American College of Neuropsychopharmacology

Check for updates

ABSTRACTS COLLECTION

ACNP 61st Annual Meeting: Poster Abstracts P271-P540

Neuropsychopharmacology (2022) 47:220–370; https://doi.org/10.1038/s41386-022-01485-0

December 4-7, 2022

Phoenix Arizona

Sponsorship Statement: Publication of this supplement is sponsored by the ACNP. All content was reviewed and selected by the Program Committee, which held full responsibility for the abstract selections. Only disclosures for presenting authors are listed. Asterisks in the author lists indicate presenter of the abstract at the annual meeting.

Abstract numbers do not correlate to poster number assigned for presentation at the Annual Meeting.

P271. Negative and Positive Urgency-Related Left Dorsolateral Prefrontal Cortex Activity During Emotion Processing Predicts Manic Symptom Changes One Year Later in Distressed Young Adults

Maya C. Schumer*, Michele A. Bertocci, Haris A. Aslam, Simona Graur, Genna Bebko, Richelle S. Stiffler, Alexander S. Skeba, Tyler J. Brady, E. Kale Edmiston, Henry W. Chase, Sheri L. Johnson, Mary L. Phillips

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

Background: There is a critical need for identifying robust biomarkers of objective risk factors associated with Bipolar Disorder (BD), particularly those predictive of mania/hypomania and mixed states, as these are pathognomonic features of BD. Moreover, identifying biomarkers of future BD risk informs not only early risk detection and targets for treatment developments, but also aids our understanding of pathophysiological processes underlying BD. Impulsivity is a widely studied characteristic of BD that is elevated during manic and mixed episodes and persistent during euthymic periods. While impulsivity is a multi-faceted trait, emotion-triggered impulsivity, or emotion-based rash action, is perhaps the most important form, as it is associated with a broad range of key outcomes in BD, including and notably suicidality. Negative and positive urgency (NU/PU), defined as the tendencies to act impulsively in response to negative and positive affect respectively, are evident in adults with BD and have been identified as risk factors for future BD, yet neuroimaging studies examining neural correlates of urgency in the context of predicting BD risk are sparse. We previously reported negative relationships between NU and PU-related blood oxygen level dependent (BOLD) activity in the left dorsolateral prefrontal cortex (L DLPFC) during approach emotion processing and lifetime mania risk in a transdiagnostic cohort (excluding young adults with BD diagnosis) of 106 young adults who were seeking treatment for psychological distress. However, it has not yet been tested whether urgency-related L DLPFC activity is a predictor of manic symptoms over time, and if the relationship is specific to mania or if it also extends to depressive symptoms.

Methods: Twenty-one distressed adults ages 18-23 (mean age 20.84 ± 1.49, 17 female), from the initial aforementioned trans-diagnostic cohort that were scanned on a 3 T fMRI while performing an implicit facial emotion processing task, completed baseline and 12-month follow-up assessments of hypo/mania and depression measured by the Moods Spectrum Scale (MOODS) Mood Manic and Depressive Domains. Angry and happy face-related activity at baseline scan was examined using an anatomical mask of regions supporting emotion processing and executive function, which included the L DLPFC. Parameter estimates were extracted from separate multiple regression models of NU and PU covarying for age and gender, $p < 0.001$, uncorrected, $k = 20$.

We performed two parallel, separate multiple linear regression models with NU and PU-related L DLPFC BOLD activity, baseline MOODS Mood Manic Domain score, age, and gender as predictors and Mood Manic Domain score at 12-month follow-up as the dependent variable.

To test whether the L DLPFC was specific to predicting mania, we performed two additional multiple regression models (one for NU, one for PU) controlling for baseline depression, with 12-month MOODS Mood Depressive Domain score as the dependent variable.

Results for both the primary analysis with mania and the specificity analysis with depression were each considered significant at $p < 0.025$, Bonferroni-corrected for two models.

Results: The initial models with baseline demographic variables explained 18% of the variance in 12-month MOODS Mood Manic Domain score. The full model including NU-related L DLPFC activity was significant, $F(2,16)=4.88$, $p = 0.009$, with an adjusted R2 of 0.44, such that the L DLPFC explained 26% of the variance and was significantly associated with mania one year later ($\beta = -8.07$, $t = -2.95$, $p = 0.009$). The full model including PU-related L DLPFC activity was also significant, $F(2,16)=4.2$, $p = 0.016$, with an adjusted R2 of 0.39, such that the L DLPFC explained 21% of the variance and was significantly associated with mania one year later ($\beta = -7.24$, $t = -2.61$, $p = 0.019$).

In the specificity analysis with 12-month MOODS Mood Depressive Domain score, the full model including NU-related L DLPFC activity was not significant, $F(2,16)=3.56$, $p = 0.029$, with an adjusted R2 of 0.338. L DLPFC activity was not significantly

associated with depression one year later (β = -6.24, t = -2.23, p = 0.041). The full model including PU-related L DLPFC activity was not significant, F(2,16)=3.08, p = 0.047, with an adjusted R2 of 0.294. L DLPFC activity was not significantly associated with depression one year later (β = -5.53, t = -1.91, p = 0.074).

Conclusions: Attenuated activation of NU- and PU-related L DLPFC at baseline both predicted changes in mania one year later, and their significant associations were indeed specific to mania. These preliminary findings underscore the importance of urgency-related central executive network activity during emotion processing in predicting manic symptom changes over time, indicative of greater risk for future BD. Future research should further extend these findings to a larger sample and assess if these markers can additionally predict conversion to BD in at-risk individuals, as well as investigate the potential for targeting the L DLPFC for therapeutic interventions.

Keywords: Bipolar Disorder, Mania, Dorsolateral Prefrontal Cortex, Negative Urgency, Emotion Processing

Disclosure: Nothing to disclose.

P272. Systemic Inflammation Response Index (SIRI) Correlates to Inflammatory-Metabolic Markers in Treatment-Resistant Bipolar Depression (TRBDD) and May Predict Treatment Response

Nausheen Baig*, Kyle Decker, Stephen Murata, Debra Hoppensteadt, Jawed Fareed, Angelos Halaris

Loyola University Chicago Stritch School of Medicine, Oak Brook, Illinois, United States

Background: Inflammation has been associated with depressive illness and treatment resistance (Haroon and Miller, 2017; Miller et al., 2009), prompting a search for predictive biomarkers of treatment response. In a randomized, double-blind, placebo-controlled trial we demonstrated efficacy of escitalopram (ESC) + Celecoxib (CBX) compared to ESC + placebo (PBO) in TRBDD (In this secondary analysis, we characterized treatment response (HAMD17) in relation to SIRI (monocytes x neutrophils/lymphocytes). We hypothesize that treatment response to ESC + CBX is associated with baseline of and/or changes in SIRI levels.

Methods: The sample (N = 79) included healthy controls (n = 32) and TRBDD subjects (n = 47). TRBDD cohort included an ESC + CBX arm (n = 26) and ESC + PBO arm (n = 21). SIRI was calculated from complete blood count (CBC) drawn at baseline and end of treatment (week 8). Inflammatory biomarkers were measured using ELISA assays. Kynurenine pathway (KP) metabolites were measured using HLPC. HAMD17 scores were obtained weekly. Treatment remission was defined as HAMD17 < 7 by treatment week 8. Univariate correlations were explored between SIRI and inflammatory and KP biomarkers. Post-treatment HAMD17 was modeled according to treatment arm, baseline HAMD17, baseline SIRI, and relevant interactions using multivariate linear regression. Statistical analysis was conducted using R-3.6.3.

Results: In the TRBDD sample (n = 47) there were n = 14 (38%) remitters by week 8. Remission was significantly associated with ESC + CBX treatment (X2 = 8.3, p = 0.004). Group comparison by treatment group revealed no differences in sex, BMI, or blood counts; however, patients in the CBX + ESC arm were older (p = 0.032).

On univariate analysis of baseline SIRI, there were no significant correlations with treatment response at week 8 (whether measured as a continuous or categorical outcome), although baseline SIRI was correlated with baseline HAMD17 (p = 0.008). Baseline SIRI was significantly associated with higher TNF-alpha at week 8 (p = 0.007) and IL-2 at baseline (p = 0.053), elevated picolinic acid at both baseline (p = 0.034) and week 8 (p = 0.037), and lower 3-hydroxykynurenine at week 4 (p = 0.030). Notably, baseline SIRI also trended towards significant correlation with lower baseline IL-4 (p = 0.057) and higher baseline anthranilic acid (p = 0.071).

Multivariate analysis showed HAMD17 was significantly associated with SIRI at week 8, adjusting for treatment arm (beta=5.74, p = 0.007, R2/R2 adjusted=0.553/0.531). Finally, HAMD17 at week 8 was significantly associated with ESC + CBX treatment (p < 0.002) and an interaction with baseline SIRI and baseline HAMD17 (β-estimate =8.3 [95% CI (0.23, 1.44)], p = 0.008) (R2/R2 adjusted = 0.354/0.286).

Conclusions: Post-treatment HAMD17 was independently predicted by treatment arm and an interaction of baseline SIRI with HAMD17. This suggests that, although baseline SIRI may not be an independent predictor of treatment response, it may be indicative of poor prognosis amongst patients with elevated pre-treatment depressive severity. It is notable that this relationship is not dependent on treatment arm. Considering the associations with pro-inflammatory markers and KP metabolites reported here, it is plausible that the biological significance of SIRI may implicate these pathways. Future studies on SIRI with a larger sample size are warranted to clarify its potential role as a readily obtained/accessible predictive biomarker of treatment response in TRBDD.

References:

Halaris, A., Cantos, A., Johnson, K., Hakimi, M., and amp; Sinacore, J. (2020). Modulation of the inflammatory response benefits treatment-resistant bipolar depression: A randomized clinical trial. Journal of Affective Disorders, 261, 145–152. https://doi.org/10.1016/j.jad.2019.10.021

Haroon, E., and amp; Miller, A. H. (2017). Inflammation Effects on Glutamate as a Pathway to Neuroprogression in Mood Disorders. Modern Trends in Pharmacopsychiatry, 31, 37–55. https://doi.org/10.1159/000470805

Mazza, M. G., De Lorenzo, R., Conte, C., Poletti, S., Vai, B., Bollettini, I., Melloni, E. M. T., Furlan, R., Ciceri, F., Rovere-Querini, P., and amp; Benedetti, F. (2020). Anxiety and depression in COVID-19 survivors: Role of inflammatory and clinical predictors. Brain, Behavior, and Immunity, 89, 594–600. https://doi.org/10.1016/j.bbi.2020.07.037

Mazza, M. G., Palladini, M., De Lorenzo, R., Magnaghi, C., Poletti, S., Furlan, R., Ciceri, F., COVID-19 BioB Outpatient Clinic Study group, Rovere-Querini, P., and amp; Benedetti, F. (2021). Persistent psychopathology and neurocognitive impairment in COVID-19 survivors: Effect ofinflammatory biomarkers at three-month follow-up. Brain, Behavior, and Immunity, 94,138–147. https://doi.org/10.1016/j.bbi.2021.02.021

Miller, A. H., Maletic, V., and amp; Raison, C. L. (2009). Inflammation and its discontents: The role of cytokines in the pathophysiology of major depression. Biological Psychiatry, 65(9), 732–741. https://doi.org/10.1016/j.biopsych.2008.11.029

Zahorec, R. (2021). Neutrophil-to-lymphocyte ratio, past, present and future perspectives. Bratislavske Lekarske Listy, 122(7), 474–488. https://doi.org/10.4149/BLL_2021_078

Zhang, Y., Xing, Z., Zhou, K., and amp; Jiang, S. (2021). The Predictive Role of Systemic Inflammation Response Index (SIRI) in the Prognosis of Stroke Patients. Clinical Interventions in Aging, 16, 1997–2007. https://doi.org/10.2147/CIA.S339221

Keywords: Bipolar I Depression, Systemic Inflammation, Neuroprotection, Neuropharmacology

Disclosure: Nothing to disclose.

P273. The Continuous-Performance Emotion-Regulation (CERT) Task: A Novel fMRI Paradigm to Investigate Inter-Neural Network Connectivity Shifts During Affect-Regulation in Depressed and Healthy Subjects

Jungwon Cha, Amit Anand*

Harvard Medical School, Boston, Massachusetts, United States

Background: Inter-network connectivity shifts are part of dynamic changes in brain configuration while it interacts with the environment. Hence, a continuous design corresponds more to real-life conditions. Furthermore, shifts in internetwork connectivity corresponds more accurately to brain function as it reconfigures to different emotion regulation conditions rather than static measurement during a single state. We have designed a novel resting state fMRI paradigm – the continuous-performance emotion-regulation task (CERT) to capture brain network re-configuration during emotion regulation. This study used CERT and Group-level Independent Components Analysis (ICA) to investigate differences in inter-neural network connectivity shifts during maintenance and regulation of negative emotions to distinguish between (i) Bipolar Depressed (BDD) and Major Depression (MDD), (ii) Patients (PA = BDD + MDD) and Healthy Controls (HC), and (iii) MDD patients who at high risk (HRMDD) and at low risk (LRMDD) for bipolar disorder.

Methods: Subjects: Data is included from a large sample of N = 298 medication-free participants recruited from the out-patient psychiatry clinics at the Indiana University School of Medicine and Cleveland Clinic, and by advertisement. All BDD and MDD participants were required to be medication-free for at least 2 weeks. The final analyses included 249 participants: 50 BDD (18 BDI, 32 BDII), 116 MDD (35 HRMDD, 45 LRMDD, 36 unspecified); and 83 HC subjects.

CERT Imaging: Functional MRI data were acquired in two different conditions: during separate runs of 5.25 minutes each, subjects were continuously shown negatively valences pictures while they either tried to maintain emotional response to the pictures or in the following set tried to suppress their emotional response using reappraisal techniques. Data collected from each of the runs was corrected for motion and physiological noise and band-pass filtered to retain frequencies between 0.008 - 0.08 Hz.

Internetwork Connectivity: Analysis: Group-level Independent Components Analysis (ICA) was performed using GIFT toolbox. The preprocessed resting state functional MR images of all subjects, both for maintenance condition (249 images) and regulation condition (249 images), were decomposed into independent components. Fourteen independent components were selected for Resting State Network (RSN) including dorsal and ventral Default Mode network (dDMN and vDMN), anterior and posterior Salience Networks (ASN and VSN), right and left Executive Control Network (ECN), Sensorimotor network (SMN), Precuneus network (PN), Visual and Auditory Networks (VN and AN), and the Language Network (LN). With the selected set of components, univariate tests were performed to investigate the inter-network connectivity difference between each individual group (BD, MDD, HRMDD, LRMDD, PA and HC) in the two different conditions (Maintain vs. Regulate) through Repeated-measure ANOVA (RMANOVA).

Results: Condition Effect: Within group - HC, MDD, and LRMMD exhibited differences in internetwork connectivity shifts during Maintain and Regulation Conditions for several pairs of networks including the DMN and SN. However, BDD and HRMDD groups did not show significant connectivity shift between the two conditions.

Group x Condition Effect:

a. BDD vs. MDD The interaction between group and condition was significant (FDR corrected $p < .01$) for inter-network connectivity between SMN-AN, SN-ECN and SN-VN.
b. PA vs. HC: The interaction between group and condition showed a significant difference (FDR corrected $p < .01$) in several pairs of FNC including vDMN-AN, dDMN-AN, and dDMN-ECN correlations.
c. HRMDD vs. LRMDD: significant interaction between the group and condition was seen for AN-VSN internetwork connectivity ($p < 0.01$, uncorrected).

Group Effect: No differences between groups were seen individually within Maintain and Suppress conditions.

Conclusions: Using a novel continuous emotion maintenance and regulation paradigm, differential internetwork connectivity-shifts distinguished between depressed subjects and healthy controls, MDD and BDD groups, and high and low-risk for bipolar disorder depressed subjects. Hence, internetwork connectivity-shifts across emotion regulation conditions rather than static measurement of internetwork connectivity in a single condition may serve as a more efficient biomarker for depression and depression sub-types.

Keywords: Brain Networks, Dynamic Functional Connectivity, Continuous Performance Task, Affect Regulation, Depression

Disclosure: Nothing to disclose.

P274. The Heinz C Prechter Bipolar Research Program: A Comprehensive Ontological Approach to the Study of Bipolar Disorder

Sarah Sperry*, Paul Jenkins, Kelly Ryan, Ivy Tso, Melvin McInnis

University of Michigan, Ann Arbor, Michigan, United States

Background: Bipolar disorder (BD) is a complex multidimensional illness characterized by pathological swings in moods and emotions that vary over time. The etiology of BD is not known, but causality is considered to be plural. The HC Prechter Bipolar Research Program at the University of Michigan is a collaborative network of research projects focused the discovery of mechanisms underlying the etiology of BD as well as the study of predictive patterns of outcomes of disease. The core features of the program include deep phenotyping and ongoing longitudinal collection of clinical data. The data are organized into 7 ontological platforms: 1) Neurocognitive functioning; 2) Personality, 3) Motivated behaviors; 4) Sleep and Circadian patterns; 5) Life Story and experiences; 6) Treatment and outcomes patterns; 7) Disease states.

Methods: There are currently 1,385 participants in the Prechter Cohort (mean age at enrollment is 39.2 years, 62% female) including 874 individuals with a BD and 282 controls. The median longitudinal follow-up is 9 years (range: 0 – 16). Deep clinical phenotyping gathers clinical data from each of the 7 ontological platforms. Standardized interviews, assessment batteries, and self-report questionnaires are used to assess neurocognitive functioning (e.g., visual memory, fine motor dexterity), personality (e.g., NEO PI-R), motivated behaviors (e.g., Alcohol Use Disorder Identification Test), sleep and circadian rhythms (e.g., PSQI, Munich Chronotype Questionnaire), life story (e.g., Life Events Checklist, Childhood Trauma Questionnaire), treatment and outcomes patterns (e.g., medication review, mood measures). Biological modelling of disease states uses induced pluripotent stem cell (iPSC) methods. To identify patterns between baseline characteristics and longitudinal course of illness, we employ

multilevel modeling, dynamic structural equation modeling, path analysis, and mathematical modeling approaches.

Results: We present primary findings from each of the ontological platforms herein. Neurocognitive Functioning: Individuals with BD have poorer cognitive performance compared to controls on visual memory ($t = 3.31$, $p = .001$) and fine motor dexterity ($t = 4.21$, $p < .001$) at baseline, one- and five-year follow ups. However, there is no overall difference in the rate of decline in neurocognition between BD and controls. Personality: Individuals with BD have higher neuroticism which tends to remain stable over time. Stability of neuroticism has a moderately strong association with depression over time (effects range from 0.21 to 0.52, ps < .001). Motivated Behaviors: Individuals with BD tend to increase their alcohol use over time (Est = .33, 95% credibility interval [.30,.35]). Increases in alcohol use appear to increase risk of depression 6 months later (Est = .04, 95% CI [.02, .07]) and greater mood instability in the following year (Est = .12, 95% CI [.05, .18]). Even mild levels of alcohol use predicted greater impairment to friendships ($\beta = .01$, $p = .02$) and work-related functioning (($\beta = .01$, $p = .04$). Sleep and Circadian Patterns: Circadian timing, as estimated from self-report chronotype appears to affect longitudinal outcomes, with those identifying as late chronotype experiencing a greater frequency of depressive symptoms over time (Est=1.29, 95% CI [1.14,1.47]). Life Story and experiences: Disruptions in childhood maternal attachment and current romantic attachment in the context of a history of childhood trauma were associated with greater depression severity (accounting for 12% of the total effect of childhood trauma on depression severity). Treatment and outcomes patterns: Mathematical modeling of longitudinal mood data suggests that individuals with BD are characterized by significant affective instability, wherein mood states do not follow a rhythmic process – BD episodes arise in the context of persistent instability with failure to return to normal states once perturbed. Disease States: Synaptic activity within iPSC cells differ significantly between BD and controls ($p < 0.05$); exosomal source / cargo significantly influences synaptic activity. The addition of control derived exosomes significantly increases the synaptic density in bipolar (iPSC) derived neurons ($p < 0.01$).

Conclusions: We have identified 7 ontological platforms that are critical to the understanding of longitudinal course and heterogeneity in BD. Baseline characteristics across these platforms seem to robustly predict severity and course of depressive symptoms. The organizational structure of the data benefits from consideration of an ontological approach in order to study static, categorical, and dynamic longitudinal features of the illness. There are biological, neurocognitive, personality, behavioral, sleep, life story, and outcome differences in comparing BD with controls and between individuals with bipolar that provides the basis for the study of underlying mechanistic differences in subtypes of illness. Future research with the HC Prechter Bipolar Research Program aims to use data-driven approaches to identify strata of individuals based on these 7 ontological platforms to implement personalized, and precision-medicine based interventions.

Keywords: Bipolar Disorder, Longitudinal Study, Deep Phenotyping, Mood and Cognition

Disclosure: Nothing to disclose.

P275. Behavioral and Electrophysiological Profiles along the Continuum of Suicide Risk: A Potential Role for Ketamine in Suicide Prevention

Steven Lamontagne*, Jessica Gilbert, Carlos Zarate, Elizabeth Ballard

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

Background: Suicide is a leading cause of death worldwide, accounting for at least one million fatalities per year (World Health Organization, 2021). Despite increasing rates of suicide, risk mitigation strategies have been largely unsuccessful. Two major barriers hinder these efforts: (1) a paucity of objective markers that probe suicidal states, constraining suicide research to self-report and (2) limited understanding of the neurobiology of suicide, precluding the development of biological interventions. Although progress has been made on these fronts, there is considerable heterogeneity in the characterization of suicide risk across clinical trials and neuroimaging studies. Thus, there is an urgent need to identify neural correlates associated with varying levels of suicide risk, as well as develop novel fast-acting therapeutics to modulate activity within these neural networks.

Methods: Seventy-five male and female adults (Mage=39.89, range 19-65) were recruited through a suicide-focused research protocol (NCT02543983). Whereas most studies broadly define suicide risk based on lifetime history of attempt(s), we examined parameters along a continuum of risk: (1) those with a suicide attempt in the past two weeks and/or lifetime suicidal ideation with intent (High Risk; HR) ($n = 15$), (2) those with a history of attempt, but no suicidal behavior or ideation with intent in the past year (Moderate Risk; MoR) ($n = 18$) (3) those with anxiety or mood symptoms, but no suicide history (Low Risk; LR) ($n = 19$), and (4) those without psychiatric or suicide history (Minimal Risk; MinR) ($n = 23$). We used a CTF 275-channel whole-head magnetoencephalography (MEG) scanner to examine electrophysiological correlates of suicide. During MEG scanning, participants completed a modified Life-Death Implicit Association Task, which is a computer task measuring associations of oneself to either life or death based on reaction times to words that represent each category (faster reaction time denotes stronger implicit bias) (Nock et al., 2010). The behavioral outcome of interest was the D-score, defined as the difference in mean reaction times between self-death and self-life trials divided by the standard deviation of all trials (positive D-scores reflect a stronger self-death association). MEG data were source-localized in the gamma (30-58 Hz) frequency, a proxy measure of excitation-inhibition balance, using a 1 s window from word onset and a linearly constrained minimum variance beamforming algorithm. A linear mixed-effects model implemented in AFNI was used to evaluate differences in gamma power between self-life and self-death word pairings. As a proof-of-concept open-label pilot study, we examined the effects of subanesthetic-dose ketamine, a N-methyl-D-aspartate (NMDA) receptor antagonist, known to have rapid anti-suicidal ideation properties, on gamma power in a small group of HR participants ($N = 5$).

Results: Behavioral results showed that D-scores in the HR group did not differ from zero ($p = 0.78$), but were significantly higher compared to the MoR, LR, and MinR groups (ps<0.01). D-scores for the latter three groups did not differ from each other (ps>0.43) and were significantly lower than zero (ps<0.001), denoting a self-life bias. Across groups, a linear mixed effects model showed enhanced gamma power for self-death compared to self-life conditions in the posterior cingulate cortex (PCC) ($p < 0.05$), a region associated with self-referential processing. A significant group-by-condition interaction ($p < 0.05$) revealed group differences in gamma power within the PCC, right insular cortex, and orbitofrontal cortex (OBF). Extracting gamma power estimates in these regions showed higher gamma power for self-death compared to self-life trials in the OBF for the HR group ($p < 0.01$) and the insula and PCC for the MinR group (ps<0.05). No differences in gamma power between self-life and self-death trials emerged for the LR group (ps>0.05). In the ketamine pilot study, D-scores were not affected by ketamine administration ($p = 0.57$); however, a session-by-condition interaction ($p < 0.05$) revealed enhanced gamma power for self-death trials in the left insula after ketamine administration compared to baseline ($p < 0.001$). Post-

ketamine insular gamma power for self-death trials inversely correlated with D-score ($r = -0.89$, $p < 0.05$), suggesting the insula might be an important biomarker for implicit cognitions about death in HR individuals.

Conclusions: These findings point to differential implicit cognitive processing of life and death depending on suicide risk, highlighting the need for patient-centered risk assessment in suicide prevention. Our findings also implicate a role for pharmacotherapies that modulate gamma activity, particularly in the PCC and insula, in risk mitigation. Toward this end, our preliminary data show promising effects of ketamine in modulating neural correlates of suicide-related cognitive processing.

Keywords: Suicide Risk Factors, Implicit Association Test, Ketamine, Magnetoencephalography

Disclosure: Nothing to disclose.

P276. The COVID19 Pandemic Disrupts Associations Between Neural Reward Connectivity and Affective and Anxiety Symptoms in Young Adults

Kristen Eckstrand*, Erika Forbes, Michele Bertocci, Henry Chase, Jeanette Lockovich, Richelle Stiffler, Haris Aslam, Alex Skeba, Simona Graur, Genna Bebko, Mary Phillips

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: The COVID19 pandemic has imposed significant morbidity and mortality, including the worsened mental health outcomes of higher depression and anxiety severity. Experiences of young adults during the pandemic - such as social isolation and increased stress - limit exposure to rewarding experiences and are associated with poorer mental health. Altered connectivity between neural reward regions, such as the ventral striatum (VS), dorsal anterior cingulate cortex (dACC), and ventrolateral prefrontal cortex (vlPFC), with other neural regions involved in salience detection and executive control has been implicated in depression and anxiety and may be a mechanism by which the COVID19 pandemic has worsened affective and anxiety symptoms in young adults. The purpose of this cross-sectional study was to examine whether the COVID19 pandemic altered relationships between neural reward connectivity and affective and anxiety symptoms.

