms

61.1 Characteristics and Effects of the Duration of Untreated Psychosis on Outcomes in Patients With Schizophrenia: Results From a Large-Scale Meta-Analysis

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Background: In schizophrenia, a limited number of moderators and mediators of outcomes have been identified. Among these factors, the duration of untreated psychosis (DUP) is one of the most consistently reported moderators of poor outcomes. However, a comprehensive meta-analysis of a) the characteristics and time or regional trends of DUP and b) effects of DUP on a wide array of outcomes in patients with schizophrenia is missing.
Methods: We conducted a systematic literature search in PubMEd, PycInfo and Embase for studies in first-episode / early-phase schizophrenia or psychosis. Studies that reported on a) DUP or b) DUP in relationship to outcomes were included in a random effects meta-analysis as well as metaregression analyses. If more than one study from the same sample existed, the largest study for each given outcome was chosen and overlapping samples with $>=50\%$ of the same sample were excluded.
Results: Of 9,745 hits, we identified 312 individual studies ($n=22{,}619$, mean age 26.8 ± 3.2 years, males: 61.4%, White: 29.7%) with DUP data. 41.5% were cross-sectional studies. The mean Positive and Negative Syndrome Scale (PANSS) total score at baseline was 72.2 ± 18.8 points. The mean duration of untreated psychiatric illness was 5.7 ± 2.4 years; the mean DUP was 1.2 ± 0.9 years. In preliminary linear regression analyses, earlier year of study publication ($p=0.026$) and higher total PANSS score were related to longer DUP, whereas sex, age, race where not. Detailed results on correlates of mean as well as median DUP and on the relationship between DUP and baseline characteristics as well as short-, medium- and long-term outcomes will be available in the full data set and presented at the time of the ACNP Meeting. Additional analyses will compare samples with study defined "short" and "long" DUP, as well as before and after interventions aimed at reducing DUP.
Conclusions: DUP remains inappropriately high across different times and regions in the world, including the US. Additional results on the correlates of DUP and the relationship between DUP and specific outcomes, available at the meeting, will help identify targets and develop interventions aimed at shortening DUP, which will hopefully help improve overall outcomes in schizophrenia.
Disclosure: Part 1: Alkermes, Consultant, Allergan, Consultant, Bristol-Myers Squibb, Honoraria, Gerson Lehrman Group, Honoraria, IntraCellular Therapies, Advisory Board, Janssen/J&J, Consultant, LB Pharma, Consultant, ProPhase, Consultant, Lundbeck, Consultant, Medavante, Consultant, Medscape, Consultant, Neurocrine, Employee, Otsuka, Consultant, Pfizer, Consultant, Sunovion, Consultant, Takeda, Consultant, Teva, Consultant,
Part 2: Bristol-Myers Squibb, Honoraria, IntraCellular Therapies, Honoraria, Janssen/J&J, Honoraria, Lundbeck, Honoraria, Otsuka, Honoraria, Pfizer, Honoraria, Sunovion, Honoraria,
Part 3: Bristol-Myers Squibb, Consultant, IntraCellular Therapies, Honoraria, Janssen/J&J, Honoraria, Lundbeck, Honoraria, Otsuka, Honoraria, Pfizer, Honoraria, Sunovion, Honoraria, **Part 4:** Teva, Honoraria.

61.2 Hippocampal Atrophy, Duration of Untreated Psychosis and Molecular Biomarkers During Initial Antipsychotic Treatment of First Episode Psychosis