Methods: 98 young adults (23.5 ± 2.9 years; 63 F/35 M sex) were included in this cross-sectional sub-study derived from a larger longitudinal study examining the development of mood and anxiety symptoms in young adulthood. Participants were divided into two pandemic groups, those who completed the study prior to the COVID19 pandemic ($n = 39$) and during the pandemic ($n = 59$), using the national lockdown date (March 13, 2020). Participants completed self-report measures of depression (Mood and Anxiety Symptom Questionnaire (MASQ) – Anhedonia Depression), anxiety (MASQ-Anxious Arousal), anhedonia (Snaith Hamilton Pleasure Scale), lifetime trauma exposure (Trauma History Questionnaire) and underwent fMRI scanning during a standardized monetary reward task. fMRI data were preprocessed using fMRIPrep 20.2.6 and 1st level analyses were completed in SPM12. The 1st level GLM included reward expectancy (RE), reward prediction error (RPE), and uncertainty expectancy (UE) as regressors. Wholebrain functional connectivity during RE and RPE was assessed using bilateral VS, dACC (Brodmann area 32), and vlPFC seeds. Six two-group 2nd level models (3 seeds x 2 connectivity conditions) were performed in SPM12 comparing neural connectivity between COVID19 pandemic groups. Each model included independent variables of self-reported depression, anhedonia, and anxiety, interacting with pandemic group. Age, sex, IQ, psychotropic medication load, bipolar diagnosis, and lifetime trauma exposure were included as covariates. Significance was defined in SPM12 as punc<0.001, cluster corrected at pFWE<0.05.

Results: Participants who completed the study during the COVID19 pandemic reported higher anhedonia ($t = -2.112$, $p = 0.019$) and higher anxiety ($t = -2.789$, $p < 0.003$) severity, but not depression severity, compared to those who completed the study before the COVID19 pandemic. The COVID19 pandemic impacted associations between affective symptoms and both dACC and vlPFC connectivity during RE and RPE. During RE, lower dACC-mPFC (kE=108 voxels, pFWE=0.015) connectivity was associated with higher anhedonia severity during the COVID19 pandemic compared to lower anhedonia severity before the COVID19 pandemic. Lower vlPFC connectivity with the left cerebellum (kE=131 voxels, pFWE=0.009), bilateral thalamus (kE=165 voxels, pFWE=0.002), right parietal lobule (kE=171 voxels, pFWE=0.002), middle cingulate cortex (kE=94 voxels, pFWE=0.044), and right precentral gyrus (kE=133 voxels, pFWE=0.008) during RE was associated with higher anxiety severity during the COVID19 pandemic compared to lower anxiety severity before the COVID19 pandemic. During RPE, higher vlPFC connectivity with the right temporoparietal junction (kE=119 voxels, pFWE=0.022), right dorsomedial prefrontal cortex (kE=176 voxels, pFWE=0.003), dACC (kE=98 voxels, pFWE=0.002) and middle cingulate cortex (kE=181 voxels, pFWE=0.002) during RPE was associated with higher anhedonia severity during the COVID19 pandemic compared to lower anhedonia severity before the pandemic. Higher dACC connectivity with the left vlPFC (kE=169 voxels, pFWE=0.001) and right ventral postcentral gyrus (kE=144 voxels, pFWE=0.003) was associated with higher anxiety severity during the COVID19 pandemic compared with lower anxiety severity before the COVID19 pandemic.

Conclusions: The COVID19 pandemic changed the direction of associations between anhedonia and anxiety severity and reward circuitry connectivity. Higher anhedonia severity was associated with lower dACC connectivity with salience regions during RE and higher vlPFC connectivity with the central executive network during RPE, suggesting a diminished ability to anticipate rewards and overregulation of reward response may have contributed to higher anhedonia severity during the pandemic. The opposite pattern emerged for anxiety, where higher anxiety severity was associated with a combination lower vlPFC connectivity with the central executive network during RE and higher dACC connectivity with salience regions during RPE. This indicates a lesser ability to evaluate and attend to potential rewards, combined with a heightened sensitivity to rewards, impacted anxiety severity during the pandemic. Combined, opposing and ineffective compensatory relationships between reward circuitry and both salience and central executive networks during the pandemic may be a mechanism for the elevated anxiety and depression severity observed during the pandemic.

Keywords: The COVID-19 Pandemic, Reward Circuitry, Depression and/or Anxiety, Young Adults

Disclosure: Nothing to disclose.

P277. Altered Patterns of Central Executive, Default Mode and Salience Network Activity and Connectivity are Associated With Concurrent and Future Depression Risk in Two Independent Young Adult Samples

Michele Bertocci*, Yvette Afriyre-Agyemang, Satish Iyengar, Renata Rozovsky, Richelle Stiffler, Haris Aslam, Genna Bebko, Mary Phillips

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: A range of subsyndromal-syndromal affective symptoms underlie emotional distress and are often early

predictors of future depression in young adults. Greater functional connectivity (FC) among the central executive network (CEN), supporting emotional regulation (ER) subcomponent processes such as working memory (WM), the default mode network (DMN), supporting self-related information processing, and the salience network (SN), is thought to interfere with cognitive functioning and predispose to depressive disorders. These patterns of neural network function are therefore potential early, objective neural markers of depressive disorders; yet, few studies have examined how activity within, and FC among, these neural networks during WM and ER tasks are associated with depression severity in young adults. We aimed to: 1. elucidate relationships among activity and FC in these networks and concurrent depression, using a paradigm designed to examine WM and ER capacity; 2. examine the extent to which these relationships were specific to depression versus mania/hypomania; 3. test whether findings in a first, "discovery" sample could be replicated in a second, "test" sample of young adults; and 4. test whether such relationships also predicted future depression and/or mania/hypomania severity.

Methods: We examined relationships among concurrent depression and mania/hypomania severity and neural activity and FC during the above paradigm in two independent samples of young adults: a discovery sample: n = 90, 60 female, age=21.7 (2.0) and a test sample: n = 96, 65 female, age=21.6(2.1). Depression severity was assessed using the Hamilton Depression Rating Scale, and mania/hypomania severity, using the Young Mania Rating Scale. We examined activity and FC using an emotional regulation neural mask comprising key regions in each network: the dorsolateral prefrontal cortex and caudate in the CEN; the precuneus in the DMN; and the dorsal anterior cingulate cortex and amygdala in the SN (FWE p = .001, k > 10). Elastic net variable selection was performed using GLMNET followed by negative binomial regression to determine relationships among clusters of significant activity and FC in this mask and concurrent depression and mania/hypomania severity in the discovery sample. Negative binomial regression analyses determined whether the significant neural activity and FC-affective symptom relationships in the discovery sample were replicated in the test sample. We then examined relationships among neural activity and FC and future depression and mania/hypomania severity in a subsample derived from both samples (n = 61, 45 female, age =21.6 (2.1)) who were clinically assessed for up to 12 months after the scan.

Results: In the discovery sample during working memory: dlPFC activity (Odds Ratio (OR):1.80, CI:1.42-2.28, qFDR < .001), precuneus activity (OR:1.43, CI:1.12-1.82, p = .004, qFDR=0.004), were positively associated with concurrent depression severity. These relationships were replicated in the test sample: dlPFC activity (OR:2.90, CI:2.31-3.64, qFDR < .001) and precuneus activity (OR:1.88, CI:1.54-2.29, qFDR < .001). These measures were also positively associated with future depression severity: dlPFC activity (OR:2.81, CI:2.21-3.56, qFDR<0.001), and precuneus activity (OR:1.49, CI:1.21-1.85, qFDR<0.001). In the discovery sample during emotional regulation: dlPFC activity (OR:4.27, CI:2.09-8.72, qFDR < .001), precuneus-dACC FC (OR:1.64, CI:1.06-2.53, qFDR = .046), precuneus-dlPFC FC (OR:1.67, CI:1.22-2.29, p = .002), and dlPFC-dACC FC (OR:1.49, CI:1.09-2.04, qFDR = .028) were positively associated with concurrent depression severity. These relationships were replicated in the test sample: dlPFC activity (OR:2.65, CI:1.94-3.63, qFDR < .001), precuneus-dACC FC (OR:11.22, CI:7.26-17.34, qFDR<0.001) and dlPFC-dACC positive FC (OR:2.61, CI:1.89-3.60, qFDR<0.001). These measures were also positively associated with future depression severity: dlPFC activity (OR:3.55, CI:2.52-5.0, qFDR < .001) precuneus-dACC FC (OR:6.26, CI:3.62-10.83, qFDR < .001) and dlPFC-dACC FC (OR:3.65, CI:2.34-5.68, qFDR < .001). There were no relationships among any neural measures and concurrent mania/hypomania severity in the discovery sample.

Conclusions: Identifying and replicating largescale neural network predictors of concurrent and future depression severity during working memory and emotional regulation paradigms is a step toward identifying objective markers of risk for future depressive disorders, and can provide neural targets to better guide and monitor early interventions in at-risk young adults.

Keywords: Mood Disorders, Emotional Regulation, Working Memory fMRI, Default Mode Network (DMN), Salience Network

Disclosure: Nothing to disclose.

P278. Neonatal Maternal Separation-Induced Alternation of MiRNAs and Their Possibility as Biomarkers of Major Depressive Disorder

Shuken Boku*, Hiroyuki Toda, Minori Koga, Naoto Kajitani, Minoru Takebayashi

Kumamoto University, Kumamoto, Japan

Background: As major depressive disorder (MDD) impairs a patient's life over a long period, early detection of MDD is essential for minimization of loss and suffering due to MDD. However, early detection of MDD remains difficult because of the lack of clinically useful biomarkers of MDD. A lot of studies have shown that early-life stress (ELS) is involved in the vulnerability and treatment-resistance of major depressive disorder in adults. In addition, recent studies have reported that miRNAs may be involved in the biological effects of ELS. These suggest that ELS-associated miRNA may be a potential biomarker of MDD. Therefore, here we examined the possibility of ELS-associated miRNAs as biomarkers of MDD.

Methods: The possible candidates of ELS-associated miRNAs were identified with neonatal maternal separation (NMS) in rats, a common animal model of ELS. The levels of miRNAs in peripheral blood of MDD patients (N = 64) and healthy controls (N = 75) were estimated with quantitative RT-PCR. Receiver operating characteristic (ROC) curve analyses were performed to estimate the possibility of the possible candidates of ELS-associated miRNAs.

Results: we performed the microarray analysis of miRNAs derived from peripheral blood of NMS rats and identified four miRNAs which were significantly altered in NMS rats and expressed in both rats and human. ROC analyses showed that area under the curve (AUC) of each miRNA was under 0.7000, which means that each miRNA has only limited diagnostic power of MDD. However, AUC of the combination of these four miRNAs was 0.9444 with sensitivity of 0.8750 and specificity of 0.9200, which means that the combination of these four miRNAs has the high diagnostic power of MDD.

Conclusions: The combination of the four ELS-associated miRNAs is expected as a clinically useful biomarker of MDD.

Keywords: Major Depressive Disorder (MDD), Biomarker, miRNA, Early life stress (ELS)

Disclosure: Nothing to disclose.

P279. Anhedonia is Associated With Paraventricular Nucleus of Thalamus to Nucleus Accumbens Resting-State Functional Connectivity

Bianca Leonard*, Steven Granger, Joren Adams, Liv McMillan, Michael Yassa

University of California Irvine, Irvine, California, United States

Background: The paraventricular nucleus of the thalamus (PVT) is gaining attention for its roles in integrating salient experiences,

arbitration of motivational conflict, retrieval of long-term fear memories, and behavioral regulation. The PVT responds to salient stimuli of both positive and negative valence, likely due to its reciprocal connections with the nucleus accumbens (NAc) and amygdala (AMY). Recently, our lab defined the resting state functional connectivity (RSFC) patterns of the PVT in humans using resting state fMRI and established that a similar PVT network to that found in rodents exists in humans. Interruption of the PVT-AMY-NAc circuitry in rodents is associated with depressive- and anxiety-like behavior in rodents - notably anhedonia as a function of sex. Here, we investigated if anhedonia was associated with changes in PVT-NAc and PVT-AMY RSFC.

Methods: We collected self-reported depression and anxiety scores (Beck Depression Inventory-II and Beck Anxiety Inventory), and resting-state functional magnetic resonance imaging (fMRI) scans in a human sample ($n = 63$, 48 females) that endorsed a range of depression and anxiety symptom severity. Latent factor analysis applied to the depression and anxiety symptom scores empirically differentiated symptom classes in this sample. We derived four latent factors from this analysis, including an anhedonia factor. RSFC was calculated as the time-series correlations between the activity of the PVT with the AMY and NAc. PVT-NAc and PVT-AMY FC were correlated with the anhedonia factor scores using Spearman's rank correlation coefficient.

Results: PVT-NAc FC was positively associated with anhedonia factor scores in participants with psychiatric symptoms (PS, $R = 0.34$, $p = 0.018$), but not in participants without psych symptoms (NPS, $R = 0.23$, $p = 04$). Males had a significant positive relationship between PVT-NAc FC and anhedonia ($R = 0.65$, $p = 0.0082$), but females did not ($R = 0.13$, $p = 0.38$). PS males had the strongest correlation between PVT-NAc FC and anhedonia factor scores (0.77, $p = 0.0054$) where females did not quite reach significance ($R = 0.3$, $p = 0.075$). PVT-AMY and anhedonia factor scores did not correlate significantly among any of these analyses.

Conclusions: These findings suggest that changes in PVT-NAc circuitry may relate to severity of reported anhedonia and support the use of latent factor analysis to better understand patient symptom reporting and categorization. Future work will include analysis to investigate if this is occurring in a sex-specific way and test remaining factors collected in the study with PVT FC.

Keywords: Paraventricular Nucleus of the Thalamus, Resting State Functional Connectivity, Anhedonia, Sex-specific Effects, Latent Factor Analysis

Disclosure: Nothing to disclose.

P280. Dorsal Attention Network Neural Activity During Explore-Exploit Decision Making is Linked to Mood-Dependent Impulsivity in Older Adults

Angela Ianni*, Beatrice Langer, Michael Hallquist, Alexandre Dombrovski

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: The decision to attempt suicide may result from an impaired ability to select among alternative options in a state of intense negative emotions. Personality traits such as neuroticism and impulsivity have been implicated suicidal behavior, and mood-dependent impulsivity (urgency) may play an important role in the suicidal crisis, however little is known about how trait predispositions impact decision-making. Reinforcement learning provides a framework for behavioral and neural studies of the interface between personality factors and decision-making. Explore-exploit decisions are one example, where the decision is to utilize known information to take advantage of (exploit) an option known to be good or explore other lesser-known options. Optimal behavior requires adequate learning from sampled rewards and appropriate generation and comparison of alternative options. While prior work has linked suicidal behavior to impaired reward-guided learning, the neural basis of aberrant decision making in the acute suicidal crisis is not well understood.

The dorsal attention network (DAN) is involved in the voluntary orienting of visuospatial attention, allowing for generation of a set of actions that can be taken in a particular situation. The DAN is comprised of several functionally connected brain regions including the intraparietal sulcus, superior pre-central sulcus, inferior pre-central sulcus, and motion-sensitive area MT complex (MT+). Prior work in the lab has shown that these subregions play distinct roles in explore-exploit decision making behavior. Furthermore, computational models suggest importance of effective information compression to limit cognitive burden and optimize behavior in explore-exploit decision-making. Parameters to characterize reward-guided learning and decision-making in this type of task include entropy (measure of information content), change in entropy, and reward prediction error. We hypothesized that the UPPS personality dimensions negative urgency and lack of premeditation would be linked with aberrant behavior and DAN neural activity during explore-exploit decision making.

Methods: 146 cognitively intact individuals, aged 49-80 years, were recruited from four groups: healthy volunteers and three groups with depression (previously attempted suicide, suicidal ideation but no suicide attempts, and no suicidal ideation or suicide attempts). Individuals completed several cognitive and personality assessments including the UPPS-P impulsive behavior scale. Subscale dimensions of negative urgency (tendency to act rashly under extreme negative emotions) and lack of premeditation (tendency to act without thinking) were used for imaging analyses due to hypothesized association with suicidal behavior.

Individuals also completed an explore-exploit "clock task" during an fMRI scan, where action values varied along a continuous interval marked by visuospatial and time cues. Participants were instructed to explore the interval extensively to discover the most rewarding options. We utilized the SCEPTIC selective maintenance reinforcement learning model, previously developed in the lab, to calculate trial-by-trial action values across discretized intervals of time, reward prediction errors, entropy (measure of information content), and change in entropy. We used the Matlab VBA toolbox for model fitting and extracted trial-by-trial parameter values for model-based fMRI analyses. fMRI images were pre-processed and then extracted signal from four DAN subregion ROIs (premotor, caudal posterior parietal cortex, rostral posterior parietal cortex, and MT+) was deconvolved using a leading hemodynamic deconvolution algorithm to estimate neural activity, which was stimulus locked and averaged across trials. Multilevel modeling implemented in R was used to test for significance between the model-derived behavioral parameters and neural data, with significance thresholding of $p < 0.05$, FDR corrected.

Results: We found a positive effect of UPPS negative urgency on the neural response to increases in the number of potentially useful options in all regions of the DAN (all $p < 0.05$, FDR corrected) with the greatest effect in the caudal posterior parietal cortex. This effect remained significant in all regions when controlling for age, education, total UPPS, and anxiety diagnosis. Furthermore, we found the negative urgency was also positively correlated with responses to unsigned prediction error (surprise) in the caudal posterior parietal cortex and MT+ but not in other DAN regions when controlling for age, education, total UPPS, and anxiety diagnosis. Mood-independent impulsivity measured by UPPS lack of premeditation did not modulate DAN encoding of reinforcement.

Conclusions: Mood-dependent impulsivity is associated with exaggerated responses to reinforcement when the best option is harder to identify. This enhanced sensitivity to global uncertainty

could represent a neural mechanism underlying aberrant decision-making in the suicidal crisis.

Keywords: Explore-exploit Dilemma, Computational Models of Decision-making, Impulsivity, Suicide

Disclosure: Nothing to disclose.

P281. Dimensional Antidepressant Response to Repetitive Transcranial Magnetic Stimulation Can Be Predicted Using Pretreatment Resting-State Functional Connectivity

Benjamin Wade*, Tracy Barbour, Kristen Ellard, Joan Camprodon

Massachusetts General Hospital, Charlestown, Massachusetts, United States

Background: Repetitive Transcranial Magnetic Stimulation (rTMS) is an effective treatment for depression. To date, however, few studies have evaluated whether antidepressant response to rTMS is predictable from pretreatment neuroimaging measures. Depression is, moreover, a symptomatically heterogenous disorder. Previous work from our group has demonstrated that prospective prediction of antidepressant response to other antidepressant treatments (i.e., ketamine and electroconvulsive therapy) is improved by predicting symptom changes along latent dimensions of depression rather than changes in the overall severity of depressive symptoms. Here, we compared the performance of machine learning models using pretreatment patterns of resting-state functional connectivity (RSFC) to predict rTMS-related symptom changes along the total score of the 17-item Hamilton Depression Rating Scale (HDRS), the HDRS-6 subscale, and three previously identified latent dimensions of the HDRS: core mood and anhedonia (CMA), somatic disturbances (SoD), and insomnia. We hypothesized that predictions made for changes in the CMA dimension and the HDRS-6 would be more accurate than those for the HDRS-17 total score.

Methods: Patients ($n = 26$; mean age [SD] = 41.1 [14.1] years; percent male=50%) received rTMS to the left DLPFC. Stimuli were delivered at 120% resting motor threshold at 10 Hz (3000 stimuli per session, 36 sessions total). All patients underwent resting-state fMRI (rs-fMRI) scans within 1 week of their first rTMS session and depressive symptoms were assessed with the HDRS-17 every 2 weeks, including the first and last day of treatment. Pretreatment rs-fMRI scans were parcellated into 200 regions using the Schaefer atlas. For each subject, regional global connectivity values were computed using the graph theoretic node degree, calculated as the sum of pairwise correlations surviving a Fisher Z-transformed correlation threshold ($Z \geq 0.4$). Regional global connectivity values were used as predictive features in random forest regression (RFR) models to predict changes along the HDRS-17, HDRS-6, CMA, SoD, and insomnia outcome measures. All RFR models were trained and validated using 10-repeated 10-fold cross validation with a nested grid search for parameter optimization. The performance of models was evaluated based on the coefficient of determination (i.e., the fraction of explained variance) in the out-of-sample test data. The significance of each model's fit was assessed using permutation tests ($B = 1000$ permutations) and multiple comparisons were adjusted across the set of all outcome measures using a Bonferroni correction, yielding a critical value of $0.05/5 = 0.01$.

Results: The mean coefficient of determination (i.e., fraction of explained variance) in the prediction of change in the CMA dimension was significantly above zero ($R^2 = 0.19$, $q = 0.005$) and was significantly higher than the HDRS-17 total score ($R^2 = -0.07$). Elevated global connectivity of a division of the right somatomotor network (SMN), right dorsal prefrontal cortex division of the default mode network (DMN), and right precuneus/posterior cingulate DMN division all predicted poorer symptom reduction while elevated connectivity components of the right dorsal attention network (DATTN) predicted more reduced CMA symptoms following rTMS. Changes along other HDRS subscales were not predicted above chance levels (all $q > 0.05$).

Conclusions: Our findings support that the pretreatment global connectivity of components of the DMN, SMN, and DATTN are predictive of dimensional antidepressant response to rTMS. Previous reported on DMN connectivity as predictive of antidepressant response in other pharmacological and behavioral treatments. Interestingly, changes in core mood symptoms (depressed mood, interests, psychomotor retardation, and weight loss) were predicted more accurately than those for somatic or sleep disturbance symptom clusters. These findings align with related work showing improved prediction of treatment outcomes using latent dimensional outcomes in ketamine and electroconvulsive therapy. Future work will seek to replicate these findings in a larger sample.

Keywords: Repetitive Transcranial Magnetic Stimulation, Depression, Resting-state Functional Connectivity, Machine Learning, Symptom Dimensions

Disclosure: Nothing to disclose.

P282. Deriving Candidate TMS Targets for Bipolar Disorder With Brain Lesions Causing Mania or Depression

Joseph Taylor*, Summer Frandsen, Amit Anand, Faith Gunning, David Silbersweig, Katherine Burdick, Amy Brodtmann, Maurizio Corbetta, Gonçalo Cotovio, Natalia Egorova, Sophia Gozzi, Jordan Grafman, Andrew Naidech, Albino Oliveira-Maia, Thanh Phan, Joel Voss, Michael Fox, Shan Siddiqi

Brigham and Women's Hospital, Harvard Medical School, Boston, Massachusetts, United States

Background: Patients with bipolar disorder need better treatments. Transcranial magnetic stimulation (TMS) shows promise, but the optimal brain regions to stimulate are unknown. In this study, we derived a map that is causally implicated in mood valence based on the functional connectivity of brain lesions causing mania versus depression.

Methods: We combined seven independent lesion datasets for mania and depression into a single model. First, we estimated the connectivity of each lesion location using a normative connectome ($n = 1000$). Second, we created a mood valence map from the differential functional connectivity patterns of mania versus depression lesions. Third, we used permutation testing to determine whether the absolute mean voxel value in the mood valence map was stronger than chance (10,000 permutations). Fourth, we assessed specificity of the mood valence map with control lesions not associated with mood disturbance. Finally, we assessed whether the average coordinates of common TMS targets (i.e., bilateral 5 cm, anti-subgenual, dorsomedial) were preferentially connected to the mood valence map.

Results: Mania is more likely to be associated with lesions functionally connected to right prefrontal cortex, and depression is more likely to be associated with lesions functionally connected to left prefrontal cortex. This mood valence map was stronger than expected by chance ($p < 0.05$) and did not change when multiple control lesions were added into the model (spatial correlation $r > 0.99$). The left anti-subgenual TMS target was preferentially connected to the negative valence map ($p < 0.01$), and the right dorsomedial prefrontal cortex was preferentially connected to the positive valence map ($p < 0.005$).

Conclusions: Mood valence shows left-right frontal lobe asymmetry, consistent with existing literature. However, the

topography of this asymmetry could guide optimization of TMS treatment targets for bipolar depression versus mania. Future analyses will search for a candidate treatment target functionally connected to both mania and depression lesions, which may be relevant for mood stabilization.

Keywords: Bipolar Disorder, Brain Networks, Lesion, TMS Targeting, Human Connectome

Disclosure: Nothing to disclose.

P283. Decrease in Calculated Brain Age Following ECT

Michael Henry*, Tracy Barbour, Kristen Ellard, James Luccarelli, Isabella Conner, Amanda Yang, Joan Camprodon

Massachusetts General Hospital, Boston, Massachusetts, United States

Background: The cognitive side effects of electroconvulsive therapy (ECT) have long caused concerns that it causes damage to the brain. Instead, structural imaging studies have found that brain regions that exhibit plasticity show an increase in tissue volume following a course of ECT. Recently, machine learning based algorithms have been developed that use regional brain volumes to estimate the person's physiologic brain age. We hypothesized that the increase in volume seen following ECT in depressed individuals would be sufficient to decrease the brain age calculated by the BrainAgeR machine learning based algorithm. We report the results of this pilot study below.

Methods: Structural magnetic resonance imaging (MRI) scans from 44 subjects with major depressive disorder (MDD) who had undergone imaging as part of other studies on the imaging changes induced by ECT were assessed for inclusion in this analysis. 37 subjects, aged 19-65, 20 males, were found to have completed a pretreatment and post treatment scan suitable for analysis. Depression rating scales obtained prospectively included the Quick Inventory of Depressive Symptomology (QIDS) and the 28-item Hamilton Depression Rating Scale (HAMD-28). Physiologic brain age was estimated for each scan using the BrainAgeR algorithm. Since we had a priori hypothesized that calculated brain age would decrease, the results were analyzed using a one-tailed paired t-test. Given the exploratory nature of this study, the results were not corrected for multiple comparisons.

Results: Across participants, there was a significant decrease in estimated physiologic brain age after a course of ECT (mean change = -0.79 years, $p = 0.037$). When stratified according to clinical response, the within-group change in the responder and non-responder groups did not reach significance. When stratified by remitters versus non-remitters, for both the QIDS and the HAMD-28, the non-remitter group showed a significant decrease in calculated age (QIDS: mean change = -0.99 years, $p = 0.018$; HAMD-28: mean change = -0.95 years, $p = 0.033$), while the remitters did not.

Conclusions: These data suggest that the BrainAgeR algorithm for calculating physiologic brain age is sufficiently sensitive to detect the effects of an acute treatment for depression. Also, consistent with our hypothesis, the mean calculated physiologic age decreased following ECT in a cohort of depressed subjects. When analyzed according to clinical response, the subjects who did not remit showed a statistically significant decrease in calculated age while the remitters did not. Given the small sample size and the preliminary nature of these findings, replication in a larger study sample is indicated.

Keywords: Brain Age, Electroconvulsive Therapy, MRI

Disclosure: Roche Pharmaceuticals: Employee (Spouse)

P284. Depression as a Disease of White Matter Network Disruption: Characterizing the Relationship Between White Matter Lesions and Depression in Patients With Multiple Sclerosis

Erica Baller*, Matthew Cieslak, Timothy Robert-Fitzgerald, Sydney Covitz, Melissa Martin, Matthew Schindler, Amit Bar-Or, Ameena Elahi, Michael Fox, Abigail Manning, Clyde Markowitz, Nebojsa Mirkovic, Christopher Perrone, Victoria Rautman, Donovan Reid, Guy Schultz, Shan Siddiqi, Sunil Thomas, John Detre, Russell Shinohara, Theodore Satterthwaite

University of Pennsylvania, Philadelphia, Pennsylvania, United States

Background: Multiple sclerosis (MS) is an immune-mediated neurological disorder that affects one million people in the United States, with up to 50% of patients experiencing a lifetime depression. However, mechanisms of depression in MS remain under-investigated. Previous research using lesion network mapping has demonstrated that strokes in gray matter associated with depression disrupt a reproducible depression network. However, such methods have not been used to investigate how white matter lesions (WMLs) relate to depression in MS. This study aims to define how depression in adults with MS is associated with white matter lesion (WML) location and burden in a retrospective sample.

Methods: Participants with MS were identified from the electronic medical record. The depressed individuals (DI) included persons with evidence of depression as indicated by a ICD-10 depression diagnosis (F32-F34.*), a prescription for antidepressant medication, or screening positive via PHQ2/9 ($n = 232$, age (SD) = 49 (12), % females = 86). The age- and sex-matched non-depressed comparators (NDC) included persons with no prior depression diagnosis, psychiatric medications, and were asymptomatic on PHQ2/9 ($n = 148$, age (SD) = 47 (13); % females=79). Structural MRI was obtained as part of routine care at 3 T using a research-quality protocol. WMLs were automatically segmented using the algorithm Method for Inter-Modal Segmentation Analysis and projected onto a standard template. Seventy-seven white matter tracts (WMT) were evaluated. The volume of WMTs intersecting each lesion was computed via streamline filtering in DSI Studio. Total volume of lesions irrespective of tract was also calculated and compared between diagnostic groups. Age and diagnostic effects were assessed with general linear models and T-tests. Enrichment of effects in a previously described depression network by Siddiqi et al., 2021, was also evaluated. Multiple comparisons were controlled using the false discovery rate ($Q < 0.05$).

Results: Streamline filtering recapitulated previously known patterns of MS disease, with high proportions of streamlines impacted in the optic radiations, inferior fronto-occipital fasciculi, medial longitudinal fasciculi, and corticopontine tracts. Greater age was associated with higher disease burden in 54/87 WMTs ($P_{FDR} < 0.05$). Total lesion volume was not significantly different between the two groups ($P = 0.07$, NS). However, when using streamline filtering, DIs had a higher mean disease burden across all WMTs ($P < 0.05$, Cohen's $d = 0.17$), which was driven by a greater burden of disease within the depression network.

Conclusions: We present a novel approach for calculating the relationship of WML to depression disease burden. We demonstrate that overall lesion burden, irrespective of location, does not differ between diagnostic groups. Rather, when looking at disease burden in streamlines that intersect with the lesions, we show that DIs have greater overall disease burden as compared NDCs, which is driven by a higher burden of disease in fibers that connect areas of the depression network. Future work using white matter lesion

network mapping in MS could also advance our understanding of the mechanisms of depression more broadly.

Keywords: Depression, Multiple Sclerosis, Electronic Medical Record, Brain MRI

Disclosure: Nothing to disclose.

P285. Tianeptine Improves Anticipation for Reward in a Mouse Model of Early Developmental Exposure to Fluoxetine

Emily Cambre, Elena Christenfeld, Gabriella Sahyoun, Jonathan Javitch, Sarah Canetta*

Columbia University, New York, New York, United States

Background: Deficits in reward processing, commonly referred to as anhedonia, are a hallmark of multiple psychiatric disorders. In the context of depression, anhedonia is one of two core symptoms needed for diagnosis, and often predicts more severe outcomes. Although selective serotonin reuptake inhibitors (SSRIs) are the frontline treatment for depression, they are ineffective for many individuals, and in certain cases can even exacerbate symptoms including anhedonia. Therefore, there is a clear gap in our understanding of how antidepressants affect reward processing, and why they may be variably effective across individuals.

Methods: Male and female pups on a C57BL/6 J (C57) background were injected with either FLX (10 mg/kg, i.p) or saline from postnatal (PN) day 2 to 11. Motivation and hedonic perception were measured in adult PN FLX or vehicle (PN VEH) exposed mice using a progressive ratio (PR) and lickometer task, respectively. During PR testing, mice were food restricted to 90% of their baseline weight and during lickometer testing, mice were maintained on ad libitum citric acid water with food removed 12 hours prior to testing. Cages with co-housed PN FLX and PN VEH mice were subsequently randomized to receive either chronic FLX or vehicle control (water) as adults. FLX (18 mg/kg) was administered through the drinking water for three weeks and then continuously throughout behavioral testing. A two-week washout period followed completion of post-FLX behavioral testing before beginning TIA administration. Mice that had previously received FLX, received TIA and the mice receiving control remained the same. A 30 mg/kg solution of TIA NaCl dissolved in 0.9% sterile saline, or saline as a control, was injected intraperitoneally twice per day for 14 days. TIA injections continued throughout behavioral testing.

Results: We found that as adults, PN FLX animals show decreased motivation to pursue rewards, manifest as a decrease in total presses (Mann-Whitney, $p = 0.0017$, $n = 31$ PN VEH, 29 PN FLX), session time (t-test, $p = 0.0011$, $n = 31$ PN VEH, 29 PN FLX), break point (Mann-Whitney, $p = 0.0018$, $n = 31$ PN VEH, 29 PN FLX) and rewards retrieved (t-test, $p = 0.0475$, $n = 31$ PN VEH, 29 PN FLX). These mice also show decreased latency to approach the lever and begin pressing (Mann-Whitney, $p = 0.0005$, $n = 31$ PN VEH, 29 PN FLX), which may reflect decreased reward anticipation. Strikingly, in the lickometer test, there were no differences in the number of times the PN FLX and PN VEH mice licked the freely available reward (Mann-Whitney, $p = 0.9796$, $n = 31$ PN VEH, 29 PN FLX) suggesting no change in hedonic perception. Interestingly, we found that adult FLX administration did not alter any of these variables in PN FLX mice. By contrast, adult TIA administration increased reward anticipation (Mann-Whitney, $p < 0.0001$, $n = 14$ PN FLX-SAL, 15 PN FLX-TIA) but did not consistently alter variables associated with motivation including total presses (Mann-Whitney, $p = 0.1456$, $n = 14$ PN FLX-SAL, 15 PN FLX-TIA), session time (t-test, $p = 0.8056$, $n = 14$ PN FLX-SAL, 15 PN FLX-TIA), break point (Mann-Whitney, $p = 0.1486$, $n = 14$ PN FLX-SAL, 15 PN FLX-TIA) and rewards retrieved (t-test, $p = 0.8641$, $n = 14$ PN FLX-SAL, 15 PN FLX-TIA).

Conclusions: Our results confirm prior work demonstrating that early developmental exposure to FLX in mice, during a period of brain development occurring during the third trimester in a human, produces adult animals that show decreased motivation in adulthood. We expand on this phenotype to show these effects also include reward anticipation, while reward learning and hedonic perception remains intact. Our results also demonstrate that these deficits in motivation and reward anticipation in PN FLX animals are resistant to subsequent FLX administration in adulthood. By contrast, the atypical antidepressant TIA improves reward anticipation, although motivation remains unaltered. Overall, these findings suggest that PN FLX mice may represent a model of SSRI-resistant affective behavioral changes and that TIA may be a promising alternative treatment from SSRIs for humans with suspected in utero early developmental exposure to SSRIs, and more broadly in a subset of people with SSRI-resistant depression.

Keywords: Reward Processing, SSRI, Tianeptine

Disclosure: Nothing to disclose.

P286. Early and Late Predictors of Depression Treatment Response in the Clinic: An Observational Study

Kathryn Ridout*, Mubarika Alavi, Samuel Ridout, Constance Weisner, Esti Iturralde

The Permanente Medical Group, Santa Rosa, California, United States

Background: Major depressive disorder (MDD) is a chronic, recurrent illness impacting 20.6%1 of the U.S. population, causing significant disability, and costing $326.2 billion annually. Patient risk for subsequent depressive episodes increases from 50% to 90% after the first compared to the third episode. Incomplete depressive episode treatment response is the strongest predictor of depression relapse and reoccurrence and increases the likelihood of work productivity loss and disability. As such, improving treatment response during a patient's first clinical presentation for a depressive episode would have the strongest impact on decreasing depressive disorder prevalence, chronicity, and associated disability. Evidence suggests that MDD treatment response is improved with early detection, intervention, and appropriate treatment. However, treatment response varies between patients, and, to date, there are no tools to predict which patients will respond to treatment. This study aimed to examine clinical predictors of depression treatment response early and later in the course of treatment.

Methods: Retrospective, observational study in a large, integrated health system. Participant inclusion criteria include age ≥ 18-years-old, a Patient Health Questionnaire-9 score of ≥ 5, and seeking depression outpatient care between 3/2020-12/2021. Exclusion criteria include acute suicide risk, diagnoses of bipolar, psychotic, dementia, or active substance use disorders, current hospice, or home-based palliative care, residing in a skilled nursing or assisted living facility, or non-health plan member. Study variables were extracted from the electronic health record at 6 weeks and 6 months after starting depression treatment. Predictors of depression treatment response were chosen based on prior literature. Based on the previous literature and our sample size, we estimated power to be able to detect small effect sizes (f2 of 0.02).

Results: Of the 27,858 patients, 68% were female, 48% were White, 19% were LatinX, 14% were Asian, 8% Black, and 11% other race. The majority of patients were age 18-39 (61%), followed by 40-59 (27%), 60-69 (8%), then 70 or older (4%); neighborhood deprivation index was equally distributed across categories (23% to 26%). Most patients had a Charleson comorbidity index of 0 (84%) and a body mass index of ≤24.9 (22%). The mean baseline

PHQ-9 was 10.7 ± 3.9; the average wait time to first treatment appointment was 2.4 ± 5.2 days. Most patients have no history of outpatient mental health treatment (63%) or antidepressant therapy (79%) in the year prior. EHR predictors significant at both timepoints included baseline PHQ-9 score; previous psychiatry referral; patient outreach to psychiatry; outpatient psychiatric encounter for condition other than depression; previous antidepressants for disorders other than depression; gender; age; and BMI ($P < .0001$ for all). EHR predictors that differed by timepoint included race (lower treatment response at 6 months for LatinX populations; $p < .0001$); Charlson comorbidity index (lower response at 6 months with higher CCI; $p < .0001$); neighborhood deprivation index (lower response at 6 weeks for higher deprivation; $p < .0001$); and antidepressant start during depression treatment (higher response at 6 months with antidepressant; $p < .0001$).

Conclusions: While many clinical variables predicted depression treatment outcomes both early and later in the course of treatment, some variables appeared to differentiate treatment response at each time point. This study provides the foundation for future work examining utilization of treatment course predictors in prospective treatment decision making.

Keywords: Treatment Outcome Prediction, Personalized Medicine, Depression Treatment Response

Disclosure: Nothing to disclose.

P287. Melancholic Versus Non-Melancholic Features in Bipolar I Disorder

Jake Stenzel*, Tim Bigdeli, Michele Pato, Carlos Pato, Ayman Fanous

University of Arizona College of Medicine-Phoenix, Phoenix, Arizona, United States

Background: Melancholic features, as a specifier for Major Depressive Episode in both major depressive disorder and bipolar I disorder (BD-I), is characterized by depressive episodes with marked psychomotor disturbances, anhedonia, weight loss, and feelings of guilt as defined in the Diagnostic and Statistical Manual of Mental Disorders, 5th ed. (DSM-V) (American Psychiatric Association, 2013). While the validity of melancholia as a diagnostic concept has been widely debated, there is evidence to suggest that melancholia exhibits differences in pathophysiology and severity of disorder. Such distinctions include increase hypothalamic-pituitary-adrenal axis dysregulation, pro-inflammatory cytokines IL-5 and IL-4, suicide risk, and psychotic features.

While the majority of investigations have focused on melancholia in the context of depression, our study investigated melancholic features amongst a patient population with BD-I from the Genomic Psychiatry Cohort (GPC) (Pato et al., 2013). The aims our study were to identify the prevalence of melancholic features in a large population of patients with BD-I ($n = 4025$) and compare clinical features and socio-demographic characteristics between melancholic and non-melancholic BD-I patients.

Methods: The GPC has gathered a large cohort of patients with schizophrenia, bipolar disorder, and healthy controls from across the United States and select sites abroad. All enrolled participants were asked to complete a screening questionnaire to assess their psychiatric history. Participants enrolled as probable cases were interviewed by mental health professionals using the Diagnostic Interview for Psychosis and Affective Disorders (DI-PAD). Information on ascertainment and diagnosis of the study population are provided in a previous publication (Bigdeli et al., 2020).

Diagnosis of psychiatric disorders was obtained from the DI-PAD. Patients were included in this study if they completed all necessary DI-PAD and screener material needed for each analysis and had a diagnosis of bipolar I disorder (with or without psychosis). DSM-V criteria for melancholic features were assessed using the DI-PAD and screening questionnaire. Socio-demographic factors and severity of illness were derived from the DI-PAD and screening questionnaire.

Results: Participants with a diagnosis of BD-I who completed the DI-PAD and screener questionnaire for all measured variables resulted in a N of 4025). Those fulfilling criteria for melancholia comprised 74.04% ($n = 2980$). Compared to non-melancholic BD-I, melancholic BD-I was more prevalent in females (77.05% vs 70.52%, $OR = 1.40$; $p < 0.0001$). Having a family history of psychiatric disorders was associated with melancholic BD-I as compared to non-melancholic BD-I (67.15% vs 61.05%, $OR = 1.30$; $p < 0.001$). In addition, melancholic BD-I was significantly associated with having suicidal ideation lasting at least one week (79.73% vs 60.86%, $OR = 2.55$; $p < 0.0001$) and suicidal ideation lasting greater than one month (69.62% vs 23.12%, $OR = 7.62$; $p < 0.0001$) when compared to non-melancholic BD-I. No significant association was found with the presence of psychosis, nicotine use (defined by having smoked over 100 cigarettes in their lifetime), or history of recreational drug use.

Conclusions: When compared to non-melancholic BD-I, patients with melancholic BD-I were more likely to be female and to have a family history of psychiatric disorders. Melancholic BD-I patients were also found to have a marked increase in periods of suicidal ideation lasting greater than one week and greater than one month. This observation is in accordance with previous studies on melancholic features in major depression (Dold et al., 2021; Tondo et al., 2020). Our study relied on DSM-V criteria for determination of melancholic features, whereas alternative instruments may have resulted in varied prevalence. Additional exploration of comorbidities and both GWAS and NGS data are planned and may provide further insight into melancholic features in BD-I.

Keywords: Bipolar I Disorder, Psychiatric Disorders, DSM-5, Clinical Psychiatry

Disclosure: Nothing to disclose.

P288. Single Nucleus RNA Sequencing of Mouse Ventral Tegmental Area Neurons Following Input-Specific Stimulation Reveals Differential Activation Patterns

Rhiana Simon, Joshua Yee, Koichi Hashikawa, Garret Stuber, Larry Zweifel, Marta Soden*

University of Washington, Seattle, Washington, United States

Background: The ventral tegmental area (VTA) is home to a heterogeneous population of dopamine neurons as well as significant populations of glutamatergic and GABAergic cells. The VTA receives inputs from over two dozen brain regions. We found previously that specific inputs differentially innervate VTA subregions, and stimulation of select inputs leads to varying behavioral outcomes and distinct spatial patterns of cellular activation, as measured by Fos protein expression. This implies that different subsets of VTA neurons are activated by distinct inputs to drive different behavioral modalities, but the specific genetic identity of these activated neurons is not known.

Methods: To address this question, we used Cre-dependent viral expression of channelrhodopsin (ChR2) to stimulate three distinct VTA inputs in the mouse: GABAergic inputs from the lateral hypothalamus (LH), GABAergic inputs from the nucleus accumbens (NAc), and glutamatergic inputs from the prefrontal cortex (PFC). Male and female Vgat-Cre (Slc32a1) or Vglut1-Cre (Slc17a7) mice were injected with AAV1-FLEX-ChR2-YFP (or AAV1-FLEX-YFP as a control) in one of the indicated regions and fiber optics were implanted above the VTA. Following 20 Hz blue light

stimulation we collected tissue and performed single-nucleus RNA sequencing (snRNAseq). We integrated the sequencing data from all four groups to generate a comprehensive analysis of VTA cell types based on differential gene expression and examined expression of immediate early genes (IEGs) to determine which cell clusters were preferentially activated by stimulation of each input. Spatial expression of a selection of marker genes was validated using multi-plex in situ hybridization.

Results: From approximately 40,000 cells sequenced we identified over 9,000 neurons with high quality gene expression data. These neurons segregated into 3 clusters of primarily dopaminergic cells, identified by expression of canonical markers such as Th, Slc6a3 (DAT), and Ddc. We also identified multiple clusters of primarily GABAergic cells, clusters of primarily glutamatergic cells, and one major cluster that expressed a mix of GABAergic, dopaminergic, and glutamatergic markers. These clusters showed differential expression patterns of critical genes important for determining cellular physiology and connectivity, including ion channels and neurotransmitter and neuropeptide receptors. We confirmed spatially distinct expression patterns of a selection of marker genes using multi-plex in situ hybridization and found that the three dopaminergic clusters tended to segregate along the lateral to medial axis, consistent with established differences in function between known dopaminergic subpopulations. Stimulation of LH-GABA, NAc-GABA, or PFC-glutamate inputs all led to increased IEG levels compared to control mice. Analysis of the percentage of cells in each cluster that showed IEG expression revealed unique patterns of cluster activation for each stimulation group compared to control (Fisher's exact test, $P < 0.05$). Notably, the patterns of dopamine cell cluster activation were consistent with the spatial innervation patterns of each input (i.e., only LH-GABA inputs, which innervate the lateral VTA, induced significant activation in the most lateral dopamine cluster). We also observed variability in IEG expression in VTA glutamatergic clusters, with NAc-GABA inputs inducing the most activation.

Conclusions: This data set represents a large and comprehensive single-nuclear sequencing analysis of the mouse VTA. Analysis of differentially expressed genes between identified cell clusters provides valuable information regarding the organization of this critical brain region and the cellular properties and functions of specific neuronal subsets. We also provide proof-of-concept of a novel approach to analyzing activation of cell clusters following optogenetic stimulation and demonstrate differential activation of VTA cell types by distinct inputs. These findings advance our understanding of the circuit architecture governing inputs to the midbrain dopamine system and the regulation of dopamine-dependent behaviors.

Keywords: Dopamine, Ventral Tegmental Area (VTA), RNAseq

Disclosure: Nothing to disclose.

P289. Analysis of Mitochondrial DNA Variants and Their Association With Brain Structural Measures in Bipolar Disorder

Ana Mendes-Silva*, Suyi Shao, Lucas Taniguti, Mikaela Dimick, Clement Zai, James Kennedy, Benjamin Goldstein, Vanessa Gonçalves

Centre for Addiction and Mental Health, Toronto, Canada

Background: Mitochondria are the main energy source for neurons and other brain cells and play crucial roles in various neural processes, such as neurogenesis, neuroplasticity, and neurotransmission. The dysfunction in those organelles may alter critical neuronal processes underlying abnormal brain development and cognitive impairment in psychosis because of a lack of energy and higher levels of inflammatory molecules. Several clinical, genetic, and neuroimaging studies implicate mitochondrial dysfunction to play a crucial role in bipolar disorder (BD) pathophysiology. The present study aimed to characterize the mtDNA variants in youth diagnosed with BD and investigate the association between two common mitochondrial DNA (mtDNA) variants with structural brain changes.

Methods: We included 96 European-Caucasian adolescents (54 BD and 42 healthy controls (HC)), within the age range 13-20, from both sexes. Psychiatric diagnoses were determined based on semi-structured diagnostic interviews. Mitochondrial DNA was extracted from saliva and the MiSeq platform was used to sequence the samples. mtDNA variants were identified using the mtDNA-Server pipeline and common variants were selected with a minor allele frequency (MAF) of at least 5%. Functional analysis was performed by Mutserve to identify mtDNA variants potentially harmful. The MutPred, Selection Score and Mito tool algorithms assigned for each potentially harmful variant identified by functional analysis were summed to build a pathogenicity score. Further, the pathogenicity score and individual variants were tested with the phenotypes using logistic regression in R. Subcortical volumes, cortical thickness and cortical surface area were determined for a-priori determined regions of interest (ROIs) (anterior cingulate cortex (ACC) and amygdala) and associations with each mtDNA variants and pathogenic score were tested using general linear model in SPSS, adjusting for age, sex, and intracranial volume (ICV) when needed. To account for multiple tests, a Bonferroni-corrected significance threshold of 0.0125 was applied.

Results: A total of 1382 homoplasmic variants were identified in our sample, of which 67 were common variants (MAF > 0.05). Functional analysis revealed that 9 of the 67 common variants were non-synonymous, and the variants (MT-ND2):m.4917 A > G, and (MT-ND1):m.4216 T > C were classified as potentially harmful. Logistic regression analysis showed the association of mtDNA pathogenicity score and 4216 T > C variant with BD (OR: 1.57 [95% CI: 1.04, 2.38], $p = 0.031$; and OR: 1.99 [95% CI: 1.04, 3.81], $p = 0.037$, respectively). Regression analysis, adjusted for sex and age, showed that both pathogenicity score and the individual variant (MT-ND1):m.4216 T > C were nominally associated with amygdala volume in the whole cohort (ß43.07, p0.037, and ß82.78, p0.024, respectively). No effect of pathogenicity score or individual variants were found for cortical thickness, surface area or volume in ACC. None of these associations were significant after adjustment for multiple testing.

Conclusions: Our findings indicate that youth with BD have a greater presence of the two reported pathogenic complex I variants m.4917 A > G and m.4216 T > C. Moreover, higher pathogenicity score and the variant m.4216 T > C were associated with greater amygdala volume, which could reflect neuroinflammation caused by mitochondrial dysfunction. Further studies in other populations are required to support these findings.

Keywords: Mitochondria, Mitochondrial DNA, Bipolar Disorder, Youth, Amygdala

Disclosure: Nothing to disclose.

P290. RNA Sequencing of the Limbic System in Major Depressive Disorder

Fernando Goes*, Leonardo Collado-Torres, Peter Zandi, Joel Kleinman, Daniel R. Weinberger, Ran Tao, Daniel Weinberger, Thomas Hyde

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

Background: Major Depressive Disorder (MDD) is the second leading cause of disability worldwide and a significant risk factor for suicide. Novel treatments are urgently needed, yet the development of new types of antidepressants has been hindered

by limited understanding of the pathophysiology of MDD. In the current study, we perform the largest and most comprehensive molecular study of MDD in post-mortem brain samples

Methods: We have performed expression profiling on a large cohort of post-mortem brains with MDD and healthy controls. RNA sequencing using a ribozero protocol (median depth 132 million reads) was performed comprising a total of 432 males and female samples from the Anterior Cingulate Cortex (ACC) and 429 samples from the Amygdala. Differential expression analyses were conducted using limma voom, while accounting for typical post-mortem confounds, including a measure of differential susceptibility to degradation. Control for multiple testing was performed using a false-discovery rate of 5%.

Results: A total of 630 and 106 genes were differentially expressed in the ACC and Amygdala respectively. The mean fold change was modest (OR ~ 1.1) and the most highly differentially expressed gene in both the ACC and Amygdala was FUS, an RNA-binding protein previously associated with neurodegeneration. Moreover, using a transcriptional-wide association study approach in the Anterior Cingulate and Amygdala, we have found 88 genes meeting Bonferroni levels of significance and 309 genes meeting an FDR < 5% level of significance. Among the Bonferroni corrected genes are several previously associated at GWAS significance levels (for example, SORCS3, ZKSCAN7, LIN28B, RMT61A, RANGAP1, FADS1, TMEM258, FNIP2, NRG1, among many others) as well as numerous novel genes, such as SIRT1, which was identified in a GWAS of severe MDD in China and now finds convergence evidence for association in a European Ancestry sample

Conclusions: Major Depressive Disorder is associated with several hundred differentially expressed genes, consistent with a large polygenic contribution across molecular features. Defining the molecular landscape of gene expression in brain regions associated with the pathophysiology of MDD may lead to the identification of novel therapeutic targets, particularly in conjunction with risk loci identified in large-scale genetic studies

Keywords: Transcriptome, Postmortem Human Brain Tissue, Major Depressive Disorder (MDD), Anterior Cingulate Cortex (ACC), Amygdala

Disclosure: Nothing to disclose.