Donald Goff

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Background: Duration of untreated psychosis (DUP) has been linked to clinical course in schizophrenia, but the mechanism underlying this relationship remains unknown. We chose to study hippocampal volume in relation to DUP because hippocampal volume loss occurs early in the illness, correlates with outcomes and may "drive" progression of illness.
Methods: 71 medication-naïve individuals with nonaffective psychosis (FEP) and 73 age and gender-matched healthy controls (HC) were studied at baseline. FEP subjects were treated with second generation antipsychotics and after approximately 8 weeks 31 FEP subjects and 32 HC were re-studied. The primary outcome measure was left hippocampal volumetric integrity (LHVI), an automated estimate of the parenchymal fraction in a standardized hippocampal volume of interest.
Results: At baseline, LHVI was reduced in FEP subjects compared to HC ($p=.001$) and decreased at a median annualized rate of 4.1% compared to an increase of 1.3% in HC ($p=.001$). DUP inversely correlated with the change in LHVI ($r=-.61$, $p=.002$). Change in LHVI also inversely correlated with baseline BPRS agitation score ($r=-.45$, $p=.03$) and change in BPRS negative symptoms ($r=-.41$, $p=.05$). An exploratory LASSO regression found significant interactions in predicting baseline LHVI between DUP and concentrations of peripheral inflammatory markers (INFγ and IL8) and genotypes for NOS1, BDNF, ZFN804A, and COMT. Significant interactions were also found between DUP and peripheral concentrations of markers of astrocytic injury and oxidative stress (S100B and thioredoxin) and with NOS1 genotype in predicting the change in LHVI. Cumulative antipsychotic exposure did not correlate with LHVI change.
Conclusions: Hippocampal volume loss during early treatment with antipsychotics was highly correlated with DUP in our sample and may play a role in the relationship between DUP and clinical course. Results from our exploratory analysis require replication, but suggest that an effect of untreated psychosis on hippocampal volume loss may be mediated by inflammation, oxidative stress, BDNF, and dopamine transmission or metabolism.
Disclosure: Part 1: Avinar Pharmaceuticals, Grant, **Part 4:** Avanir, Grant.

61.3 Duration of Untreated Psychosis and Functional Neuroimaging in Early-Phase Schizophrenia: A Focus on Central Executive Connectivity and Activation

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Background: Longer duration of untreated psychosis (DUP) in patients with early-phase schizophrenia is associated with poorer response to antipsychotic treatment. Our understanding of the neurobiology underlying this phenomenon remains largely unknown. Meanwhile, functional neuroimaging has not widely been applied to examine neural correlates of DUP. In the present analysis, we characterized DUP-related variation in functional measures with a focus on the central executive network (CEN). We combine the results from our previous work (Sarpal et al. 2017), which examined patients with resting-state functional connectivity of the striatum, with evidence from an independent cohort of patients who were scanned during a working memory (WM) paradigm.
Methods: In our previous work, we included early-phase schizophrenia patients (N = 83) who underwent resting-state scanning while entering clinical trials with second-generation antipsychotic medications. A seed-based functional connectivity analysis of the striatum was performed. In a separate site, patients with first-episode psychosis (N = 31) were scanned during performance of a visual WM task. DUP was calculated via patient interview and normalized via log transformation. Activation of a priori CEN regions during maintenance and increasing WM load was examined along with DUP.
Results: In our prior work, we found that longer DUP associated with overall decreased functional connectivity between the striatum and cortical regions, largely located within the CEN. In a post-hoc analysis, these connectivity results mediated the negative association between DUP and response to antipsychotic treatment (Sarpal et al. 2017). Supporting these findings, we observed a negative relationship between length of DUP and activation of CEN regions, including the dorsolateral prefrontal cortex, bilaterally, during WM maintenance (left: $p = 0.006$; right: $p = 0.04$).
Conclusions: In conjunction, findings from both independent sites demonstrate converging evidence for an association between DUP and CEN functioning. Future work will be necessary to link these findings with prospective functional imaging studies focused on treatment outcomes.
Disclosure: Nothing to Disclose.