P291. Immunometabolic Gene Pathways are Associated With Anhedonia and Altered Following a TNF-Alpha Antagonist in Patients With Depression and High Inflammation

Mandakh Bekhbat*, Ebrahim Haroon, Sarah Etuk, Jennifer Felger, Andrew Miller, Michael Treadway

Emory University, Atlanta, Georgia, United States

Background: Inflammation and altered glucose metabolism are two pathways implicated in the pathophysiology of anhedonia in major depressive disorder (MDD). These pathways are hypothesized to synergize within circulating immune cells, whereby inflammatory activation can shift metabolic demand and reprogram cellular energy sources to fuel pro-inflammatory activities, including systemic inflammation and its effects on the brain and reward processing to contribute to symptoms of anhedonia. We have previously shown relationships between symptoms of anhedonia and a whole-blood gene expression pattern consistent with increased reliance on glycolysis (as opposed to oxidative phosphorylation [OXPHOS]), a hallmark of metabolic change in activated cells, but only among MDD patients with high inflammation (plasma C-reactive protein [CRP] > 3 mg/L). We have also reported associations between combined plasma inflammatory and glucose metabolism biomarkers and reduced willingness to expend effort, a key index of motivational anhedonia, in MDD patients with CRP > 3 mg/L undergoing anti-inflammatory challenge. These findings suggest that immunometabolic shifts may contribute to, and serve as intervention targets for, anhedonia symptoms in depressed patients with high inflammation. The tumor necrosis factor (TNF)-alpha antagonist infliximab was previously found to improve symptoms of anhedonia, including effort-based motivation, selectively among MDD patients with higher inflammation. Here, we analyzed microarray data form a cohort of MDD patients with high inflammation before and after a single anti-inflammatory challenge with infliximab versus placebo in order to discover novel immunometabolic signatures for infliximab's effects on anhedonia.

Methods: N = 42 medically stable, medication-free depressed patients aged 21-65 with high inflammation (CRP > 3 mg/L) were studied before and two weeks after a single infusion of infliximab (5 mg/kg body weight) or placebo. Anhedonia was assessed using the subscale from the Inventory of Depressive Symptoms-Self Report (IDS-SR) and whole blood gene expression was profiled via the Clariom S (ThermoFisher) gene array platform at baseline and Week 2 post-infusion. Microarray data were SST-RMA-normalized and batch-corrected. Differential expression analysis was conducted using linear models for microarray data (limma) to assess treatment x time interaction effects while controlling for clinical covariates (age, sex, and race). Functional pathway enrichment was queried using the Kyoto Encyclopedia of Genes and Genomes (KEGG), and WikiPathways databases.

Results: At baseline, anhedonia severity was negatively associated with 117 gene probes ($R < -0.30$, $p < 0.01$) which were enriched for the Mitochondrial Complex I of the OXPHOS system pathway ($p < 0.05$, $q < 0.1$). Among those who received infliximab, patients who responded with >50% reduction of anhedonia had greater baseline expression of 82 gene probes which enriched IL-17 and NF-kappa B signaling pathways ($p < 0.05$, $q < 0.1$) compared to non-responders. Longitudinal gene expression and behavioral data were available from fifteen infliximab- and fourteen placebo-treated patients. At two weeks following a single infusion of infliximab, there was a significant decrease in the expression of 44 gene probes that enriched PI3K/AKT/mTOR as well as Glycolysis and gluconeogenesis pathways ($p < 0.05$, $q < 0.1$). In contrast, only 36 genes were significantly changed following placebo, which enriched broad, non-specific pathways.

Conclusions: Our results indicate that anhedonia in MDD patients with high inflammation is associated with altered energy metabolism pathways, including lower OXPHOS at baseline, which may reflect a pro-glycolytic shift characteristic of activated immune cells. In addition, increased expression of inflammatory pathways at baseline predicted reduced anhedonia scores following infliximab, consistent with our previous findings. Finally, the effects of infliximab involved reductions in glycolysis and upstream mTOR signaling pathways. These findings highlight the metabolic reprograming in circulating immune cells that are associated with systemic inflammation in MDD, which may lead to identification of new metabolic and anti-inflammatory targets for treatment of anhedonia and/or MDD with increased inflammation.

Keywords: Immune Modulation, Immunometabolism, Anhedonia, Precision Medicine for Depression, Microarray

Disclosure: Nothing to disclose.

P292. Pharmacogenomic Testing and Symptom Remission in Mood Disorders: Systematic Review and Meta-Analysis

Lisa Brown*, Joseph Stanton, Kanika Barthi, Abdullah Al Maruf, Daniel Mueller, Chad Bousman

Great Scott! Consulting, Brooklyn, New York, United States

Background: Pharmacogenomic (PGx) testing has emerged as a compelling strategy that clinicians can use to inform medication

selection and dosing, but the clinical efficacy of this strategy has been questioned. Here, we sought to systematically review and meta-analyze prospective, controlled clinical trials for an association between PGx testing and depressive symptom remission in patients with major depressive disorder (MDD).

Methods: We reviewed PubMed and the bibliographies of systematic reviews published up to July 12, 2022. We included prospective, controlled clinical trials examining the association between PGx testing and depressive symptom remission in adults and were available in English. The study adhered to the 2020 PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) reporting guidelines and was registered in PROSPERO (CRD314807). Extracted information included trial publication year, trial design, sample size, trial eligibility criteria, trial duration, participant's characteristics (i.e., age, gender, ancestry), remission measure used, and description of the genes included in the PGx test performed. Data was extracted by two independent reviewers and a third reviewer determined discrepancies. Each trial was assessed for risk of bias and a random-effects model was used to estimate pooled risk ratios. Depressive symptom remission was defined as a Hamilton Depression Rating Scale-17 score ≤ 7, Clinical Global Impression scale ≤ 2, or Patient Health Questionnaire ≤ 5.

Results: Thirteen trials including 4767 patients were analyzed, including 10 randomized controlled trials (RCTs) and three open label trials. Across all included trials, those that received PGx-guided antidepressant therapy (n = 2395) were 1.41 (95% CI: 1.15 - 1.74, p = 0.001) more likely to achieve remission compared to those that received unguided antidepressant therapy (n = 2372). Pooled risk ratios for randomized controlled trials and open label trials were 1.46 (95% CI: 1.13 - 1.88) and 1.26 (95% CI: 0.84-1.88), respectively

Conclusions: PGx testing is associated with a modest but significant increase in depressive symptom remission in adults with MDD. Heterogeneity in PGx test composition and accompanying prescribing recommendations across trials are likely contributing to uncertainty about the efficacy of PGx testing in the literature.

Keywords: Pharmacogenomics, Depression, Pharmacogenetics

Disclosure: Great Scott Consulting: Founder (Self), Tempus Labs: Employee (Self), Myriad Genetics: Stock / Equity (Self)

P293. Whole and Single Cell Human Cortical Spheroid Transcriptomics in Response to Low-Dose Ketamine

Mark Niciu*, Natalia Schmidt, William Guiler, Amanda Decker, Brian Kinnaird, Kornel Schuebel, Carlos Zarate, David Goldman

University of Iowa Health Care, Iowa City, Iowa, United States

Background: Major depressive disorder (MDD) has a ~16% global lifetime prevalence and is associated with extensive morbidity and mortality including suicide. Standard first-line antidepressant medications are effective for some but not all MDD patients, as exemplified by numerous real-world effectiveness trials including the Sequenced Treatment Alternatives to Relieve Depression (STAR*D) and the International Study to Predict Optimized Treatment for Depression (iSPOT-D). As also exemplified in these trials, first- and second-line antidepressant medications have monoaminergic mechanisms of action, so it is not surprising that MDD patients who fail a first trial are at greater risk of failing subsequent medication trials of medications due to their pharmacodynamic similarities. On the other hand, the N-methyl-D-aspartate receptor antagonist and glutamate modulator ketamine at subanesthetic doses, has rapid-acting antidepressant and anti-suicidal efficacy in such patients. The mechanisms of action of ketamine, e.g., molecular, and cellular response, have been studied in the brains of preclinical model organisms and peripheral human tissue but have yet to be reported in human central nervous system-like tissue. In this study, we report whole and single cell transcriptomic data from human stem-cell derived cortex-like spheroid/organoid cultures exposed to low-dose ketamine and its bioactive metabolites in vitro.

Methods: For stem cell lines created in the laboratory, whole blood samples were collected from subjects participating in clinical studies at the National Institutes of Health Experimental Therapeutics and Pathophysiology Branch. Peripheral blood mononuclear cells (PBMCs) were then isolated and reprogrammed into human induced pluripotent stem cells (hiPSCs) via Sendai virus-mediated transient transfection of plasmids containing Yamanaka factors, e.g., Klf4, c-Myc, Oct-3/4, and Sox2. Other validated stem cell lines, e.g., PENN025i-71-58 and STAN062i-164-2, were purchased from WiCell (Madison, WI, U.S.A.). Human pluripotent stem cells (hPSCs) were then differentiated into human cortical spheroids (hCSs), closely adhering to the methodologies pioneered by Sergiu Pasca's laboratory (Stanford University, Palo Alto, CA, U.S.A.). After reaching maturity (>40 days in vitro), hCSs were exposed to low-dose ketamine and other bioactive ketamine metabolites, e.g., 2 R,6R-hydroxynorketamine (HNK). Briefly, we followed the next-generation Ion Torrent RNA Sequencing Pipeline (ThermoFisher Scientific, Waltham, MA, U.S.A.) and the 10x Genomics Chromium Single-Cell System (10x Genomics, Pleasanton, CA, U.S.A.) for the whole and single cell RNA-Seq analyses, respectively.

Results: For the whole cell transcriptomics, no genes were ≥1.5x up-regulated by brief (1 hour) exposure to low-dose (10 mcM) racemic ketamine, relative to vehicle control. 10 genes were ≥1.5x down-regulated ($p < 0.05$) but did not survive False Discovery Rate (FDR) correction. In response to (2 R,6 R)-hydroxynorketamine (1 hour, 5 mcM), 13 genes were ≥1.5x up-regulated with FDR corrected $p \leq 0.05$, and 4 genes were ≥1.5x down-regulated with FDR-corrected $p \leq 0.05$. For the up-regulated genes, pathway analyses revealed 3 clusters: axon guidance (microtubule-mediated), transcriptional regulation (histones), and translational regulation (ribosomal proteins). In subsequent experiments, hCSs were briefly exposed to multiple low doses of racemic ketamine. Single cell RNA-Seq was performed in collaboration with the Iowa Institute of Human Genetics Genomics Division. At the time of abstract submission, this data is being processed with anticipated presentation for the first time at the 2022 American College of Neuropsychopharmacology's Annual Meeting (Phoenix, AZ, U.S.A.)

Conclusions: We have identified several genes/clusters altered by low-dose ketamine and/or its bioactive metabolites in hPSC-derived cortical-like organoid cultures. In the whole cell transcriptomic analyses, 2 R,6R-hydroxynorketamine, which is hypothesized to have non-NMDA receptor antagonist properties [Zanos et al. (2016) Nature 533(7604):481-6, PMID: 27144355], robustly upregulated several genes involved in cytoskeletal reorganization, e.g. stathmin-like 2 (STMN2), beta-2B tubulin and microtubule-associated proteins 1B and 2. Due to the known effects of low-dose ketamine on synaptic plasticity, it appears logical that cytoskeletal genes appear to be rapidly up-regulated. One major future direction is to determine if these such transcriptional alterations correlate with antidepressant efficacy. This may be assessed by stratifying at "efficacy bookends," e.g., hCSs derived from ketamine non-responders and ketamine remitters, in response to brief, low-dose incubation with racemic ketamine and/or its bioactive metabolites.

Keywords: Transcriptomics, Ketamine, (2 R,6 R)-Hydroxynorketamine, Stem Cells, Brain Organoids

Disclosure: Johnson and Johnson-Janssen: Contracted Research (Self)

P294. Neurodevelopmental Signature of a Transcriptome-Based Polygenic Risk Score for Depression

Yuliya Nikolova*, Amy Miles, Fernanda Dos Santos

Centre for Addiction and Mental Health, Toronto, Canada

Background: Transcriptomic studies in post-mortem human brain tissue have begun to shed light onto the molecular mechanisms implicated in Major Depressive Disorder (MDD). Yet, it remains unclear how these transcriptomic shifts may impact brain structure or function in vivo. We recently leveraged common cis-eQTL single nucleotide polymorphisms (SNPs) to create a transcriptome-based polygenic risk score (tPRS) reflecting developmental shifts towards depression-like corticolimbic gene expression patterns, based on 76 genes differentially expressed in MDD. We identified distinct sex-specific neurofunctional and neurostructural signatures associated with higher tPRS and MDD risk in young adults, independent of the effects of traditional MDD PRS emerging from genome-wide association studies (GWAS). To elucidate the neurodevelopmental impact of tPRS and provide insight into when these risk phenotypes may first emerge, we set out to map an expanded version of tPRS, comprising 332 genes differentially expressed in MDD, onto individual differences in brain structure and depressive symptoms in children participating in the Adolescent Brain Cognitive Development (ABCD) study.

Methods: This study used tabulated neuroimaging data, collected at baseline, from 5124 non-Hispanic white participants in the ABCD study (2737 male, 2387 female; 9.9 ± 0.6 years of age (range: 8.9 – 11.0)). We used the tool MetaXcan and the CommonMind Consortium reference transcriptome to impute relative gene expression in the dorsolateral prefrontal cortex (dlPFC) at the individual level. Individual SNP contributions were determined based on weighting in a tissue-specific elastic net prediction model (DLPFC_newMetax.db). Using this model, we were able to impute 9347 genes, which included 332 out of the 566 genes previously identified in a meta-analysis of case-control post-mortem brain corticolimbic transcriptome datasets ('metaA-MDD genes'). tPRS was computed as the sum of the imputed expression values of the 332 imputed genes, each weighted by the direction of effect in the original post-mortem meta-analysis. Separate linear mixed effects models were used to test main effects of tPRS and tPRS-by-sex interaction effects on volume in each subcortical segmentation ($n = 14$), and cortical thickness and surface area in each Desikan-Killiany atlas-based cortical parcellation ($n = 68$). Within each analysis, FDR-correction was applied to account for testing in multiple regions. In each case, tPRS, sex, age, estimated total intracranial volume (eTIV, not included in cortical thickness analyses) and 10 genetic components were modeled as fixed effects, while site was modeled as a random effect. Depressive symptoms were indexed using the t-score from the depressive syndrome scale of the parent-reported Child Behavior Checklist (CBCL).

Results: We identified a significant tPRS-by-sex interaction effect on volume in the right hippocampus (HPC.R: $t = -3.087$, $pFDR = 0.028$) and right pallidum (PAL.R: $t = -2.808$, $pFDR = 0.035$), wherein higher tPRS was associated with lower volumes of both structures in females (HPC.R: $t = -3.058$, $pFDR = 0.002$; PAL.R: $t = -3.112$, $pFDR = 0.002$) but not in males (HPC.R: $t = 1.379$, $pFDR = 0.227$; PAL.R: $t = 1.208$, $pFDR = 0.227$). Higher tPRS was further associated with greater cortical thickness in the left posterior cingulate cortex when tested in the full sample ($t = 3.739$, $pFDR<0.001$). No other neurostructural effects emerged. Lower right hippocampal and pallidum volume further showed trend-level associations with higher depressive symptoms in the female subsample (HPC.R: $t = -1.942$, $p = 0.052$, $pFDR = 0.052$; PAL.R: $t = -2.121$, $p = 0.034$, $pFDR = 0.052$), where the right pallidum volume was also a significant mediator of an indirect effect of tPRS on elevated depressive symptoms (PAL.R, $p = 0.016$). In contrast, posterior cingulate cortical thickness was not associated with depressive symptoms when tested in the full sample ($t = 0.009$, $p = 0.993$). All effects' significance remained unchanged when a more conventional GWAS-based measure of MDD PRS was included as an additional covariate.

Conclusions: Our results suggest that genetic variants associated with shifts towards a depression-like corticolimbic transcriptome may have a distinct sex-specific neurodevelopmental signature affecting the morphology of corticolimbic regions, which may indirectly increase risk of depression, particularly in female participants. Future studies will examine the effects of tPRS on brain maturation and depressive symptom change across adolescence to identify trajectories of risk amenable to early prevention efforts.

Keywords: Depression, Polygenic Scores, MRI, ABCD Study, Brain Structure

Disclosure: Nothing to disclose.

P295. Correlation Between Gene Expression of SIRT2 - SIRT7 and Cognitive Function in Recurrent Major Depressive Disorder

Angelos Halaris*, Maria Filip, Piotr Galecki

Loyola University School of Medicine, Maywood, Illinois, United States

Background: Sirtuins are proteins found in all aerobic organisms. They are enzymes that regulate important biological functions and are involved in several pathways. There are seven known sirtuins in humans (Sirt1-Sirt7). They are present in all organisms, including bacteria. Sirtuins have been associated with caloric restriction, aging, metabolism, cancer, transcriptional silencing, chromosomal stability, cell differentiation, stress response, inflammation, apoptosis, DNA repair, and prevention of age-related ocular diseases. Sirt1 is involved in gene silencing, cell cycle, fat and glucose metabolism and cellular oxidative stress. A genetic study has received considerable attention for association with depression and determined that Sirt1 is a potential gene target. The study conducted by the CONVERGE Study Cconsortium (China, Oxford and Virginia Commonwealth University Experimental Research on Genetic Epidemiology) identified two loci that contributed to the risk of MDD on chromosome 10: one is close to the Sirt1 gene (P52.53310210) and the other is localized in one of the introns of the LHPP gene (P56.45310212). A case-control investigation in Japan showed that one SNP (rs10997875) in the Sirt1 gene could play a role in MDD pathophysiology. It was also found that there is a link between the Sirt1 gene (rs3758391) and depressive disorders. Further, it has been shown that Sirt1 expression in the peripheral blood from individuals with depression is significantly less than healthy subjects. Sirt1 expression is markedly down-regulated in the blood of MDD patients when compared with control subjects and those with remitted MDD. Given the relative paucity of studies in depressive disorders, we conducted the present study.

Methods: Seventy-two newly admitted hospitalized patients who met diagnostic criteria for recurrent major depressive disorder (MDD) and inclusion/exclusion criteria, were enrolled in the study after signing informed consent. A comparative group consisted of 74 healthy volunteers with a negative history of any mental disorder. Complete clinical and laboratory data about these subjects were retrieved from the Department's database. The following data were collected: sirtuin 2-7 mRNA expression levels, results of 21 cognitive assessment tests, clinical parameters

describing the patient's depressive disorder. The data were analyzed in an effort to detect possible differences in sirtuin 2-7 mRNA expression levels between the group of patients and the healthy controls, correlations between levels of expression of sirtuin 2-7 mRNA, and the results of cognitive assessment in the group of MDD patients, correlation between levels of sirtuin 2- 7 mRNA expression and clinical data describing the course of the disease in the group of patients. The study was approved by the Bioethics Committee No. RNN/137/17/ KE and No. RNN/303/18/ KE, and the collection of biological research material was approved by the Bioethics Committee No. RNN /566/08/ KB.

The specific aims of the study included: 1. Determine whether gene expression of sirtuins 2-7 differed between patients and healthy controls. 2. Establish whether there were correlations between gene expression levels for sirtuins 2-7 and the severity of cognitive impairment in these patients. 3. Establish whether there any correlations between gene expression levels of these genes and clinical variables in these patients.

Results: The levels of gene expression of sirtuins 2-7 differed statistically significantly between the two study groups. The levels of expression of the sirtuin 2, sirtuin 3, sirtuin 4, sirtuin 5 and sirtuin 7 genes were significantly higher in patients compared to healthy controls. Conversely, the level of expression of the sirtuin 6 gene was statistically significantly higher in healthy controls compared to the patient group. There were statistically significant correlations between the level of expression of the sirtuin 4 gene and cognitive function on the Stroop B scale for time, and the level of expression of the sirtuin 7 gene and cognitive function in the California Language Learning Test CVLT2 and in scale SIET raw. Lastly, there were statistically significant correlations between the level of gene expression of two sirtuins, 3 and 6, and the Hamilton Depression score on the day of admission to the hospital.

Conclusions: Expression of genes for sirtuins 2-7 in patients with recurrent MDD is significantly different from the expression of these genes in healthy subjects. There were three significant correlations between gene expression for sirtuins 2-7 and the severity of cognitive impairment in patients versus healthy controls. The gene expression of sirtuins may prove to be a biomarker for cognitive impairment in this patient population if confirmed in future studies.

Keywords: Major Depressive Disorder (MDD), Biomarker, Genes, SNP, Sirtuins

Disclosure: Nothing to disclose.

P296. Genome-Wide Methylation Study of Suicide Attempt in Bipolar Disorder Suggests Epigenetic Pathway to Peripheral Immune Alterations

Salahudeen Mirza*, Camila De Carvalho-Lima, Alexandra Del Favero-Campbell, Alexandre Rubinstein, Brenda Cabrera-Mendoza, Consuelo Walss-Bass, Joao Quevedo, Jair Soares, Gabriel Fries

Institute of Child Development, University of Minnesota, Minneapolis, Minnesota, United States

Background: Suicide is a leading cause of death worldwide and has shown extensive comorbidity as well as genetic correlation with bipolar disorder (BD). BD patients show a 10-30-fold increase in risk for suicide. DNA methylation (DNAm) patterns involve both genetic and experiential contributions and may be promising biological substrates for suicide attempt (SA) risk. Previous studies have considered DNAm in SA, but often lack matched diagnosis reference groups for better disentanglement between the pathophysiologies of SA and psychiatric disorder. We conducted an extensive epigenome-wide association study of SA in BD.

Methods: DNA from seventy-nine BD patients with a lifetime history of SA (BD/SA) and eighty-four BD patients without a lifetime history of SA (BD/non-SA) was isolated from the buffy coat fraction of whole blood. 500 ng of DNA were bisulfite converted and hybridized to the Illumina Infinium EPIC BeadChip, which queries methylation at over 850,000 cytosine-preceding-guanine (CpG) sites. Analyses considered genome-wide methylation differences between BD/SA and BD/non-SA. False discovery rate (FDR) significant ($q < 0.05$) differentially methylated positions (DMPs) were detected using linear models with the limma package in R, and Šidák correction significant (Šidák $p < 0.05$) differentially methylated regions (DMRs) were detected using the comb-p algorithm within the ENmix package in R. Area under the curve (AUC) of the receiver operating characteristic (ROC) curve was used to evaluate the prediction capability of a binomial generalized linear model predicting BD/SA from the beta-values for the FDR significant DMPs. Gene ontology (GO) pathway enrichment analysis using the DMPs at $p < 0.001$ was performed using the gometh function in the missMethyl R package. Weighted gene co-methylation network analysis (WGCNA) using the DMPs at $p < 0.05$ and located at transcriptional start sites was performed in the R package WGCNA, and modules were evaluated for relation to BD/SA with age and sex adjusted binomial generalized linear models, as well as subjected to GO pathway enrichment analysis. GWAS enrichment analysis was performed using a gene set consisting of the DMPs at $p < 0.05$ and the summary statistics for recent GWAS of SA. DMP and DMR analyses were adjusted for age, sex, the first three genomic ancestral principal components, DNAm-based white blood cell count proportion estimates, and DNAm-based smoking score estimates.

Results: Six FDR-significant DMPs were related to BD/SA, with the leading site nearest to the IL8 gene (hypomethylated in BD/SA; logFC = -0.33, pFDR= 0.03). AUC for the ROC curve predicting BD/SA from the six FDR significant DMPs was 83.5% (CI = 77.4-89.59%). Nine significant DMRs, ranging from 3 to 19 CpG sites, were identified, with the leading DMR nearest to the CLDN9 gene (Šidák $p = 7.09 \times 10\text{-}9$). GO pathway enrichment analysis from DMPs implicated biological processes modulated by calcium. In the WGCNA analysis, six co-methylated modules, ranging from 188 to 7237 CpG sites, were identified. All six modules were significantly related to BD/SA, with one module ($\beta = 7.15$, pFDR = 0.004) being enriched for genes related to immune processes in GO analysis. GWAS enrichment analysis revealed no gene set enrichment in summary statistics for SA in BD, SA in psychiatric disorders, and SA in the general population.

Conclusions: Results suggest an epigenetic signature associated with SA in BD. These biological factors may be useful to delineate a more severe subgroup of BD patients prone to suicidal behaviour. DMP and WGCNA analyses converge on immune-related processes, which parallels existing literature documenting peripheral immune alterations in suicidal behaviour. The divergence between genetic and epigenetic correlates of suicidal behaviour suggests that risk for suicidal behaviour may involve different biological mechanisms at each of these levels. Therefore, integrating genetic and epigenetic information together may offer further clarity in understanding the pathophysiology of suicidal behaviour. On both fronts, replication studies are needed to understand the reliability of identified (epi)genetic markers, if they are to be incorporated into (preventive) intervention efforts.

Keywords: Epigenome Wide Association Studies, Suicide Attempt, Bipolar Disorder (BD), Peripheral Blood Marker, DNA Methylation

Disclosure: Nothing to disclose.

P297. Differential MicroRNA Expression Profiling of Neural-Derived Extracellular Vesicles in Bipolar Disorder Patients: A Preliminary Analysis

Gabriel R. Fries*, Salahudeen Mirza, Camila Nayane de Carvalho Lima, Wei Zhang, Giselli Scaini, Amit Srivastava, Zhongming Zhao, Benson Mwangi, Jair C. Soares, Joao de Quevedo

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

Background: Bipolar disorder (BD) is a globally prevalent psychiatric disorder associated with functional impairment and elevated risk for suicide. There is an urgent need to identify biological mechanisms underlying BD to facilitate risk assessment, prevention, and intervention efforts. Epigenetic processes may be one pathway by which experiential risk and genetic liability interface to drive risk for BD. In particular, microRNAs (miRNAs) have recently been highlighted as important candidates in the study of psychiatric disorders due to their dynamic expression and ability to regulate the expression of many target genes. However, peripherally sampled miRNAs do not directly reflect the expression of miRNAs in the brain. We isolated extracellular vesicles (EVs) from plasma which were tagged for neural origin to identify a peripheral marker of brain gene expression. We then conducted a differential miRNA expression analysis between BD patients and healthy controls (HC) to identify miRNAs which may be differentially expressed (DE) in the brain of living BD patients.

Methods: This preliminary analysis included 20 patients with BD and 20 age- and sex-matched HC. EVs were isolated from plasma with the ExoQuick® ULTRA EV Isolation Kit (Systems Biosciences) and further immunoprecipitated for the L1 cell adhesion molecule (L1CAM) neural adhesion protein, a validated marker for neural-derived EVs. Total RNA was isolated from the neural–derived EVs with the exoRNeasy Midi Kit (Qiagen) and prepared for RNA sequencing on the Illumina NextSeq. Sequencing reads were submitted to the ERCC exceRpt small RNA-seq pipeline (v.4.6.2) with default settings (adapter trimming, alignment to miRBase hg38). After filtering steps, 157 miRNAs remained for differential expression analysis in DESeq2 (v.1.36). The design was adjusted for age (median split) and sex, with HC as reference. Benjamini-Hochberg false discovery rate (FDR) procedure corrected for multiple comparisons. FDR significant DE miRNAs were inputted to the miEAA 2.0 web tool to identify implicated gene ontology (GO) processes (miRTarBase, miRWalk), diseases (MNDR), pathways (KEGG, miRWalk), and target genes (miRTarBase). We focused on the pathways, processes, and target genes which implicated the largest number of FDR significant DE miRNAs.

Results: The differential expression analysis revealed 27 FDR significantly differentially expressed miRNAs in patients, led by hsa-miR-301a-3p (log2FC = 3.88, pFDR = 1.32 ×10-6). Variance stabilized hsa-miR-301a-3p expression classified BD patients with an AUC of the ROC curve of 70% (CI = 54-86%). Sixteen miRNAs were upregulated and 11 miRNAs were downregulated at the FDR significance threshold. Implicated GO processes included enzyme binding, cell proliferation, neuron projection, and positive regulation of protein phosphorylation (miRTarBase); positive regulation of transcription from RNA polymerase II promoter, extracellular region, blood coagulation, and endoplasmic reticulum membrane (miRWalk). Diseases included carcinoma, ductal, breast; hepatocellular carcinoma; breast cancer; and colorectal cancer. Pathways included yersinia infection, regulation of actin cytoskeleton, growth hormone synthesis, secretion, and action, AGE-RAGE signaling pathway in diabetic complications (KEGG); cancer, focal adhesion, integrated pancreatic cancer pathway, and angiogenesis (miRWalk). Target genes included NUFIP2, PTEN, WASL, and MBNL1.

Conclusions: Our preliminary results support an epigenetic biosignature to BD and validate the potential for neural-derived EVs to characterize miRNA expression patterns in BD and other psychiatric disorders. Established relevance of target genes to nervous system function as well as psychiatric disorders is promising. Further investigation will be required to better understand the functional implications of the observed miRNA alterations, as well as their concordance to studies conducted in post-mortem brain tissue. In particular, given higher burden for certain diseases in BD patients, miRNA alterations may be one possible pathway to disease risk, although this hypothesis is preliminary. Regardless, findings offer a promising first step to incorporating brain-specific miRNAs to monitoring as well as intervention work.

Keywords: Bipolar Disorder, MicroRNA, Extracellular Vesicles, Epigenetics

Disclosure: Nothing to disclose.

P298. Functional Magnetic Resonance Spectroscopy of Emotional Processing at 7 T

Alexander Lin*, Katherine Breedlove, Jessica Busler, Eduardo Coello, Huijun Liao, Pamela Mahon, Pamela Mahon

Brigham and Women's Hospital, Harvard Medical School, Boston, Massachusetts, United States

Background: Functional MR Spectroscopy (fMRS) at 7 T provides increased sensitivity and spectral dispersion that enables the detection of dynamic changes in metabolite concentrations in the brain under specific functional tasks. Recently, fMRS experiments performed at 7 T have successfully detected changes in metabolite concentrations. To maximize the sensitivity of these experiments, high stability of the system and the patient are required (i.e., B0-homogeneity, negligible B0-drifts, effective water suppression, reduced eddy currents, and negligible patient motion). Moreover, special processing is required to account for different external instabilities of the experiment. This work presents a novel fMRS methodology to investigate metabolic changes related to emotional processing with improved sensitivity at 7 T. An acquisition protocol for the evaluation of a visual go-no-go task was designed. Furthermore, a processing pipeline that corrects measurement instabilities is presented.

Methods: Data acquisition: All data was acquired on a 7 T scanner (Terra; Siemens, Erlangen, Germany). Patients were scanned in a single channel transmit 32-channel receive head coil with additional dielectric padding to improve for B0 inhomogeneity. High-resolution MRI images were acquired using magnetization-prepared rapid gradient echo with different inversion times (MP2RAGE) using TR = 2.530 ms, TE1 = 1.64 ms, TE2 = 3.5 ms, TE3 = 5.36 ms, TE4 = 7.22 ms, Flip=7°, FOV = 256 cm, 0.7×0.7×0.7 mm resolution, acceleration factor of 2, for a scan time of 6 minutes. A 20x40x20mm3 volume was acquired using a semi-LASER localization sequence optimized for 7T28 with the gradient-modulated FOCI pulses to reduce maximum B1 (TE = 28 ms, TR = 5 s, 128 scans, spectral width 6 kHz). VAPOR water suppression will be interleaved with the outer-volume saturation. Unsuppressed water signal was acquired using 4 averages for eddy current correction and as an internal reference for metabolite quantification. 768 signal averages were acquired during the full experiment for a total acquisition time of 24 minutes.

Visual stimuli and fMRS paradigm: The fMRS paradigm consisted of an affective go/no-go task to assess emotional regulation. Participants were instructed to silently read each word presented

and then to button-press for words in normal font (go trial) and to inhibit this response for words in an italicized font (no-go trial). Stimuli (words) had negative, positive, and neutral valence. Performance during the task and post-task assessment of stimuli recognition and valence were used to determine participant compliance with task instructions.

Data Processing and Quantification: The pipeline used for reconstruction of the spectra was implemented in python using OpenMRSLab and included the following steps: (1) coil combination, (2) frequency alignment to water, (3) zero-order phase removal, (4) combination of 4 signal averages (sub-blocks) to increase SNR for processing, (5) water removal at each sub-block using HSVD, (6) frequency alignment to NAA, (7) zero-order phase removal, (8) filtering of the dynamic temporal signal (e.g. sliding window average, Fourier thresholding), and (9) combination of sub-blocks to obtain the desired temporal resolution to match the paradigm. Frequency/phase corrections eliminated distortions caused by external effects that affect the measurement, such as B0-drifts, eddy currents, temperature, or the BOLD effect. The reconstructed spectra, corresponding to consecutive time points, were fitted using LCModel with a simulated basis set.

Results: The analysis of the auditory fMRS data showed the presence of BOLD signal when subtracting each STIM block with its nearest REST block. Variations in metabolite concentrations were observed and enhanced using dynamic filtering. The quantified metabolite concentrations processed with the different methods superimposed with the visual paradigm are shown. The visual experiment served as a validation for the methodology as it was able to reproduce previously published experiments.

Conclusions: This is a promising development for the application of functional MRS to the study of mood disorders such as bipolar disorder. Altered performance in neural activation and neurometabolites levels has previously been correlated with this go-no-go tasks in patients with bipolar disorder. Therefore, the ability to measure dysfunction in the glutamatergic systems that may be indicative of network dysfunction observed in mood disorders is a promising application of functional MRS.

Keywords: Functional Magnetic Resonance Spectroscopy, Emotional Regulation, Ultra-high Field MRI, GoNoGo

Disclosure: Agios Pharmaceuticals, Biomarin Pharmaceuticals, Design Therapeutics, Moncton MRI: Consultant (Self), BrainSpec, Inc: Founder (Self)

P299. Fronto-Limbic Functional Connectivity as a Predictor of Emotional Arousal in Youth at Risk for Bipolar Disorder

Samantha Hu, Aniruddha Shekara, Maxwell Tallman, Rodrigo Patino, Thomas Blom, Jeffrey Welge, Jeffrey Strawn, Kaitlyn Bruns, David Fleck, Manpreet Singh, Caleb Adler*, Melissa DelBello

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

Background: Antidepressants are a first-line treatment for youth with anxiety and/or depression; however, their use can carry significant risk of adverse psychiatric events (e.g., increased irritability, aggression, impulsivity, psychosis) associated with increased emotional hyperarousal (EH). This may be particularly important in individuals with a family history of bipolar disorder (BD). Previous studies have shown that selective serotonin reuptake inhibitors (SSRIs) may disrupt fronto-limbic neural pathways critical for emotion regulation; however, the neural mechanisms of EH remain largely unknown. We used functional neuroimaging to study youth with familial risk for BD receiving treatment for anxiety and/or depression to determine whether changes in emotion-associated fronto-limbic networks are predictive of dysfunctional emotional arousal.

Methods: Seventy-five youth with a family history of BD and a current depressive or anxiety disorder were recruited at the University of Cincinnati and Stanford University. All participants received escitalopram and during the treatment period of up to 16 weeks, 14 youth developed EH. All participants completed a functional magnetic resonance imaging (Fmri) scan at baseline. A second scan was obtained at the time of EH occurrence. Ten youth on escitalopram matched by time on treatment, age, sex, and site who did not develop EH during the 16-week period were also scanned for comparison.

EH was defined by clinician assessment, which prioritized observation of arousal-like behaviors in a clinical setting, but also incorporated parental and child self-reports, such as the Treatment-Emergent Activation and Suicidality Assessment Profile (TEASAP). During fMRI scans, participants viewed a series of stimuli and were asked to press a button when presented with an infrequent target (circles) in the context of infrequent distractors and more frequent, standard items (squares). Distractors were either emotional or neutral images.

Analysis of fMRI data was performed using the CONN toolbox 21a in MATLAB. ROI-to-ROI functional connectivity analyses were performed using the Harvard-Oxford Atlas-based insular cortex (INS), orbitofrontal cortex (OFC), amygdala (AMY), inferior frontal gyrus (IFG) pars opercularis and pars triangularis, and anterior cingulate cortex (ACC) seed regions. Differences in functional connectivity between emotional and neutral distractors were compared at baseline and between baseline and EH scans. Contrasts were analyzed with repeated measures ANOVA and random-effects mixed ANOVA. Significant ROI-ROI connectivity differences were reported with cluster-level threshold p-FDR < 0.05.

Results: Functional connectivity changes in participants who developed EH after treatment:

At baseline, participants who developed EH while receiving escitalopram displayed no statistically significant differences in ROI-ROI functional connectivity when comparing emotional and neutral stimuli. However, over time, participants with EH showed increased connectivity between left IFG pars opercularis and left insula (p-FDR = 0.0105) and between left IFG pars opercularis and right insula (p-FDR = 0.0105) when comparing emotional with neutral stimuli.

Functional connectivity changes in participants who developed EH and matched patients after treatment: Compared with matched subjects, participants who developed EH did not have significant baseline differences in functional connectivity when contrasting emotional and neutral stimuli. Over time, participants who developed EH exhibited increased connectivity between ACC and left IFG pars triangularis (p-FDR = 0.0075) when compared to matched subjects.

Conclusions: Youth with family history of BD who developed EH during escitalopram treatment did not demonstrate any differences in fronto-limbic functional connectivity at baseline. However, there are longitudinal changes in emotional processing circuitry over the course of treatment with escitalopram, implying that SSRIs may play a role in mediating disruptions in these fronto-limbic neural circuits. Due to the small sample size of 14 EH participants, further study is necessary to confirm these findings. Our results suggest that a larger sample of at-risk youth is necessary to elucidate the mechanisms underlying treatment-induced EH and uncover potential targets for pharmacotherapy in this patient population.

Keywords: Bipolar Disorder, fMRI, Task-based Functional Connectivity, Emotional Arousal

Disclosure: Janssen Pharmaceuticals: Speakers Bureau (Self), Janssen Pharmaceuticals: Advisory Board (Self), Otsuka: Speakers Bureau (Self)

P300. Examining Resting State Functional Connectivity in Suicidal Ideation and Attempts

Patricia Burhunduli, Zhuo Fang, Katie Vandeloo, Pierre Blier, Jennifer Phillips*

University of Ottawa Institute of Mental Health Research, Ottawa, Canada

Background: Suicide remains one of the most common preventable causes of death, although predicting and reducing suicide-related behaviours remains challenging. Studies have shown that suicide cannot be solely explained as a catastrophic outcome of major depressive disorder (MDD), but risk factors are multifaceted and neurobiological processes play an important role. Studies have shown that aberrant neural networks may be involved in suicide-related thoughts and behaviours. This study examined resting-state functional connectivity (FC) differences between suicidal ideation (SI) and suicide attempts (SA) in patients with treatment-resistant MDD.

Methods: The sample consisted of N = 40 patients with treatment-resistant MDD (n = 21 suicide ideators, n = 19 suicide attempters). Resting state functional magnetic resonance imaging (fMRI) data were acquired at 3 T and pre-processed using the default CONN Functional Connectivity pipeline. Group-level differences in resting-state functional connectivity between patients with SI and SA were completed using a region of interest (ROI)-to-ROI approach. SI and SA were clinically characterized using the Columbia Suicide Severity Rating Scale (C-SSRS); comparisons were statistically significant at pFDR≤0.05.

Results: Resting-state functional connectivity of the right hippocampus and the default mode network (DMN) (bilateral lateral-parietal lobe) were increased in patients with SI compared to SA (pFDR= 0.02). The connectivity of the DMN (right lateral-parietal lobe) and regions within the salience network, such as the anterior cingulate cortex (ACC) and the bilateral supra marginal gyrus (SMG), were significantly decreased in the SI group (pFDR=0.03). Functional connectivity between the caudate and the hippocampus/posterior parahippocampus was negatively correlated with current SI severity in the full patient sample (N = 40; pFDR=0.05), and in the attempter group specifically (n = 19; pFDR=0.008).

Conclusions: Our preliminary resting state fMRI analyses shows significant differences in DMN functional connectivity in patients with a suicide attempt history compared to those with suicidal ideation only. There were significant associations between limbic network functional connectivity and current suicidal ideation severity. These findings support potential neural circuits involved in the neurobiology of suicidal ideation and suicide attempt history in patients with treatment resistant MDD.

Keywords: Resting State Functional Connectivity, Suicide Attempt, Suicidal Ideation, Treatment-resistant Depression, Magnetic Resonance Imaging

Disclosure: Nothing to disclose.

P301. Regional Grey Matter Volume Correlates With Anxiety, Apathy and Resilience in Geriatric Depression

Beatrix Krause, Prabha Siddarth, Hanadi Ajam Oughli, Lisa Kilpatrick, Katherine Narr, Helen Lavretsky*

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

Background: Geriatric depression (GD) is associated with significant medical comorbidity, cognitive impairment and brain atrophy, premature mortality, and suboptimal treatment response. While apathy and anxiety are common comorbid symptoms in GD, resilience has been found to be a protective factor. Understanding the relationship between brain morphometry and symptoms and resilience in GD could inform clinical treatment targets. Gray matter volume (GMV) is altered in GD but correlations with mood symptoms and particularly resilience are less available.

Methods: Forty-nine older adults over 60 y.o. (38 women) diagnosed with major depressive disorder undergoing concurrent antidepressant treatment participated in the study. Depressive symptoms, as well as apathy, anxiety and resilience were assessed along with an anatomical T1-weighted MRI scan. We used a general linear model (GLM) to identify clusters in which GMV correlated with each measure. Freesurfer 6.0 was used to process T1-weighted images and perform voxel-wise whole-brain GLMs using age and sex as covariates. Partial Spearman correlations controlling for age and sex were used to investigate the association between clinical variables.

Results: Greater depression severity was, as expected, associated with greater anxiety (r = .53, p = .0001). Greater resilience was associated with lower depression (r = -.33, p = .03) and reduced apathy (r = .39, p = .01). Greater GMV in widespread, partially overlapping clusters across the brain was associated with reduced anxiety and apathy, as well as increased resilience

Conclusions: Our results suggest that GMV in extended brain regions is a potential marker for resilience in GD and overlaps in specific smaller regions with GMV markers for depression and anxiety. Since resilience is a protective factor against depression, future studies should examine GMV changes in these regions with treatment.

Keywords: Depression, MRI, Resilience, Apathy, Anxiety

Disclosure: Nothing to disclose.

P302. Reduced Prefrontal γ-Aminobutyric Acid and Glutamate Levels in Major Depression: Results of a Proton Magnetic Resonance Spectroscopy Study

Chris Ritter, Andreas Buchmann, Sabrina Müller, Martin Volleberg, Melanie Haynes, Carmen Ghisleni, Ralph Noeske, Ruth Tuura, Gregor Hasler*

University of Fribourg, Villars-sur-Glâne, Switzerland

Background: In MDD, perturbations of the major inhibitory and excitatory neurotransmitters, γ-aminobutyric acid and glutamate respectively, as well as Glx (glutamate + glutamine) have been extensively reported in a multitude of brain areas, but few studies have examined changes in glutamine, the metabolic counterpart of synaptic glutamate. In this study we investigated changes in glutamate, GABA, Glx, as well as glutamine levels in a voxel in the left dorsolateral prefrontal cortex of participants with no, past and current major depressive disorder in a large, unmedicated sample, recruited from the general population.

Methods: Cross-sectional design using 3-T 1H-MRS in participants recruited from the community. Our sample consisted of 251 healthy controls, 98 subjects with a history of past major depressive disorder, as well as 47 subjects who met the diagnostic criteria for current major depressive disorder. Diagnostic groups were comparable regarding age, education, income and diet. Our main outcome measures were GABA, glutamate and glutamine concentrations in left dorsolateral prefrontal cortex.

Results: Participants with past major depressive disorder had lower glutamate (r = .162, p = .010, n(Healthy; Past MDD) = 234, 93) and GABA (r = .184, p = .002, n(Healthy; Past MDD) = 236, 92), as well as higher glutamine (r = .165, p = .043, n(Healthy; Past MDD) = 153, 66) compared to participants without major depressive disorder. GABA concentrations were negatively associated with acute depressive symptoms (HAMD: rho = -.117, p = .022, n = 322, MADRS: rho = -.125, p = .018, n = 362, BDI: rho = -.150,

p = .005, n = 363), while glutamine was positively associated with neuroticism (rho = .202, p = .002, n = 240).

Conclusions: In a large, unmedicated community sample, reduced prefrontal GABA concentrations were associated with past and current major depressive disorder, consistent with histopathologic studies reporting reduced glial cell and GABA cell density in the prefrontal cortex in depression. Patients with major depressive disorder also demonstrated increased glutamine levels, indicative of increased synaptic glutamate release, adding to previous evidence for the glutamate hypothesis of major depressive disorder.

Keywords: Gutamate, Glutamine, GABA, Depression

Disclosure: Nothing to disclose.

P303. Impaired Coronary Microvascular Reactivity in Youth Bipolar Disorder

Benjamin Goldstein*, Nilesh Ghugre, Idan Roifman, Brian McCrindle, Bradley MacIntosh, Kody Kennedy

Centre for Addiction and Mental Health, Toronto, Canada

Background: Cardiovascular disease (CVD) is excessively prevalent and premature in bipolar disorder (BD), even after controlling for traditional cardiovascular risk factors. The increased risk of CVD in BD may be subserved by microvascular dysfunction. We set out to extend the prior literature on coronary microvascular dysfunction in psychiatric populations by focusing on BD in youth.

Methods: Participants were 86 youth, ages 13-20 years (n = 39 BD, n = 47 controls). Coronary microvascular reactivity (CMVR) was assessed using blood-oxygen-level-dependent T2-weighted magnetic resonance imaging including an established breath-holding paradigm. Images were acquired during normal breathing (baseline), following 60-seconds of hyperventilation, and every 10-seconds during a 40-second breath-hold. Measure of left ventricular structure (e.g. mass, volume) and function (e.g. ejection fraction) were evaluated based on 12-15 short- and long-axis cardiac-gated cine images. A linear mixed-effects model controlling for age, sex, and body mass index assessed for between-group differences in CMVR (i.e., a time-by-group interaction).

Results: The breathing paradigm induced a significant change in T2-relaxation time in the overall sample (i.e., CMVR; β = 0.36, p < 0.001). CMVR was significantly lower in BD vs HC (β = -0.11, p = 0.004). Post-hoc pairwise analyses revealed that significant between-group differences were evident after 30- and 40-seconds of breath-holding (p = 0.003 and p < 0.001, respectively). Measures of left ventricular structure and function were within normal ranges and did not significantly differ between groups.

Conclusions: There was evidence of coronary microvascular dysfunction, despite normal gross cardiac structure and function, in youth with BD. These findings converge with prior findings of adults with major depressive disorder and post-traumatic stress disorder. Future studies integrating larger samples, prospective follow-up, and blood-based biomarkers are warranted.

Keywords: Bipolar Disorder, Cardiac Reactivity, BOLD fMRI Signal, Adolescent

Disclosure: Nothing to disclose.

P304. Connectome-Based Prediction of Mood Lability in Youth at Familial Risk for Bipolar Disorder

Danella Hafeman*, Feldman Jamie, Mary Phillips

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: Mood lability, defined as frequent and marked changes in mood, is a predictor of mood disorder onset, poor psychosocial functioning, and suicidal thoughts and behaviors. Given the clinical relevance, it is critical to better characterize this phenotype, including the neural underpinnings. In recent years, whole-brain predictive approaches (e.g., Connectome Predictive Modeling) have emerged as useful tools for assessing the degree that edge strengths predict a given phenotype (e.g., anxiety or irritability), using a cross-validation approach. However, the interpretability of the networks that emerge from these analyses are limited. Network-Based Statistic (NBS) is an approach developed over a decade ago that identifies networks associated with a phenotype; recently, this approach has been adapted to a predictive context, using k-fold cross-validation (NBS-Predict). The combined use of NBS and NBS-Predict have the potential to identify reproducible and interpretable networks associated with a mood lability phenotype.

Methods: We recruited 69 youth 10-15 years old with family history of bipolar disorder; 33 of which were selected to have elevated mood lability (defined as an average of >10 on the parent- and self-reported Child and Adolescent Lability Scale, CALS). We also scanned 42 age- and sex-matched healthy controls. Family history was confirmed via either the Structured Clinical Interview for DSM-IV (mania and depression sections) and/or medical records. Youth were assessed using Kiddie Schedule for Affective Disorders and Schizophrenia, present and lifetime; they were also administered several questionnaires, including the CALS. Youth had an MRI scan on a 3 T scanner (Verio or Prisma), which included a 6-minute resting state protocol. Data were analyzed using fmriprep (20.2.6 LTS) and xcp-d (stable version), adjusting for motion, global signal, and derivatives (36p model). Participants with excessive motion (mean framewise displacement > .5 mm or max motion > 3 mm; n = 14) or artifact observed on visual inspection (n = 1) were excluded, yielding 96 participants with acceptable data (58 at-risk, 38 healthy controls). Using nilearn, we extracted timeseries from the Shen parcellation (268 parcels) and generated connectivity matrices. We next used NBS-Predict to assess predictive models for risk status (at-risk vs. healthy) and mood lability; all models were adjusted for scanner, mean framewise displacement, age, and sex. Models were tested using 5-fold cross validation (10 repetitions), using the optimized prediction model (established in the training set) and thresholds of p < .01 and p < .05, with 5000 permutations. NBS (v1.2) was used for additional exploratory models, including testing signed networks separately and testing CALS as a dichotomous predictor.

Results: Using an individual edge threshold of p < .05, we identified a network that predicted mood lability (correlation: 0.335 (0.299, 0.371), p = 0.017), explaining 10.3% of the variance. Limiting to the most influential edges (weight threshold=1), we found 52 edges (15 positively correlated; 37 negatively correlated). Visual networks (esp. Visual II) were highly represented in selected edges. Visual II connectivity with motor, salience, and medial frontal networks was correlated with lower mood lability, while Visual II-frontoparietal network connectivity was correlated with greater mood lability. Testing networks in NBS, we found a significant network negatively correlated with mood lability (5000 permutations, p = .004); a positively correlated network approached significance (5000 permutations, p = .055). These networks were largely overlapping with those identified by NBS-Predict, but with more nodes in medial frontal and frontoparietal networks. Dichotomizing mood lability at a median split (10), we found a significant network negatively predicting mood lability. We were not able to classify at-risk versus healthy controls using NBS-Predict; similarly, NBS did not identify any significant networks between these two groups.

Conclusions: We find that a connectome-based approach identified a network (consisting of both negative and positive edges) that predicts mood lability in a sample of healthy controls and at-risk youth. While we hypothesized that subcortical and medial frontal networks would be most correlated with mood

lability, we were surprised to find that primary visual cortex was heavily represented in the predictive network. Most strikingly, more connectivity between visual and motor context (anatomically consistent with the visual dorsal stream) was associated with less mood lability. Interestingly, previous work has shown that the visual dorsal stream is activated by naturalistic emotional stimuli and may be important for action preparation; in this way, higher connectivity within the dorsal stream may enhance emotion regulation strategies, and thus less mood lability. While NBS and NBS-Predict generated similar edges, one important difference is that networks prominent in NBS (e.g. medial frontal) did not feature so prominently in NBS-Predict results; this may be related to their out-of-sample performance. While k-fold cross-validation enhances reproducibility of findings, the gold standard approach is out-of-sample replication; in the future, we will plan to test this model in an independent sample of at-risk youth.

Keywords: Mood Dysregulation, Resting-state fMRI, Familial Risk of Bipolar Disorder

Disclosure: Nothing to disclose.

P305. Estrogen Receptor Beta Mediates Stress-Susceptibility in the Male Brain

Polymnia Georgiou*, Ta-Chung M. Mou, Liam E. Potter, Xiaoxian An, Panos Zanos, Michael S. Patton, Katherine J. Pultorak, Sarah M. Clark, Vien Ngyuyen, Chris F. Powels, Katalin Prokai-Tatrai, Istvan Merchenthaler, Laszlo Prokai, Margaret M. McCarthy, Brian N. Mathur, Todd D. Gould

University of Wisconsin Milwaukee, Milwaukee, Wisconsin, United States

Background: Depression is the leading cause of disability worldwide afflicting ~16% of the population. Although, women are diagnosed with depression twice as often as men, there are approximately 109 million men worldwide suffering from the disease. In addition, men suffering from depression are at higher risk to lose their life from suicide, one of the most common symptoms of depression, with suicide rates being four times higher in men than women. In susceptible populations, stress is a major risk factor for the development of mental disorders, including depression. While the role of estrogen receptors in the pathophysiology of depression and as treatment targets in females are widely elucidated, their role in males is not well understood.