61.4 The Relationship Between Dopamine and Glutamate and State Changes in Psychosis: Multimodal Imaging Findings

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Background: Striatal dopamine overactivity is thought to underlie the onset of psychosis, potentially driven by cortical glutamate dysfunction through cortico-subcortical projections. However, the link between dysfunction in these systems and state changes in psychosis remains to be tested. It is predicted that patients with longer duration of untreated psychosis will show more marked alterations, and that improvements in symptoms will be associated with reductions in these alterations.
Methods: We conducted a prospective case-control study in patients presenting in their first episode of psychosis. 47 volunteers (33 patients with first episode psychosis and 14 controls) received 18F-DOPA PET and [1H]-MRS imaging to index dopamine synthesis capacity and glutamate levels respectively at baseline. The patients received clinical measures and were re-scanned after receiving approximately 6 weeks of antipsychotic treatment.
Results: Shorter duration of untreated psychosis was associated with greater baseline striatal dopamine synthesis capacity (rho = 0.65, $p = 0.01$). Greater reduction in dopamine synthesis capacity with treatment was associated with greater improvement in psychotic (rho = -0.46, $p < 0.05$), and total symptoms (rho = 0.51, $p < 0.05$). In contrast, there were no relationships between baseline glutamate levels and duration of untreated psychosis, or between change in glutamate levels over time and improvement in symptoms.
Conclusions: These data do not support our hypothesis and instead suggest that a rapid presentation of psychosis is associated with more marked dopaminergic elevation, and greater plasticity in the dopamine system, and subsequent better response to treatment.
Disclosure: Part 1: Dr. Howes has received investigator-initiated research funding from and/or participated in advisory/ speaker meetings organised by Astra-Zeneca, Autifony, BMS, Eli Lilly, Heptares, Jansenn, Lundbeck, Lyden-Delta, Otsuka, Servier, Sunovion, Rand and Roche. Neither Dr. Howes or his family have been employed by or have holdings/ a financial stake in any biomedical company, Honoraria, Self.

Study Group

62. Animal Research Committee: Advocating for Animal-Based Neuropsychopharmacology Research

Sari Izenwasser*, James Jentsch, Paula Clifford, Logan France, Amanda Dettmer, Marilyn Carroll, Nancy Ator, James Jentsch

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Study Group Summary: Animal models remain central to the mission of neuropsychopharmacological research because of the crucial necessity for laboratory animals in basic and translational science research. In recognition of the need for ongoing advocacy for humane and responsible animal research, the ACNP reactivated the Animal Research Committee (ARC) with the charge being to engage, in a supportive way, with other existing scientific societies, advocacy groups and accrediting organizations for which animal research is a central focus, as well as to review and develop ACNP resources that articulate "the positive aspects of the ethical use of animals in research." This Study Group

will focus on the diverse ways that neuropsychopharmacologists can participate in this advocacy and will welcome input from the audience on ways that the ARC can be helpful to ACNP members. The panel participants include Sari Izenwasser, Ph.D. (Chair of the panel) and David Jentsch, Ph.D. (co-Chair and moderator). David is a behavioral neuroscientist who has committed himself to advocacy for animal-based research in reaction to animal rights extremism that focused on UCLA researchers. Marilyn Carroll, Ph.D. is a preclinical behavioral pharmacologist who advocates for animal research and co-founded STAR (Speaking Truth about Animal Research), a coalition of scientific societies, based in the American Psychological Association, that seeks to advocate for the value of animal research, support targeted researchers, and multiply the efficacy of scientific societies in this realm through coordinated efforts. Paula Clifford is the Executive Director of Americans for Medical Progress (AMP), an organization dedicated to protecting biomedical research, with a focus on public outreach by providing information on the important findings being discovered with humane animal research. Amanda Dettmer, Ph.D. is a behavioral neuroscientist, the representative of the American Society of Primatologists to STAR, and an editor at Speaking of Research, an organization focused on providing, through its website, accurate information about the importance of animal research and testing in medical and veterinary science. Logan France, DVM was awarded a Hayre Fellowship by AMP and used it to found BRAD (Biomedical Research Awareness Day) to raise awareness, first with veterinary students, and this year with medical students, about the important role of animals in biomedical research. Nancy Ator, Ph.D. is a behavioral pharmacologist who represents ACNP in STAR as well as on the board for AAALAC International, the association that assesses and accredits institutions involved in animal research. This panel encompasses a wide range of experience and methods for advocacy. Strategies and opportunities for advocacy will be discussed in an interactive discussion with attendees.

Disclosure: Nothing to Disclose.