Methods: We used a subthreshold social defeat stress model which consists of three cycles of two-min physical stress and fifteen min of sensory stress, in combination with immunohistochemistry, neuronal tracing, RNAscope, in vitro electrophysiology, in vivo optogenetics, in vivo chemogenetics, in vivo fiber photometry and surgical/pharmacological hormonal manipulations to investigate the role of estradiol and its receptors in the development of social avoidance and anhedonia in male and/or female mice ($n = 10$-15/experimental group). Statistical analysis was performed with ANOVAs followed by Holm-Sidak post-hoc analysis. If criteria for parametric analysis were not met, Kruskal-Wallis test was performed followed by two-stage linear step-up procedure of Benjamini, Krieger and Yekutieli.

Results: We found that absence of estrogen receptor-β (ERβ) is associated with stress susceptibility in male but not female mice following exposure to a mild stressor and infusion of an ERβ-specific agonist in the basolateral amygdala (BLA) rescues this stress susceptibility in male mice. We identified a dense ERβ-expressing projection from BLA to the nucleus accumbens (NAc) and optogenetic stimulation of this projection resulted in the development of real-time place preference for the light-paired compartment in male mice while female mice did not show any preference between the light-paired and unpaired compartments suggesting a sex-dependent effect of this circuit. In addition, we observed that activity of these neurons is reduced in male mice lacking gonadal hormones subjected to mild stress and this is associated with stress susceptibility. Moreover, we demonstrated that optogenetic activation of this circuit reverses stress-induced maladaptive behaviors and induces stress resilience in hypogonadal mice, while chemogenetic inhibition of this circuit induces a stress susceptible phenotype following mild stress in intact mice. Estradiol (E2), often considered a female specific hormone, is distributed in the male brain via aromatization of testosterone. We identified that absence of E2, but not testosterone per se, underlies susceptibility to develop maladaptive behaviors following exposure to mild stress in males. Using brain-selective delivery of E2 through administration of a prodrug, which offers a viable treatment option in males, we demonstrated that E2 prevents the development of depression-related behaviors following acute/mild stress in hypogonadal male mice.

Conclusions: Overall, our findings provide evidence for an estrogen-based mechanism underlying stress susceptibility and suggest a novel therapeutic strategy utilizing brain selective estradiol for treating depression in males

Keywords: Estradiol, Depression, Neural Circuit and Animal Behavior, Estrogen Receptor

Disclosure: A patent was filed for the development of brain selective estradiol compounds: Patent (Self)

P306. Luteal Phase Epigenetic Biomarkers Identify Premenstrual Dysphoric Disorder (PMDD) and Selective Serotonin Reuptake Inhibitor (SSRI) Response in PMDD

Liisa Hantsoo*, Zachary Kaminsky, Jennifer Payne

Johns Hopkins University, Baltimore, Maryland, United States

Background: Premenstrual dysphoric disorder (PMDD) is considered a type of reproductive affective disorder and is characterized by affective symptoms that emerge in the luteal phase of the menstrual cycle and remit in the follicular phase. PMDD's pathophysiology is poorly understood, although genetic vulnerabilities, including epigenetic changes, may contribute. Previous research has found DNA methylation variations that predict postpartum depression (another reproductive affective disorder) with up to 80% accuracy. This study explored these epigenetic markers in those with PMDD, compared with healthy controls, in the follicular and luteal phases in order to determine if these biomarkers were specific for postpartum depression or were more general biomarkers of reproductive affective disorders.

Methods: Female control and PMDD participants were recruited from the community and completed two months of daily prospective symptom ratings (Daily Record of Severity of Problems (DRSP), a standard measure for premenstrual symptoms) to assess control or PMDD status, confirmed with SCID interview. Blood was collected during the follicular (days 5-11 of the menstrual cycle) or luteal phase (days -7 to -1 of the menstrual cycle, confirmed with urine lutenizing hormone test and serum progesterone levels ≥3 ng/ml). Additionally, women with PMDD received treatment with 50 mg of the selective serotonin reuptake inhibitor (SSRI) sertraline during a subsequent luteal phase and were characterized as either "responders" or "nonresponders" based on a minimum 30% reduction in DRSP score. Blood samples underwent sodium bisulfite pyrosequencing at the TTC9B and HPIBP3 genes and our published, established PPD biomarker linear model was used to evaluate whether our postpartum depression model was able to predict PMDD status compared to controls, and, in addition, predict SSRI response in the PMDD participants.

Results: Blood samples were available for fifty-five participants (n = 26 control, n = 29 PMDD); 23 in the follicular phase and 32 in the luteal phase. Follicular samples failed to distinguish PMDD cases (N = 13) from controls (N = 9). However, luteal phase samples were able to distinguish PMDD cases (N = 10) from controls (N = 18) with an AUC of 0.71 (95% CI: 0.49-0.93). Our methylation biomarkers were also able to distinguish between SSRI responders (N = 5) and SSRI nonresponders (N = 5) using luteal phase blood with an AUC of 0.84 (95% CI: 0.57-1).

Conclusions: This study examined CpG methylation levels at two loci within the HP1BP3 and TTC9B genes. We found that methylation patterns at these two genes were able to distinguish between PMDD cases and control participants but only when luteal phase blood was used indicating that the hormonal changes that occur during the luteal phase may be important. Our methylation biomarkers were also able to discriminate between SSRI responders and nonresponders in PMDD cases when luteal phase blood was used. Research on postpartum depression, another reproductive affective disorder, suggests that these loci may mediate sensitivity to changes in estrogen that occur during and after pregnancy which suggests that they may also be identifying women who are sensitive to the hormonal changes that occur during the luteal phase of the menstrual cycle. Our results also indicate that there may be biological differences between SSRI responders and nonresponders. We hope to replicate these findings in a large sample of PMDD cases and controls as well as examine our biomarker's ability to predict case status in other types of reproductive affective disorders.

Keywords: Epigenetic Biomarkers, Reproductive Affective Disorder, Premenstrual Dysphoric Disorder, Allopregnanolone, Estradiol

Disclosure: Nothing to disclose.

P307. Differential Cellular Response to Allopregnanolone in Postpartum Depression

Sarah Rudzinskas*, Maria Mazzu, Allison Goff, Crystal Schiller, Samantha Meltzer-Brody, David Rubinow, David Goldman, Peter Schmidt

National Institute of Mental Health, NIH, Rockville, Maryland, United States

Background: Given the health consequences, societal burden, and prevalence of postpartum depression (PPD), the recent FDA-approval of Brexanolone was an encouraging advance in PPD treatment. Allopregnanolone (ALLO), the naturally occurring steroid metabolite of progesterone that is chemically identical to Brexanolone, is thought to exert its anxiolytic- and antidepressant-like effects via positive allosteric modulation of GABAA receptors. However, ALLO's mechanism of action at the molecular level, and whether that mechanism is specific to PPD, remains unclear.

Methods: We examined the consequences of ALLO exposure using transcriptomics on lymphoblastoid cell lines (LCLs) derived from women with past PPD (n = 9) and women with no history of PPD or other psychiatric illness (n = 10, i.e., Controls). All LCLs were treated for 60 hours total with either ALLO (three spikes, 100 nM/spike) or DMSO vehicle, creating four different experimental groups: Control:DMSO, Control:ALLO, PPD:DMSO, and PPD:ALLO. LCLs were then collected for AmpliSeq RNA-sequencing and analyzed for differential gene expression 1) between Control and PPD LCLs, at baseline (Control:DMSO vs. PPD:DMSO) and after ALLO treatment (Control:ALLO vs. PPD:ALLO) and 2) within either Control (Control:DMSO vs. Control:ALLO) or PPD LCLs (PPD:DMSO vs. PPD:ALLO). Quality control, unsupervised clustering, and expression analyses were performed in Transcriptome Analysis Console (TAC) 4.0, and Weighted Gene Correlation Network Analysis (WGCNA) was performed in R.

Results: Differentially expressed genes (DEGs, p(nom)<0.05, log(Fold Change)>=|1.25|) were detected after ALLO treatment within, as well as between, PPD and Control LCLs. Between Control:ALLO vs. PPD:ALLO, 269 DEGs were observed, with Enrichr revealing many that were related to synaptic activity. Amongst these DEGs was Glutamate Decarboxylase 1 (GAD1), which was significantly (p(nom)<0.019) decreased in PPD:ALLO compared to Control:ALLO LCLs. Technical replication via qRT-PCR confirmed that GAD1 expression was decreased in PPD LCLs compared to Controls (Diagnosis: $F(1,34)=5.25$, $p = 0.0283$), but ALLO did not significantly effect expression (Treatment: $F(1,34)=0.006121$; Interaction: $F(1,34)=0.0108$). Overall patterns of gene expression also demonstrated that regardless of ALLO treatment, diagnosis was the primary driver of expression differences, suggesting a robust effect of PPD on gene expression, in line with previous data (Rudzinskas et. al, under review). Surprisingly, substantially more DEGs were induced with ALLO treatment in control LCLs (Control:DMSO vs. Control:ALLO, 265 DEGs), as compared to in PPD LCLs (PPD:DMSO vs. PPD:ALLO, 98 DEGs), with only 11 of these 363 total ALLO-related DEGs overlapping. Correspondingly, Gene Set Enrichment Analysis using MSigDB revealed networks linked to PPD:DMSO vs. PPD:ALLO DEGs were unique from, and occasionally opposite of, Control:DMSO vs. Control:ALLO DEGs. Similarly, WGCNA revealed statistically significant modules related to either treatment or diagnosis; no modules significant for both PPD and ALLO were observed.

Conclusions: Together, these data highlight both ALLO-independent and -dependent molecular responses in PPD. While the Control:ALLO vs. PPD:ALLO comparisons of the transcriptional response reflect literature supporting ALLO's modulation of synaptic signaling and GABA-related activity, the ultimately weak response of PPD LCLs to ALLO-treatment, especially compared to control LCLs, warrants further investigation. Furthermore, the striking lack of overlap in DEGs between diagnostic groups after ALLO-treatment suggests that ALLO ultimately activates unique and potentially divergent cellular pathways in women with PPD compared to controls. Thus, it may be beneficial to recognize ALLO's ability to play distinct, diagnosis-dependent cellular roles when considering its therapeutic potential.

Keywords: Postpartum Depression, Allopregnanolone, Transcriptomics

Disclosure: Nothing to disclose.

P308. Menstrual Cycle Brain Plasticity: Ultra-High Field 7 T MRI Reveals Changes in Human Medial Temporal Lobe Volume in Female Adults

Rachel Zsido, Angharad Williams, Claudia Barth, Bianca Serio, Luisa Kurth, Frauke Beyer, Veronica Witte, Arno Villringer, Julia Sacher*

Emotion NeuroimaGinG (EGG) Lab, Max Planck Institute for Human Cognitive and Brain Sciences, Clinic for Cognitive Neurology, University of Leipzig, Leipzig, Germany

Background: Female-specific variables, such as pregnancy, menopause, and the menstrual cycle, influence the brain and the risk for mental health disorders, with women at higher risk for cognitive and affective disorders when ovarian hormones rapidly fluctuate and decline. Only 3 percent of studies in the neurosciences, however, are conducted exclusively in females, creating a huge data gap. Here, we provide the first longitudinal high-field MRI dataset of the hippocampus across the menstrual cycle, demonstrating the natural variance of female brain structural plasticity in a region instrumental to memory and affect regulation.

Methods: We performed longitudinal mapping of medial temporal lobe subregion morphology at 6 timepoints across the menstrual cycle in vivo using a dense-sampling protocol, ultra-high field neuroimaging and individually derived segmentation analysis in 27 healthy female participants (19-34 years).

We acquired a total of 138 high resolution images for volumetric calculations of the hippocampus and medial temporal lobe. Using this well-powered within-subject design, we performed novel segmentation analysis of high resolution 7Tesla MRI scans to investigate whether ovarian hormone-modulated volumetric changes manifest differently across subregions of the medial temporal lobe complex. We also quantified cerebral spinal fluid and cerebral blood flow to confirm any volumetric changes were not driven by hormone-related water shifts or blood flow changes.

Results: As hypothesized, linear mixed-effects modeling showed positive associations between estradiol levels and whole hippocampus volume ($\beta = 108.26$, 95% CI = 27 to 190, random effects SD = 174.47, $p = 0.009$). In MTL subregions, the addition of the estradiol*progesterone interaction to the model significantly improved model fit only for the CA1 ($\chi2(1) = 7.691$, $p = 0.006$). Estradiol was positively associated with CA1 volume ($\beta = 42.87$, 95% CI = 21 to 65, $p < 0.001$), progesterone was negatively associated with CA1 volume ($\beta = -150.02$, 95% CI = -249 to -51, $p = 0.003$), and we observed a significant interaction of estradiol and progesterone with CA1 volume ($\beta = 53.06$, 95% CI = 16 to 90, random effects SD = 44.03, $p = 0.005$), such that at higher progesterone levels, the positive effect of estradiol on CA1 volume was attenuated. Progesterone was positively associated with subiculum volume ($\beta = 13.12$, 95% CI = 4 to 22, random effects SD = 43.29, $p = 0.006$) and with Area 35 volume ($\beta = 11.98$, 95% CI = 2 to 21, random effects SD = 44.01, $p = 0.014$). Finally, estradiol was positively associated with parahippocampal cortex volume ($\beta = 24.33$, 95% CI = 10 to 39, random effects SD = 32.48, $p = 0.001$).

Conclusions: We found unique associations between ovarian hormones and CA1, perirhinal Area 35, subiculum, and para-hippocampal cortex volumes across the menstrual cycle. These findings suggest that ovarian hormones alter structural brain plasticity in brain subregions that are differentially sensitive to hormones. Recognizing female brain health as more than just how female brains differ from male brains requires shining a spotlight on female-specific variables, such as the menstrual cycle, as a primary variable of interest. By providing detailed neural phenotyping of brain areas substantially implicated in cognitive and neurodegenerative disease, we establish an important reference: an integrative benchmark for evaluating female MRI biomarkers in health and disease and a prerequisite to ultimately assess depression and dementia risk later in life, pathologies which affect females twice as often as males and more frequently during times of a changing ovarian hormone environment.

Keywords: Human Neuroimaging, Hippocampus, Menopause, Mood, Childhood Stress, Neuroendocrinology, Women's Health, Sex Differences

Disclosure: Nothing to disclose.

P309. Mast Cell Activity Induces Fosb as a Negative Feedback Mechanism to Limit Neuroinflammation and Anaphylactic Response

Natalia Duque-Wilckens*, Dimitri Joseph, Maradiaga Nidia, Srinivasan Vidhula, Szu-ying Yeh, Hari Subramanian, Eric Nestler, Adam Moeser, Alfred Robison

Michigan State University, Bath, Michigan, United States

Background: Brain inflammation plays a central role in neurodegenerative and psychiatric disorders, but the mechanisms by which events such as peripheral infection or environmental stressors result in persistent central neuroimmune activation remain unclear. Mast cells, innate immune cells mostly known for their role in allergy, are uniquely positioned to play a key role in amplifying inflammatory responses leading to chronic brain diseases. In addition to multiple peripheral tissues, mast cells reside in the brain and meninges, are highly sensitive to immune challenges and stress (which can permanently change their activity) and can release an impressive variety of bioactive molecules ranging from pro- and anti-inflammatory cytokines to proteases and neurotransmitters, directly affecting brain physiology. Surprisingly, the transcriptional mechanisms controlling acute-and long-term mast cell responses remain largely unexplored. Based on initial findings that stress and antibody-mediated activation of mast cells dramatically increase the expression of FosB, which encodes the FosB and ΔFosB transcription factors critically involved in long-term modulation of neuronal activity, we used transgenic mice in combination with in vitro and in vivo experiments to test the hypothesis that FosB plays a fundamental role in regulating stimulus-induced mast cell activation and mediator release.

Methods: 1) To create the first mice in which FosB expression is ablated specifically in mast cells (MCFosB-), we crossed the Mcpt5-Cre with the Cre-dependent floxed FosB mouse lines. 2) For in vitro experiments, bone marrow progenitor cells were harvested from femurs of WT and MCFosB-mice and cultured in media supplemented with stem cell factor and Il-3 for 6 weeks to generate bone marrow derived mast cells (BMMC). Next, several approaches were used to assess BMMCs responses to IGE-antigen or lipopolysaccharide (LPS) stimulation: electron microscopy, intracellular Ca2+ measurements, and mediator release. Finally, to confirm the role of FosB in mast cell activity, we overexpressed ΔFosB or its dominant negative binding partner ΔJunD into WT and MCFosB- BMMCs 3) For in vivo experiments, WT and MCFosB-male and female mice were exposed to passive systemic anaphylaxis or LPS injections and rectal temperature, peripheral and central release of inflammatory mediators, and activation levels of mesenteric mast cells were assessed. 4) Finally, to uncover regions of ΔFosB binding in mast cell chromatin, we performed CUT and RUN using baseline and IGE-antigen activated BMMCs, and further combined these data with RNAseq analysis to find potential genes directly regulated by ΔFosB after stimulation. 5) Statistical analyses: samples sizes were n = 3-6 for in vitro studies and 8-10 for in vivo studies. T-test, two-way ANOVAS, or repeated measures ANOVAS were used depending on experimental design.

Results: 1) BMMCs derived from MCFosB- do not express FosB or ΔFosB and, compared to WT, show increased activated appearance at baseline and after stimulation as well as heightened stimulus-induced Ca2+ mobilization and release of proinflammatory mediators ($p < 0.001$). Preliminary data suggest that overexpression of ΔFosB inhibits stimulus-induced mediator release in both WT and MCFosB- BMMCs while ΔJunD exerts the opposite effect. 2) MCFosB- mice show exaggerated hypothermic responses ($p < 0.01$), increased plasma and hypothalamic content of inflammatory cytokines (both $p < 0.05$), and heightened activation of mesenteric mast cells ($p < 0.05$) The overlap of CUT and RUN and RNAseq data suggest that ΔFosB functional targets include dual specificity phosphatase Dusp4 and thymic stromal lymphopoietin TSLP, known regulators of mast cell activity. Currently, we are validating these findings using gene and protein expression analyses.

Conclusions: Taken together, these data show that FosB products exert an inhibitory effect on mast cell activation and proinflammatory mediator release and suggest that this effect could be partly mediated by DUSP1 mediated dephosphorylation of mitogen-activated protein kinases (MAPKs). These results provide a novel negative feedback mechanism of mast cell

regulation relevant to brain and peripheral disorders associated with exacerbated inflammation.

Keywords: Neuroinflamation, ΔFosB, Mast Cells

Disclosure: Nothing to disclose.

P310. Gestational Stress Exposure Effects on Microglia-Synaptic Interactions in the Rat Prefrontal Cortex and Nucleus Accumbens Across the Peripartum Period

Kathryn Lenz*, Courtney Dye, Dominic Franceschelli, Amanda Ringland, Benedetta Leuner

The Ohio State University, Columbus, Ohio, United States

Background: Pregnancy confers a period of heightened vulnerability to mood disorders, with an estimated 20% of new mothers experiencing Postpartum Depression (PPD). The mechanisms contributing to mood dysregulation and impaired maternal care in PPD are not well understood, but stress during pregnancy is a strong risk factor. We previously discovered pregnancy drives microglial changes in brain regions regulating mood and maternal care, including the medial prefrontal cortex (mPFC) and nucleus acccumbens (NAc). Pregnancy also induces widespread neuroplasticity to facilitate motivation and maternal care, and because microglia have been linked to synaptic remodeling in both stressed and non-stressed states, we hypothesize that aberrant microglia-mediated synaptic remodeling may contribute to PPD.

Methods: We used a rodent model of gestational stress that induces a PPD-like phenotype (i.e., behavioral despair, anhedonia, impaired maternal care). Rats underwent chronic variable stress or control handling ($n = 8$-10/group/endpoint) from gestational days (GD)7-20 and were sacrificed on GD21 or postpartum day (PD) 8 and brains collected. In one cohort, qPCR was performed on mPFC and NAc tissue to measure a select panel of synaptic transcripts. In another cohort, brains were sectioned, and immunofluorescent staining performed against a postsynaptic glutamatergic synaptic marker (PSD-95), a marker extracellular matrix protein marker for perineuronal nets (WFA), a pan-microglia marker (Iba1), and a lysosomal marker (CD68) to identify phagocytic microglia. Staining density and 3D co-localization were quantified with ImageJ and IMARIS software. Data were analyzed via 2 way ANOVA and Tukey post-hocs if significant main effects found. All experimental protocols in animal studies were approved by the Institutional Animal Care and Use Committee of The Ohio State University and were conducted in accordance with the NIH Guide for the Care and Use of Laboratory Animals.

Results: Gestational stress led to increased microglia and phagolysosome staining in the mPFC at PD8 ($p < 0.05$) and NAc at both GD21 ($p < 0.05$) and PD8 ($p < 0.0001$). In NAc, gestational stress led to decreased expression of the pre-synaptic glutamatergic marker Vgat1 at GD21 and the general pre-synaptic marker synaptophysin at PD8 ($p < 0.05$). Stress also led to a decrease in PSD-95 immunostaining in the NAc ($p < 0.05$), but no stress-induced differences in microglia engulfment of PSD-95 were observed with Imaris 3D rendering analysis. In mPFC, neither synaptic mRNA expression nor PSD95 immunostaining were impacted by stress exposure. However, we did find changes in perineuronal nets (PNNs), which are extracellular proteins responsible for synaptic stability. We found that PNNs increased from GD21 to PD8 in control animals ($p < 0.0001$), and that gestational stress led to a significant decrease in PNN levels relative to controls at GD21 ($p < 0.001$) and PD8 ($p < 0.01$). However, there were no significant stress effects on microglia engulfment of PNNs in the Imaris 3D rendering analysis. The NAc had few to no WFA-stained PNNs across any group.

Conclusions: In the PFC, PNNs, but not synaptic targets, were decreased by gestational stress exposure. In the NAc, synaptic targets were decreased by gestational stress exposure. Thus gestational stress has region-specific impacts on the synaptic elements it affects, even though both regions showed similar microglial changes. Since PNN and synaptic loss were not mediated by microglia phagocytosis, future work will assess degradation of these synaptic and stabilizing by microglia-derived secreted factors. Microglial-mediated remodeling of perineuronal nets is a novel mechanism for maternal neuroplasticity. Understanding how remodeling of perineuronal following gestational stress may contribute to the underlying pathophysiology of PPD is an exciting avenue for future research and interventional strategies.

Keywords: Postpartum Depression, Microglia, Medial Prefrontal Cortex, Stress, Synapse

Disclosure: Nothing to disclose.

P311. Serotonergic IL-1R1 Modulates Transporter and Neuronal Activity in a Sex-Dependent Manner in Response to Peripheral Inflammation

Paula Gajewski-Kurdziel*, Alaina Tillman, Hideki Iwamoto, Nicole Baganz, Matthew Robson, Ning Quan, Randy Blakely

Florida Atlantic University, Jupiter, Florida, United States

Background: Multiple neuropsychiatric disorders have been found to display immune system alterations (Kerr et al., 2005). The high incidence of comorbidity of neuropsychiatric disorders with chronic inflammatory conditions, such as rheumatoid arthritis, suggests a commonality in the manifestation of these conditions (Evans et al., 2005). Inflammatory cytokines have been found to impact multiple dimensions of neural signaling, from neurotransmitter release and transport, to neural excitability and plasticity (Capuron and Miller, 2011). Forebrain-projecting serotonin (5-HT) neurons of the dorsal raphe (DR) play a key role in regulating behaviors related to mood, fear, sleep, feeding and social interactions. Dysfunction in 5-HT neurotransmission is believed to contribute to multiple neuropsychiatric disorders. Our work is focused on identifying molecular and circuit-level mechanisms that can translate peripheral innate immune system activation into changes in behavior. We have reported that peripheral activation of the innate immune system rapidly increases the activity of the serotonin transporter (SERT), a main determinant of 5-HT neurotransmission (Zhu et al., 2010). Multiple groups, including our own, have demonstrated high level expression of IL-1R1 on 5-HT neurons (Okaty et al., 2015; Liu et al., 2019). Identifying projections and physiological consequences of 5-HT expressing IL-1R1 neurons may clarify how the immune system can influence behavior through discrete circuits and lead to refined treatments for behavioral alterations arising in neuropsychiatric disorders.

Methods: We used adult male and female transgenic mice that allow for conditional elimination of IL-1R1 (IL-1R1loxP/loxP) or restoration of IL-1R1 on an otherwise IL-1R1 knockout background (IL-1R1r/r). Mice were administered LPS (0.2 mg/kg i.p.) or saline and sacrificed by transcardial perfusion three hours after treatment ($n = 8$-12). Brain slices containing DR were immunolabeled with 5-HT and cFos and counts for colocalization were obtained by a blind observer. 5-HT projection sites were stained for cFos (DAB) and cell counts were obtained through automated counting (Nikon NES Elements software). 5-HT uptake assays on midbrain synaptosomes were performed after peripheral LPS with or without the presence of serotonergic IL-1R1. Candidate projection sites were targeted for stereotaxic surgery to inject the retrograde tracer, Fluorogold (FG), in ePet1:Cre;IL-1R1r/r mice to visualize colocalization of serotonergic IL-1R1 (via a transcriptional reporter) with FG. In vivo chronoamperometry was utilized

to determine the effect of local IL-1β (2 ng) on 5-HT clearance in the dorsal hippocampus (CA3) of wild type male mice. Acute brain slice recordings of DR 5-HT neurons were obtained using whole cell patch clamp with current injection used to produced tonic firing. IL-1β was perfused (10 ng/ml) onto the slice and recordings were taken.

Results: We demonstrate nonuniform expression IL-1R1 throughout the DR subregions containing 5-HT neurons. Serotonergic IL-1R1 shows enrichment evident in the dorsal and dorsolateral subregions of the DR. Upon peripheral LPS treatment, males showed an IL-1R1-dependent decrease in serotonergic cFos throughout the DR, whereas females only demonstrated reduced cFos levels in a subset of DR regions. Examination of the median raphe revealed female-specific decreases in serotonergic neuronal activity dependent on the expression of IL-1R1. The lateral habenula (LHb) appears to be a direct target of serotoninergic IL-1R1 neurons, and we observed a female-specific serotonergic IL-1R1-dependent decrease in LHb neuronal activity after LPS. Other brain regions revealed a dependence of serotonergic IL-1R1 expression to maintain baseline neuronal activity. Additionally, we show that the ability of peripheral LPS treatment to stimulate SERT in midbrain synaptosomes is lost in our serotonergic IL-1R1 knockout. Local application of IL-1β in the CA3 subregion of the dorsal hippocampus resulted in increased 5-HT clearance, whereas ex vivo DR slice electrophysiology revealed an inhibition of 5-HT neuron firing in response to IL-1β. Reversal potential analysis suggests a role for membrane $K+$ channel activation as underlying IL-1R1 actions.

Conclusions: Our findings support a growing appreciation that 5-HT neurons contribute to changes in CNS physiology following peripheral immune activation. Our studies are among the first to reveal a sex specificity of serotonergic IL-1R1 on the regulation of 5-HT neuron activity. Since the subregions of DR 5-HT neurons have unique projection patterns, the differences in 5-HT neuron activity post-LPS in males and females suggest a basis for sex-specific differences in projection targets that can drive distinct behavioral responses. Our findings indicate that IL-1β modulates 5-HT neurotransmission both via regulation of SERT and by suppression of 5-HT firing, and thereby 5-HT release. Our current efforts are aimed at investigating the behaviors dependent on serotonergic IL-1R1 activation, as well as elucidating the intracellular signaling cascade of serotonergic IL-1R1 in hopes of revealing targets important for modulating 5-HT intrinsic activity. Our ongoing work will further clarify links between elevated inflammatory signaling and how, by acting through 5-HT pathways, these signals can provide for both normal, health promoting behavioral responses as well as impact risk for neuropsychiatric disorders.

Keywords: Serotonin, Dorsal Raphe, Interleukin 1 Receptor, Interleukin 1beta, LPS

Disclosure: Nothing to disclose.

P312. Type I Interferon Signaling Mediates Chronic Stress-Induced Synapse Loss and Behavioral Deficits

Ashutosh Tripathi*, Alona Bartosh, Danny Perez Sierra, Anilkumar Pillai

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

Background: Inflammation and synaptic deficits have been associated with pathophysiology of many neuropsychiatric disorders. Chronic stress is a major risk factor for many neuropsychiatric conditions and is known to induce inflammation and behavior deficits. Type I interferons (IFN-I) are key players in peripheral inflammatory response and are responsible for mood and behavior deficits. Findings from our published study identified a crucial role of complement component 3 (C3) in IFN-I-mediated changes in neuroinflammation and behavior under chronic stress conditions. C3 is the central hub of complement activation pathways and is known to tag synapses to be eliminated. The receptor of C3 (C3ar1) is highly expressed on microglia and infiltrating monocytes/macrophages. In the present study, we hypothesized that C3ar1 activation is involved in IFN-I signaling-mediated chronic stress-induced synapse loss and behavior deficits.

Methods: To test our hypothesis, we conducted experiments using C3ar1 knockout mice. To block IFNAR signaling, mice were administered intraperitoneally with anti-IFNAR antibody. We used the chronic unpredictable stress (CUS) paradigm in mice. Three chambers social behavior test, Golgi staining for spine density, immunohistochemistry to study microglial activation and synapse pruning, flow cytometry for monocyte infiltration, and qRT-PCR for cytokine measurements were conducted in our experiments. Data were analyzed using two-tailed Student's t-tests (for two-group comparisons) or Analysis of Variance (ANOVA; for multiple-group comparisons). $p < 0.05$ was considered significant. Bonferroni's posthoc test was performed within the comparison.

Results: Our findings show that CUS significantly increased serum IFNβ levels and social behavior deficits. Increased microglial activation and synapse loss were found in the prefrontal cortex (PFC) following CUS. In addition, decreased sociability index was correlated with increased inflammation and decreased synapse numbers in CUS-exposed mice. Peripheral blockade of IFN-I signaling attenuated the above CUS-induced neuro-immune alterations and behavioral deficits. Furthermore, C3ar1 mediates systemic IFNβ-induced neuroinflammation and social behavior deficits.

Conclusions: Our findings identify a key role of C3ar1 in IFN-I-mediated changes in neuro-immune alterations under chronic stress. Collectively, these results support the rationale that targeting peripheral IFN-I pathway represents a promising therapeutic option especially for patients with an elevated immune profile as seen in many depressed subjects. Additional studies are warranted to investigate the brain cell types responsible for the C3ar1-mediated effects.

Keywords: Interferon, Neuroinflammation, Complement Component 3, Synapse Loss, Behavior Deficits

Disclosure: Nothing to disclose.

P313. Chronic Stress Exposure Alters the Gut Barrier Integrity: Sex-Specific Effects on Microbiota and Jejunum Tight Junctions

Ellen Doney, Laurence Dion-Albert, Francois Coulombe-Rozon, Natasha Osbourne, Renaud Bernatchez, Sam Paton, Fernanda Neutzling Kaufmann, Roseline Olory Agomma, Jose Luis Solano Lopez, Raphael Gaumond, Katarzyna Dudek, Joanna Kasia Szyszkowicz, Signature Consortium, Manon Lebel, Alain Doyen, Audrey Durand, Flavie Lavoie-Cardinal, Marie-Claude Audet, Caroline Menard*

Université Laval, Quebec City, Canada

Background: Major depressive disorder (MDD) is currently the most prevalent mood disorder and a leading cause of disability worldwide. However, 30-50% of patients are unresponsive to commonly prescribed antidepressants, highlighting untapped causal biological mechanisms. Dysfunction in the microbiota-gut-brain axis, the bidirectional communication between the central nervous system and gastrointestinal tract, has been implicated in MDD pathogenesis. Aligning with the neuroimmune hypothesis of depression, MDD has a high comorbidity with

inflammatory bowel diseases such as Crohn's disease and ulcerative colitis, suggesting that inflammation-driven gut barrier dysfunction may affect emotion regulation and vice versa. Exposure to chronic stress disrupts blood-brain barrier integrity in a sex-specific manner through loss of the tight junction protein Claudin-5, leading to the development of anxiety- and depression-like behaviors. Still, little is known about intestinal barrier function in these conditions particularly for the small intestine where most food and drug absorption takes place.

Methods: Thus, here we investigate how chronic social (n = 12-20/group/sex) or variable stress (n = 5-10/group/sex), two mouse models of depression, impact the jejunum (JEJ) intestinal barrier in males and females. As chronic stress is the main environmental risk factor for MDD, it is commonly used in animals to alter behaviors and investigate underlying biology. Chronic social defeat stress is a mouse model of depression based on social dominance which produces two distinct phenotypes of stress response: stress-susceptible and resilient mice. The susceptible subgroup display distinct behavioral changes reminiscent of depressive symptoms in humans with increased social avoidance, anxiety, anhedonia, despair, body weight changes, metabolic disturbances, and corticosterone reactivity. Furthermore, loss of BBB integrity occurs only in the brain of susceptible, but not resilient, mice. Another leading stress paradigm is the variable stress model, during which mice are exposed to a repetitive sequence of three stressors, namely tube restraint, tail suspension, and foot shocks. In this paradigm, females and males develop depression-like symptoms at different time points making it a strong model for investigating sex differences. Mice were subjected to stress paradigms followed by analysis of gene expression profiles of intestinal barrier-related targets by quantitative PCR, fecal microbial composition with sequencing studies, and blood-based markers using ELISAs. We also took advantage of machine learning and developed algorithms to characterize in detail tight junction morphological changes. Translation value of a potential gut health biomarker was validated on blood human samples from the Montreal Signature biobank (N = 15-29 individuals/group/sex).

Results: Altered microbial populations as well as changes in gene expression of JEJ tight junctions were observed depending on the type and duration of stress, with sex-specific effects (Claudin-3: social stress for males, two-tailed unpaired t-test $p = 0.0002$ vs $p = 0.288$ for females; variable stress for males, two-tailed unpaired t-test $p = 0.4234$ vs $p = 0.0079$ for females). We confirmed that stress-induced alterations in tight junction gene expression are also reflected at protein level. Thin 6-um slices were double stained with Cldn3 (red) and F-actin (green), and morphological analysis performed using the Imaris software (Claudin-3; two-tailed unpaired t-test $p = 0.0357$). Intriguingly, with unsupervised k-mean clustering of four features of tight junctions - ruffles, width, fragmentation, and diffusion - we identified a cluster of ruffled junctions in stressed animals. Ruffling is associated with inflammation, so we evaluated if LPS injection recapitulates stress-induced changes in the JEJ and observed profound sex differences (Claudin-3: LPS treatment for males, two-tailed unpaired t-test $p = 0.0001$ vs $p = 0.2799$ for females). Finally, LPS-binding protein (LBP), a marker of gut barrier leakiness, was associated with stress vulnerability in mice (for males $p = 0.0033$ after social stress vs for females $p = 0.0188$ after variable stress) and translational value was confirmed on blood samples from women with MDD (two-tailed unpaired t-test $p = 0.7285$ for men vs $p = 0.0434$ for women).

Conclusions: Our results provide evidence that chronic stress disrupts intestinal barrier homeostasis in conjunction with the manifestation of depressive-like behaviours in a sex-specific manner in mice and possibly, human depression. We developed tools and algorithms to analyze in detail tight junction morphological changes and identified circulating LBP as a gut leakiness potential biomarker that could help better diagnose and inform treatment strategies for mood disorders.

Keywords: Social Defeat Stress, Sex Differences, Gut Microbiome, Brain Immune Gut Axis, Cytokines

Disclosure: Nothing to disclose.

P314. Psilocybin Alters Behavior and the Intestinal Microbiota in a Wild Type Mouse Model by Mechanisms That Are Not Fully Dependent on 5HT2A and 5HT2C Receptors

Zachary Cordner*, Emese Prandovszky, Megan Pedicini, Hua Liu, Lindsey Macias, Mikhail Pletnikov, Kellie Tamashiro, Robert Yolken

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

Background: Much enthusiasm has emerged for the therapeutic potential of psilocybin, with growing evidence of remarkable benefit for depression as well as several other psychiatric disorders. However, despite promising clinical data, our understanding of psilocybin's therapeutic mechanisms remains limited. The lack of mechanistic studies appears to be driven, in part, by the assumption that psilocybin's agonism of serotonin receptors in the brain – known to be responsible for the drug's psychedelic effects – also explains its diverse therapeutic benefits, though several recent studies suggest that other mechanisms are likely involved. The microbiome-gut-brain axis, which is increasingly recognized as a driver of behavior and modulator of psychotropic drug effects, is a plausible but largely unexplored target of psilocybin. Here, we begin to assess the effects of psilocybin on behavior, the intestinal microbiota, and the dependence of psilocybin-induced changes on 5HT2A and 5HT2C receptors.

Methods: In the first study, adult male and female C57BL/6 J mice were exposed to a single dose of saline, psilocybin, the 5HT2A and 5HT2C receptor antagonist ketanserin, or psilocybin co-administered with ketanserin. The head twitch response, a validated behavioral measure of central 5HT2A receptor agonism, was measured 30 minutes after treatment. Elevated plus maze, social interaction, and forced swim behaviors were measured between 3 and 6 days after treatment. In a second cohort, male mice were again treated with saline, psilocybin, ketanserin, or psilocybin co-administered with ketanserin. Then, 3 days after treatment, behavior was assessed, and intestinal contents were collected for 16 S rRNA sequencing to determine bacterial composition and diversity. Finally, intestinal contents were collected from mice treated with saline or psilocybin then transplanted by gavage to naive male mice, followed by behavioral analysis.

Results: Psilocybin induced a robust head twitch response, increased exploratory behavior in the elevated plus maze, increased social behavior in the social interaction test, and decreased immobility in the forced swim test. Co-administration of ketanserin fully blocked the head twitch response without significantly altering psilocybin's effects on other behavioral outcomes. In a separate cohort, treatment with psilocybin produced broad alteration of the intestinal microbiome, with particularly marked changes in the large intestine that were only partially blocked by pre-treatment with ketanserin. Finally, transplantation of intestinal contents from psilocybin-treated mice to naive untreated mice resulted in behavioral changes consistent with the effects of psilocybin treatment.

Conclusions: Our findings demonstrate that a single dose of psilocybin leads to behavioral changes in mice that are relevant for studies of resilience and affective disorders. Our results further indicate that the behavioral changes may not be fully dependent on psilocybin's agonism of 5HT2A and 5HT2C receptors. Further, psilocybin appears to broadly alter the intestinal microbiome and transplantation of intestinal contents reproduces behavioral change associated with psilocybin treatment, suggesting a previously unknown microbiome-gut-brain mechanism of action.

Keywords: Psilocybin, Gut Microbiome, Mood and Anxiety Disorders, Translational Animal Models

Disclosure: Nothing to disclose.

P315. Transcriptional Machinery of Microglial Stress Response for Mental Illness Pathology

Masayuki Taniguchi, Kazutoshi Matsushita, Rei Mishima, Mitsutaka Kadota, Shigehiro Kuraku, Tomoyuki Furuyashiki*

Kobe University Graduate School of Medicine, Kobe, Japan

Background: Clinical studies have suggested the presence of neuroinflammation in depressive patients. Rodent studies have demonstrated that chronic stress activates microglia, thereby leading to depression-related behaviors via the induction of proinflammatory molecules. Microglia appear to utilize different yet overlapping molecules in multiple brain areas to cause behavioral alterations. However, histological markers for microglial activation can only capture a limited facet of microglial responses. Thus, microglial responses to chronic stress and their functions remain poorly understood.

Methods: We subjected male C57BL/6 N mice to acute or chronic social defeat stress. We categorized the mice after chronic social defeat stress into susceptible and resilient mice according to the level of social avoidance, a typical depression-related behavior. Then, we isolated microglia from multiple brain areas of these mice, including the medial prefrontal cortex (mPFC), primary sensory and motor cortices, hippocampus, nucleus accumbens (NAc), and hypothalamus, and subjected these cells to single-cell RNA-seq. In addition, we more deeply analyzed the transcriptome of microglia from the mPFC and NAc, two representative brain areas with distinct responses to social defeat stress, with bulk RNA seq. We then predicted transcription factors responsible for stress-induced changes in the microglial transcriptome and examined their functional significance using genetic and surgical manipulations.

Results: Using single-cell RNA-seq of microglia isolated from multiple brain areas in mice, we found that microglial transcriptomes manifested the brain region-specificity and individual variability of stress susceptibility after chronic social stress. Deeper transcriptome analyses of microglia from the mPFC and NAc segregated gene clusters of brain region-specific (local) and non-specific (global) responses to chronic social stress, and the global responses encoded individual variability of stress susceptibility. Epigenomic analyses with H3K27ac ChIP-seq and ATAC-seq predicted distinct transcription factors involved in the respective responses, and surgical and genetic manipulations identified a peripheral stress signal that induced the global transcriptional responses to confer microglia with stress susceptibility.

Conclusions: These findings suggest that microglia integrate local and global stress signals via transcriptional machinery to determine their activation state, promoting mental illness pathology.

Keywords: Social Defeat Stress, Depression, Microglia, Neuro-Inflammation, Medial Prefrontal Cortex

Disclosure: Nothing to disclose.

P316. From Blue to Gray: Inflammation, Aging and Neurodegeneration in Depression

Ebrahim Haroon*, Diana Beltran, Xiangchuan Chen, Evanthia Wommack, Blaine Roberts, Deqiang Qiu, Felicia Goldstein, Michael Treadway, Jennifer Felger, Andrew Miller

Emory University, Atlanta, Georgia, United States

Background: Depressed patients experience a 2-5 times higher risk of neurodegenerative disorders such as dementia (Alexopoulos, 2019; Byers and Yaffe, 2011). However, predisposing risk factors that enable clinicians to stratify risk and initiate preventive measures are unclear. We propose that chronic inflammatory activation in depression promotes and sustains this risk. Our previous data have demonstrated that increased inflammation in depression increases the risk of glutamate toxicity and leads to toxic disorganization of neural systems linked to emotional and cognitive functions (Haroon et al., 2016). Herein, we examined if increased inflammation in the brain as measured in cerebrospinal fluid (CSF) was associated with increases in CSF makers of neurodegeneration in depressed versus controls.

Methods: 54 subjects (35 depressed and 19 non-depressed control subjects) participated in the study and provided CSF samples and clinical and demographic information. Study participants were aged 35-65 and unmedicated with psychotropic medications. No patient was taken off medications for the sake of the study. CSF immune markers were assessed using methods described previously (Felger et al., 2020). CSF neurodegeneration markers were assayed using SIMOA assay on the Quanterix platform (Rissin et al., 2010). The immune marker panel included c-reactive protein (CRP), tumor necrosis factor (TNF), interleukin (IL)-6, and IL-1beta, and their circulating receptors [type 2 TNF (TNFR2), IL-6 (IL6sr), and IL-1 receptor antagonist (IL1ra). The neurodegeneration panel included neurofilament light chain protein (NFL), glial fibrillary acidic protein (GFAP), hyperphosphorylated tau-181 (Tau), abeta-(ab)42 and ab40. Ab42:40 ratio was used to differentiate neurodegenerative from immune effects on amyloid metabolism. Extended regression analyses were used to examine the relationship between CSF immune and neurodegeneration markers across groups, considering subject characteristics as covariates, including age, sex, body mass index, 10-year vascular risk, race, and educational, marital, and occupational status. In addition, structural equation modeling (SEM) was used for mediation analysis.

Results: Of the inflammatory markers, CSF TNFR2 was differentially associated with neurodegeneration markers as a function of depressed group status. Indeed, there was a significant CSF TNFR2 by depressed group interaction that was positively associated with CSF NFL (cf=0.51, $p = 0.015$), CSF GFAP (cf=0.90, $p < 0.001$), CSF Tau (cf=0.50, $p = 0.013$) and negatively associated with CSF ab42:40 ratio (cf = -0.55, $p = 0.005$). No CSF TNFR2 by control group interactions were significantly associated with neurodegeneration markers. Mediation models indicated that CSF TNFR2 indirectly mediated the negative effect of age on CSF Abeta42:40 (cf = -0.25, $p = 0.014$); and the positive effect of age on CSF Tau (cf=0.28, $p = 0.009$), CSF GFAP(cf=0.22, $p = 0.02$), and CSF NFL (cf=0.42, $p < 0.001$).

Conclusions: Ours is among the first studies to concurrently examine CSF markers of neuroinflammation and neurodegeneration in depressed and control subjects. We focused on depressed patients aged between 35 and 65 due to the confluence and build-up of neurodegenerative risk factors in midlife (Marsland et al., 2015). CSF increases in TNFR2 were associated with increases in markers of neurodegeneration in depressed but not control subjects. Profiling proinflammatory activity compounded by the combined risk of aging and depression may better predict the risk

of neurodegeneration. While sex was included as a covariate, this study was not powered to detect sex differences. Treating the risk of neurodegeneration among aging depressed patients is of great interest. Treatment with immune-modulating or neuroprotective agents in addition to well-known antidepressants may be useful in this group.

Keywords: Neuroinflammation, Neurodegeneration, Major Depression, Alzheimers, Late-Life Depression

Disclosure: Nothing to disclose.

P317. Relationship Between Interleukin-6 (IL-6) Blood Levels and Treatment Outcome in Patients With Treatment-Resistant Bipolar Disorder Depression (TRBDD)

Evan Sitar*, James Sinacore, Angelos Halaris

Loyola University School of Medicine, Maywood, Illinois, United States

Background: Immune system activation and inflammatory response have been increasingly linked in the pathophysiology of psychiatric disorders. It has been proposed that stress is associated with sustained activation of the innate immune system, leading to the release of pro-inflammatory cytokines in the nervous system (Haapakoski et al., 2015). Recent literature has identified a potential relationship between alterations in the cytokine network and depression in bipolar disorder (BD) (Halaris et al., 2021). Elevated expression of pro-inflammatory cytokine IL-6 has been positively associated with the subject's experience of depressive and anxiety symptoms, particularly in BD depression (Muneer 2016), and has been proposed to reflect symptom severity and likelihood of treatment response. Furthermore, specific single nucleotide polymorphisms (SNPs) have been shown to influence IL-6 expression and may predispose individuals to depression (Sundaresh et al., 2019). Understanding depressive illness in the context of inflammation triggered by stress may aid in intervention. For the present study, we hypothesized that depression in BD will be accompanied by an elevated blood level of IL-6, and that this elevation correlates positively with the illness severity in BD. Finally, we predicted that the addition of the cyclooxygenase-2 inhibitor and anti-inflammatory celecoxib (CBX), in conjunction with escitalopram (ESC), a selective serotonin reuptake inhibitor, will reduce IL-6 blood levels and improve treatment outcomes in BD patients with depression. We also identified carriers of a particular SNP (rs4553185) among the study's BD cohort and anticipate this SNP will influence IL-6 expression.

Methods: Data was derived from forty patients that completed a 10-week, randomized, double-blind, two-arm, placebo-controlled trial examining the efficacy of CBX combined with ESC in treating TRBDD (Halaris et al., 2020). Both sexes were included. Patients were selected after they met criteria for TRBDD, which included failure to reach remission following 8 weeks of two or more adequate trials of antidepressant drug treatment (Hidalgo-Mazzei et al., 2019). Subjects that met criteria underwent a physical exam, medical history, laboratory tests, and completed several assessment scales. The severity of depressive symptomatology was assessed via the Hamilton Rating Scale for Depression (HAM-D) and Hamilton Anxiety Rating Scales (HAM-A), Beck Depression Inventory, and Beck Anxiety Inventory at Baseline, as well as at Weeks 1, 2, 4, and 8. Patients scoring 18 or higher on the HAM-D 17 scale were randomized and treated with either CBX and ESC, or ESC alone for 8 weeks. Blood samples were obtained via venipuncture for IL-6 at Baseline, Week 4, and Week 8. Patients were either excluded by an inability to adhere to visit schedule or due to having incomplete data sets. An Independent Sample t-test was used to compare IL-6 values between BD patients and Healthy Controls. Depression and anxiety were extracted from assessment scales, and both Pearson correlations and partial correlations were used to examine the relationship between scores and IL-6 levels. Comparison of IL-6 values between CBX + ESC and ESC treatment groups, as well as with respect to treatment response or remission status, were conducted using an analysis of covariance.

Results: There was a statistically significant difference ($p = 0.007$) between mean Baseline IL-6 values in the BD group (Mean ± standard deviation) (1.51 ± 1.30) ($n = 43$) and the Healthy Control group (0.94 ± 0.69) ($n = 53$). No clinical associations were found between IL-6 levels and HAM-D, HAM-A, or other assessment scores. After controlling for Baseline IL-6, comparing the change in IL-6 values from Week 4 to Week 8, as well as the difference between Week 8 IL-6 values, rendered no statistically significant difference between BD patients treated with ESC ($n = 15$) and BD patients treated with ESC and CBX ($n = 25$). Outcome of treatment was categorized into treatment response when patients had a 50% or less reduction in HAM-D 17 score, or higher score than baseline, and into treatment remission when patients had a HAM-D 17 score ≤7. When BD patients were identified in terms of treatment outcome, the adjusted mean Week 8 IL-6 values steadily decreased ($p = 0.074$) when looking from no response (Adjusted Mean ± standard deviation) (1.65 ± 0.20) to treatment response (1.54 ± 0.25), and finally to treatment outcome (1.06 ± 0.17). We also assessed carrier status of the SNP (rs4553185) and distinguished the BD patient cohort between carriers ($n = 35$) and non-carriers ($n = 6$) of the SNP. Results will be presented in the poster.

Conclusions: The significantly elevated Baseline blood levels of IL-6 in BD patients suggests an elevated expression of pro-inflammatory cytokines in patients with depression. This finding supports the hypothesis that immunological dysregulation may play a role in depressive illness. The observation that Week 8 IL-6 values decreased as treatment outcomes improved suggests that IL-6 blood levels may have the potential to predict treatment response. While there was no correlation between IL-6 levels and severity of depression or anxiety, nor was there a difference between IL-6 values in either treatment group, recruiting a larger sample size and extending the duration of treatment may impact these findings and accentuate the role that IL-6 plays in depression.

Keywords: Bipolar Disorder, Treatment-Resistance, Depression Inflammation Cytokine, Celecoxib, Escitalopram

Disclosure: Nothing to disclose.

P318. Assessment of Complement Cascade Components and Ketamine's Mechanism of Action in Patients With Treatment Resistant Depression

Brandi Quintanilla*, Ashutosh Tripathi, Alona Bartosh, Peixiong Yuan, Dede Greenstein, Carlos Zarate, Anilkumar Pillai

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

Background: Over 300 million people world-wide suffer from major depressive disorder (MDD). Unfortunately, only 30-40% of patients with MDD receive remission after conventional monoamine antidepressant therapy. In recent years, ketamine has revolutionized the treatment of MDD, with its rapid antidepressant effects manifesting within a few hours as opposed to weeks with conventional antidepressants. Many research endeavors have sought out to identify Ketamine's mechanism of action in mood disorders, with several studies implicating ketamine's role in neuroinflammation regulation. The complement system is an important component of the innate immune response vital for the

regeneration processes, including neurogenesis. The complement pathway has been implicated in the pathophysiology of depression and studies have shown significant increases in Complement component 3 (C3) expression in the PFCs of depressed suicide subjects. Given complement's role in depression, ketamine's/complement's ability to modulate glutamatergic transmission, and the fund of research highlighting ketamine's anti-inflammatory properties; there is reason to suspect an overlay between the complement system pathway and ketamine's mechanism of action. To investigate this, we hypothesized an increase in baseline complement system levels that are subsequently attenuated by ketamine administration at varying time points.

Methods: Thirty-nine unmedicated individuals with MDD (23 F) and 25 healthy volunteers (HVs, 16 F) participated in a randomized, double-blind trial comparing intravenous ketamine (0.5 mg/kg) to placebo. Blood was obtained at baseline and at three post-infusion timepoints (230 minutes, Day 1, and Day 3). Plasma was then aliquoted into cryotubes and stored at -80 °C until thawed for assay. In this secondary analysis from a placebo-controlled double-blind inpatient crossover ketamine trial, C3a levels were determined by ELISA. Due to notable skew in our data, we log transformed C3 values. We used a linear mixed model with C3 (log ng/ML) as our outcome and included fixed effects of drug (KET, PBO) and drug*time to test our hypotheses. Models also included time as a main effect, sex, age, and baseline C3 as covariates, and a random intercept per person.

Results: We did not detect overall drug differences or differences on C3a levels at any of the time points. The model adjusted overall drug effect was (collapsed over time) (Ket - Pbo = -0.008 (SE = 0.04) t = -0.203 (df=286), p = 0.84). The Ketamine/Placebo difference at each time point was 230 min: Ket - Pbo = -0.0517 (SE = 0.065) t = -0.795(df=282), p = 0.4271; Day 1: Ket – Pbo = -0.0250 (SE = 0.0652), t = -0.384 (df=279), p = 0.7014; and Day 3: Ket - Pbo 0.0531 (SE = 0.0695), t = 0.764 (df=278), p = 0.4458.

Conclusions: Our findings did not show a significant effect of ketamine on plasma C3a levels. Additional analyses on other complement proteins and their association with inflammatory markers are in progress.

Keywords: Treatment Resistant Depression, Ketamine, Complement Pathway

Disclosure: Nothing to disclose.

P319. Serum MCP-1 Protein Levels Correlate With Depression Symptom Severity and Predict Relapse Status in Major Depressive Disorder

Qingqin Li*, Randall Morrison, Maggie Fedgchin, Vanina Popova, Wayne Drevets

Johnson and Johnson Pharmaceutical, Titusville, New Jersey, United States

Background: Major depressive disorder (MDD) is an episodic condition with a relapsing and remitting disease course. Neuroimmune mechanisms have been associated with the pathophysiology of depressive symptoms in subgroups of patients with MDD. The current study investigated the correlation between immune-related protein levels and depression symptom severity and their association with future relapse/recurrence episodes.

Methods: Serum samples were collected from participants with MDD drawn from an observational clinical study OBSERVEMDD0001 (ClinicalTrials.gov Identifier: NCT02489305, n = 422) and three interventional clinical studies SUSTAIN-1 (NCT02493868, n = 462), TRANSFORM-1 (NCT02417064, n = 320) and TRANSFORM-2 (NCT02418585, n = 420). Immune-related proteins were assayed using MRBM InflammationMap protein panel v1.0 consisting of 46 protein analytes. Samples from multiple time points were included in the analysis correlating protein levels with depression symptom severity as measured by Montgomery Asberg Depression Rating Scale (MADRS). Protein analyte level was subject to BoxCox transformation to approximate a normal distribution and Protein analytes with > 80% data missing rate were removed from the analysis. Samples with protein analyte below the lowest limit of detection (LLOD) were set to missing data. A linear mixed model adjusting sample storage age, assay batch (where applicable), subject age, sex, and symptom severity score was fit to assess the relationship between protein levels and depression symptom severity.

For the relapse/recurrence analysis, only the "baseline" stable samples from OBSERVEMDD0001 and SUSTAIN-1 were used. For OBSERVEMDD0001, the "baseline" samples when participants were clinically stable (MADRS ≤ 14) were included in the analysis. For SUSTAIN-1, the week 16 samples after subjects achieved stable remission or stable response (MADRS ≤ 12) at the end of the optimization phase but before randomizing to either continuing esketamine plus antidepressant or switch to placebo plus antidepressant for the maintenance phase were used. In the OBSERVEMDD0001 study, subjects were followed up for up to 2.8 years during which patients continued antidepressant treatment. Clinical evaluations were performed at regular time intervals to detect relapse/recurrence events. Proteins associated with relapse/recurrence in each study/treatment arm were analyzed separately. Meta-analyses were also performed by type of antidepressant or disease population (treatment-resistant depression (TRD) and non-TRD).

Results: ~28 protein analytes were detectable in >80% of the samples. Interleukin 12 subunit p40 (IL12p40) and MCP-1 were associated with MADRS symptom severity score in OBSERVEMDD0001 (β = -6.56×10-3, p = 0.0001, adjusted p-value = 0.003 for IL12p40, and β = 2.38×10-2, p = 0.005, adjusted p-value = 0.07 for MCP-1). The MCP-1 correlation with MADRS score was also observed in both TRANSFORM-1 (β = 5.10×10-3, p = 0.03) and TRANSFORM-2 (β = 4.54×10-3, p = 0.06) studies, but not in SUSTAIN-1. IL12p40 finding was not significant however in other studies.

For the relapse analysis, stable "baseline" samples from 63 relapsers and 154 non-relapsers were retained for OBSERVEMDD0001. Nine proteins were nominally associated with relapse status during follow-up (p < 0.1). For SUSTAIN-1, samples from in the esketamine arm (21 relapsers and 34 non-relapsers) and in the placebo arm (33 relapsers and 48 non-relapsers) were retained in the analysis. Five proteins and 1 protein from esketamine and placebo arm, respectively, were nominally associated with relapse status during the maintenance phase (p < 0.1). Meta-analysis across the two studies (three separate analyses) identified monocyte chemotactic protein 1 (MCP-1) to be consistently predicting relapse status among the three analyses (p = 0.003, adjusted p-value = 0.09).

Conclusions: These results are noteworthy given evidence that chemokines play major roles in mediating interactions between immune cells in the periphery and those within the CNS. We provide evidence that higher MCP-1 levels are associated with worse depression symptom severity scores, which is consistent with a recent meta-analysis indicating that MCP-1 is elevated in depressive subjects compared to healthy controls. Furthermore, baseline MCP-1 is associated with the relapse disease trajectory.

Keywords: MDD, MCP-1/CCL2, Relapse, Depressive Symptoms

Disclosure: Janssen Research and Development, LLC: Employee (Self), Johnson and Johnson: Stock / Equity (Self)

P320. Improvement in Depression and Fluid Cognition Following Accelerated Theta Burst Stimulation for Treatment-Resistant Major Depressive Disorder in Autism Spectrum Disorders

Elizabeth Blank, Steve Wu, Donald Gilbert, John Sweeney, Elizabeth Smith, Lauren Schmitt, Rebecca Shaffer, Meredith Will, Travis Larsh, Paul Horn, Kelli Dominick, Craig Erickson, Ernest Pedapati*

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

Background: Major depressive disorder (MDD) disproportionately affects individuals with autism spectrum disorders (ASD). MDD is a leading cause of functional impairment, social withdrawal, and suicide. Tragically, it is estimated that individuals with ASD have a 4X greater risk of developing MDD and a 9X greater risk of completed suicide. Despite this urgent need, intervention research specifically targeting MDD in ASD is scarce. No medication trials have focused on MDD in ASD. Standard-of-care medications for mood disorders, such as selective serotonin reupdate inhibitors, may be unpredictable or ineffective in individuals with ASD. Repetitive transcranial magnetic stimulation (TMS) is an efficacious, non-pharmaceutical treatment for MDD with strong meta-analysis support, including over 1,000 patients from randomized controlled trials (RCT), and is covered by most insurers. However, repetitive TMS therapy involves 40-minute sessions of high-intensity pulses over a period of 4 to 6 weeks. The lack of evidence in ASD, duration of therapy, and sensory concerns have severely limited the use and scalability of TMS in the ASD population. The study team is highly experienced in using TMS and has piloted an innovative accelerated low-intensity TMS protocol optimal for MDD in ASD individuals.

Methods: This is an interim analysis of an in-progress intervention trial of accelerated theta burst stimulation (aTBS) on MDD in ASD (see NCT05271357). The primary outcome was Hamilton Depression Rating Scale (17-item; HDRS), and the secondary outcome was the NIH Cognitive Toolbox. Exploratory outcomes involved EEG connectivity. The study design is a randomized, active-comparator study of unilateral or bilateral dorsolateral prefrontal cortex (DLPFC) aTBS stimulation (including mandatory two-week lead-in and waitlist control groups). ATBS sessions are administered 3X daily for ten days (30 sessions). The intervention was delivered using a Magstim Horizon (Magstim/EGI, Whitland, UK) using a 70 mm figure-eight EZ cool coil. Resting motor threshold was determined with single-pulse TMS and electromyography. Theta Burst Stimulation: Participants received intermittent theta burst stimulation (50 Hz in 5 Hz bursts) for a total of 600 pulses total at 90% of RMT for each session. We used the Beam F3 localization method. EEG recordings are acquired using Netamp 400 amplifier (MagstimEGI, Eugene, OR) and 128-channel Hydrocel nets in sound-attenuated booths. We performed source estimation using Brainstorm to compute L2-normed, depth-weighted minimum norm estimation source model to generate a current source density (CSD) map to reconstruct time series activations. Standard weighted alpha phase lag index connectivity measures of left prefrontal to right anterior cingulate cortex were estimated for each subject. Statistics: As this is an interim analysis, we examined the effect of aTBS (unilateral and bilateral) on outcome measures. For our primary outcome, we examined HDRS change from baseline immediately, and 4-weeks post-treatment using FDR-corrected paired t-tests. Secondary variables were compared using t-tests. The relationship between changes in frontocingulate connectivity and change in HDRS score was explored using Pearson correlation.

Results: The analysis below reflects the seven subjects who have completed the aTBS treatment and have had at least a 4-week follow-up visit at the time of abstract submission. Since November of 2021, we have screened 14 subjects (2 females), two subjects on the waiting list, one subject scheduled for treatment, and one subject who withdrew before treatment. Our pilot demographics include 57% Caucasian and 14% African American, Hispanic, and Asian. Primary outcome: Mean baseline HDRS ($M = 20.3$, $SD = 3.5$) was significantly reduced immediately following treatment ($M = 7.0$, $SD = 2.8$, $t = 10.5$, FDR $p = 4.4\times10\text{-}5$, Cohen's $d = 3.9$, $n = 7$) and sustained at Week 4 ($M = 5.4$; $SD = 2.7$; $t = 9.7$, FDR $p = 6.9\times10\text{-}5$, Cohen's $d = 3.7$, $n = 7$). Secondary outcome: Baseline Fluid Cognition composite score ($M = 43.1$, $SD = 4.7$) significantly improved at Week 4 following aTBS intervention ($M = 51.4$, $SD = 6.6$; $t = -3.1$, $p = .02$, Cohen's $d = 1.18$, $n = 7$). Exploratory: EEG source analysis was only available on five subjects at the time of the abstract. Reduction of prefrontal to cingulate connectivity following aTBS was predictive of improved HDRS score ($R = .82$, $p = .09$, $n = 5$).

Conclusions: We present early but promising results of using accelerated TBS in a diverse sample of individuals with ASD and MDD. We observed significant improvement in MDD symptoms leading to remission but also improvements in objectively measured Fluid Cognition. We have identified a potential neural biomarker suggesting that left prefrontal (stimulation site) and cingulate connectivity are associated with depression symptoms. The availability of a 10-day non-pharmaceutical intervention for rapid remission of MDD would be highly impactful. The negative sequelae of MDD are particularly devastating, exacerbated by the lack of evidence-based treatments and atypical responses in ASD. A TMS protocol optimized for MDD in ASD, which can be used with commercially available TMS machines, would greatly improve access to care, reduce the need for specialists, and provide rapid recovery as a mid-level option between medications and electroconvulsive therapy.

Keywords: Autism and Depression, Theta Burst Transcranial Magnetic Stimulation, Electroencephalography

Disclosure: Nothing to disclose.

P321. Acute rTMS Effects on EEG-Based Functional Cortical Networks

Camila Cosmo*, Amin Zandvakili, Thaise Toutain, José Garcia Vivas Miranda, Noah Philip

Brown University, Center for Neurorestoration and Neurotechnology, Providence VA Medical Center, Providence, Rhode Island, United States

Background: Repetitive transcranial magnetic stimulation (rTMS) is a non-invasive brain stimulation technique that generates electrical current in the cortex through magnetic pulses, modifying brain networks. Several studies have shown the effectiveness of rTMS in neuropsychiatric disorders, including pharmacoresistant depression. However, these previous trials have mostly indicated therapeutic and neurophysiological effects following a long-term treatment course. Identification of brain-based biomarkers of early therapeutic response remains an important unanswered question in the field. Here we used a novel graph-based analysis method, called Functional Cortical Networks (FCN), to examine electroencephalograpy (EEG) data, hypothesizing that changes would occur early in the rTMS treatment course.

Methods: Resting-EEG activity was measured in fifteen patients with pharmacoresistant depression following five rTMS sessions (5 Hz, 120%MT, 3,000 pulses/session, left dorsolateral prefrontal cortex). Ten minutes of eyes closed; 64-channel EEG was recorded at baseline (pretreatment; T0) and following five rTMS sessions (T1). An FCN model was constructed by applying time-varying graphs and motif synchronization. Each EEG electrode position

was identified as a node, and the synchronization between electrodes is called edge. The main outcome was the weighted-node degree; this parameter reflects how many times the nodes (i.e., EEG electrodes) were synchronized during the EEG acquisition period. Analyses were performed using the EEGLAB/MATLAB software system (Mathworks, Inc.).

Results: A paired t-test showed a significant acute effect of rTMS over the left posterior area, with an increase of 37.825 in the weighted-node degree after five rTMS sessions (95% CI, 468 to 75.181); and marginally significant at the left frontal region (95% CI, -1.663 to 111.981). No other significant changes were observed when hemispheres and additional regions were analyzed ($p \geq .05$).

Conclusions: FCN models might work as a sensitive measure of acute changes in neural mechanisms underlying therapeutic rTMS, as indicated by these pilot findings. Five rTMS sessions were enough to evoke higher synchronization between electrodes in the left posterior and frontal areas, with a statistically significant increase in the first. Our data are accordant with prior trials that have shown that increased left frontal activity might be linked to decreased negative affect, while increased central and posterior activity is thought to reflect increased arousal, which may be involved in reduced fatigue and increased motivation. Based on our findings, FCN models may contribute to further understanding of acute mechanisms underlying rTMS treatments. Future examination will investigate whether early EEG changes can serve as a potential predictor of therapeutic rTMS response.

Keywords: Repetitive Transcranial Magnetic Stimulation (rTMS), Functional Brain Network, Treatment Resistant Depression, Non-invasive Neuromodulation

Disclosure: Nothing to disclose.

P322. Subcallosal Cingulate Deep Brain Stimulation for Treatment-Resistant Depression: Findings From a Double-Blind Randomized Controlled Trial and Open-Label Extension Phase

Sidney Kennedy*, Peter Giacobbe, Sakina Rizvi, Amanda Ceniti, Rima Styra, Andres Lozano

St. Michael's Hospital, Toronto, Canada

Background: Deep brain stimulation (DBS) to subcallosal cingulate (SCG) white matter has been evaluated in 'Treatment-Resistant Depression' (TRD) with mixed findings. Despite a large number of positive findings in open-label studies, randomized controlled trials (RCTs) have so far not supported these results. The primary objective of this study was to compare the efficacy of active vs. sham DBS stimulation to the SCG in reducing depressive symptoms over a 6-month randomized, double-blinded phase, followed by open-label evaluation of outcomes for a further 18 months.

Methods: Thirty-three male and female patients with TRD were enrolled at Toronto Western Hospital, University of Toronto between 2010 and 2016 in this randomized, placebo-controlled double-blind crossover trial. Participants had a current Major Depressive Episode of more than 12 months duration and had documented resistance to at least 4 adequate depression treatments from a minimum of 3 treatment categories in the current episode. Thirty-one of these patients subsequently underwent DBS implantation. Using a Balaam crossover design, participants were randomized to one of four stimulation sequences during two consecutive 3-month treatment phases: ON-ON, ON-OFF, OFF-ON, or OFF-OFF. The primary RCT endpoint was change in Hamilton Depression Rating Scale (HDRS-17) from baseline to 3 and 6 months, with a reduction of 5 points representing a minimum clinically important difference. Response rate was calculated as ≥50% reduction in HDRS-17 score from baseline. At the end of the 6-month randomized phase, all participants received active stimulation for an additional 18 months in an open-label extension phase.

Results: In the double-blind phase, no statistically significant treatment effect was observed for either intent-to-treat ($p = 0.4725$) or as-treated ($p = 0.7397$) analyses, using a linear random-effects model. All four treatment groups demonstrated a reduction in HDRS-17 scores at 3 and 6 months, but this did not significantly differ by stimulation status. No unanticipated adverse device effects or deaths were reported. In the subsequent open-label extension phase, significant reductions in HDRS-17 scores from baseline were observed at 12-, 18-, and 24-months post-implantation (all $p < 0.0001$) across the full sample. Response rates at 12, 18, and 24 months were 64.5% 63.6%, and 71.0%, respectively. Significant and progressive improvements in functional impairment, as measured by the Sheehan Disability Scale, were also observed at 12 months ($p = 0.002$), 18 months ($p = 0.0003$), and 24 months ($p < 0.0001$) post-implantation.

Conclusions: The double-blind RCT phase failed to demonstrate a statistically significant difference between active and sham stimulation. In the open-label phase, the majority of participants responded to DBS, a particularly noteworthy finding in this TRD population. There are several potential explanations for these results: first, the Balaam crossover design may have resulted in an inadequate duration to detect a treatment effect. Second, this design led to a reduced sample size for each treatment condition. Third, the improvements observed with sham stimulation may be related to microlesion (insertion) effects, or finally, due to frequent visits with the study psychiatrist. This study also supports the safety of DBS to SCG for TRD, with reported adverse events consistent with those previously reported in DBS trials.

Keywords: DeepBrain Stimulation, Treatment-Resistant Depression, Randomized Balaam Crossover Design, Open-Label Follow-up

Disclosure: Abbvie: Consultant (Self), Boehringer-Ingelheim: Advisory Board (Self), Janssen: Consultant (Self), Janssen: Contracted Research (Self), Lundbeck: Consultant (Self), Lundbeck: Contracted Research (Self), Lundbeck Institute: Consultant (Self), Merck: Consultant (Self), Otsuka: Other Financial or Material Support (Self), Otsuka: Consultant (Self), Pfizer: Contracted Research (Self), Sunovion: Consultant (Self), Servier: Consultant (Self), Brain Canada: Grant (Self), CIHR: Grant (Self), Ontario Brain Institute: Grant (Self), Strategy for Patient-Oriented Research (SPOR): Grant (Self), Field Trip Health: Stock / Equity (Self), Abbott: Other Financial or Material Support (Self)

P323. The Astrocyte Protein, S100B's Potential Role as a Predictor of Treatment Outcome of Transcranial Magnetic Stimulation for Depression

Andrew Fukuda*, Lauren Hindley, Eric Tirrell, Linda Carpenter

Brown University, Butler Hospital, Providence, Rhode Island, United States

Background: Transcranial Magnetic Stimulation (TMS) is effective for Major Depressive Disorder (MDD) but the therapeutic mechanism of action is not fully elucidated and there is no biomarker used to inform clinicians and patients of the likelihood of a favorable clinical outcome. Astrocytes have been implicated to play role in depression pathophysiology but have not been explored in TMS. S100 Calcium Binding Protein (S100B) is ubiquitously and selectively expressed in the astrocytes and plays a variety of functions including neuroplasticity and neuroinflammation and has been shown to be increased in the serum of depressed patients compared to non-depressed controls and holds promise as a clinical biomarker in TMS. We provide

preliminary data showing association of peripheral levels of S100B and clinical outcome.

Methods: A prospective observational study at an outpatient TMS clinic from consenting patients consisting of a collection of behavioral measures and serum at pre and post TMS treatment (standard clinical treatment targeting left DLPFC daily for 6 weeks) were carried out. The Inventory of Depressive Symptomatology (IDS-SR) as a measure of overall depressive symptom severity. Serum enzyme linked immunosorbent assay (ELISA) for S100B was performed. A repeated measures general linear model (GLM) was performed to determine whether change in S100B concentration differed significantly between those who achieved clinical remission and those who did not with time (pre vs post) as the within-subjects factor and clinical outcome (remission vs non-remission) as a between-subjects factor, with gender and age as covariates. A binary logistic regression was performed to ascertain the effects of age, gender, baseline depression score, and baseline S100B concentration on the likelihood that participants will achieve remission.

Results: 66 patients (26 Males, 44 Females) had pre and post IDS-SR and S100B concentrations. The remission rate was 44%. GLM showed no statistically significant effect observed with time but there was a statistically significant difference between remitters and non-remitters ($F = 4.641$, $p < 0.05$), with a post hoc t-test showing remitters to have lower S100B concentration at baseline ($p < 0.05$, cohen's $d = 1.213$) and post-treatment ($p < 0.05$, $d = 0.445$) compared to non-remitters. The logistic regression model was statistically significant, $\chi2(4)=11.031$, $p < .05$. The model explained 20.6% (Nagelkerke R2) of the variance in achieving clinical remission and correctly classified 65.2% of the cases. Lower S100B concentration prior to treatment course was associated with increased likelihood of achieving remission.

Conclusions: Although S100B did not show a significant change with TMS treatment, those who achieved clinical remission had a lower concentration compared to non-remitters at both timepoints. Furthermore, lower S100B level at baseline was associated with increased likelihood of achieving clinical remission and holds promise as a predictive biomarker of clinical outcome. Studies are underway to look at specific symptomatic domains that S100B may be associated with in depression.

Keywords: Astrocytes, Biomarker, Repetitive Transcranial Magnetic Stimulation (rTMS), Natural Setting, Treatment Resistant Depression

Disclosure: Nothing to disclose.

P324. TMS-EEG Indices as High Potential Predictive Markers of iTBS Response in MDD

Rebecca Strafella, Reza Zomorrodi, Jennifer Lissemore, Yoshihiro Noda, Zafiris Daskalakis, Daniel M. Blumberger, Daphne Voineskos*

Centre for Addiction and Mental Health, Toronto, Canada

Background: Intermittent theta-burst stimulation (iTBS) to the L-DLPFC is effective for treatment-resistant depression (TRD). We indexed transcranial magnetic stimulation-electroencephalography (TMS-EEG) markers, the N45 and N100 amplitudes as well as overall TMS evoked potential activity (GMFA-AUC) at baseline and post-iTBS, comparing separated and contiguous iTBS schedules. TMS-EEG markers were also compared between iTBS responders and non-responders.

Methods: TMS-EEG was analyzed from a triple-blind 1:1 randomized trial for TRD, comparing a separated (54 min interval) and contiguous (0 min interval) schedule of 2×600 pulse iTBS for 30 treatments. Participants underwent TMS-EEG over the L-DLPFC at baseline and post-treatment. 114 participants had usable TMS-EEG at baseline, and 98 at post-treatment. TMS-evoked potential (TEP) components (N45, N100) were examined via global mean-field analysis.

Results: The N100 amplitude decreased from baseline to post-treatment regardless of treatment group ($F(1, 106.02 = 5.20$, $p = 0.02$). There were no changes in N45 amplitude in either treatment group. In responders, the N100 amplitude decreased after iTBS ($F(1,102.13) = 11.30$, $p = 0.001$, pcorrected$= 0.0004$). Responders showed higher post-treatment N45 amplitude than non-responders ($F(1, 94.14) = 4.11$, $p = 0.045$, pcorrected$= 0.016$). Higher baseline N100 amplitude predicted lower post-iTBS depression scores ($F(4, 106) = 6.28$, $p = 0.00014$).

Conclusions: These results further the evidence for an association between neurophysiological effects of iTBS and treatment efficacy in TRD in the largest collected pre/post rTMS TMS-EEG sample to date. Future studies are needed to solidify the predictive potential for clinical applications of TMS-EEG markers.

Keywords: Theta-Burst Stimulation, TMS-EEG, Treatment Resistant Depression

Disclosure: Nothing to disclose.

P325. Dose Dependent Effects of Transcranial Photobiomodulation on Brain Temperature in Major Depression

Akila Weerasekera, Eva Ratai, Jacek Dmochowski, Katherine A. Collins, Aura M. Hurtado, Luis De Taboada, Maia B. Gersten, Julie A. Clancy, Matthew J. Hoptman, Molly K. Irvin, Allison Sparpana, Elizabeth Sullivan, Xiaotong Song, Arwa Adib, Paolo Cassano*, Dan V. Iosifescu

Massachusetts General Hospital, Boston, Massachusetts, United States

Background: Transcranial photobiomodulation (t-PBM) with near-infrared (NIR) light penetrates the cerebral cortex and is absorbed by the mitochondrial enzyme cytochrome c oxidase (CCO), stimulating the mitochondrial respiratory chain. t-PBM also significantly increases cerebral blood flow (CBF) and oxygenation. Small-scale studies have reported that t-PBM may be an effective treatment in major depressive disorder (MDD). However, t-PBM may affect brain temperature, and those effects are unclear. Therefore, possible excessive brain warming during t-PBM is a concern and must be investigated. In this pilot study we evaluated the dose-dependent brain temperature effects of t-PBM in MDD subjects.

Methods: We enrolled 30 adult subjects (age 18-65 years) meeting DSM-5 criteria for MDD, not treatment-resistant (0-2 failed antidepressants in the current episode), either unmedicated or on stable doses of antidepressants, with no other significant medical or psychiatric comorbidities. All subjects underwent three t-PBM sessions with distinct doses (peak irradiance; low: 50, medium: 300, high: 850 mW/cm2, low and medium doses were administered in continuous wave [CW] mode, high dose in pulsed wave [PW] mode) and sham treatment. 1H-MRS data: using a 3 T Siemens Trio MRI scanner, single-voxel (SV) MRS was performed with a PRESS sequence (TE/TR = 30 ms/2 s, dynamic averages = 32×4). A voxel with a volume of 30 mm × 30 mm × 15 mm was placed on the left prefrontal region. Brain temperature (°C) was derived by analyzing the 1H-MRS spectrum chemical shift differences between the water (~4.7 ppm) and NAA (~2.01 ppm) peaks, using jMRUI software's HLSVD method and Zhu et al formula: $Tbrain\ (°C) = 36 - [103.8 \times (\Delta H2O - NAA - 2.6759)]$.

Results: After quality control procedures, the following group numbers were available for both pre- and post- temperature estimations: Sham ($n = 10$), Low ($n = 11$), Medium ($n = 10$), High

(n = 8). We did not detect significant post-irradiation temperature differences between any of the tPBM-active or sham groups (unpaired t-test; p value range 0.105-0.781). We also tested for potential differences in the pre-post variability of brain temperature in each group. As for t-PBM active groups; lowest fluctuation (variance) was observed for the medium-dose (σ2 = 0.29), next for the low-dose (σ2 = 0.47) and the highest fluctuation was for the high-dose (σ2 = 0.67). The sham condition showed the overall lowest fluctuation (σ2 = 0.11).

Conclusions: Overall our MRS thermometry results show that no significant brain temperature elevations occur during the t-PBM application procedure. The brain temperature variations observed pre- and post- t-PBM sessions were not statistically significant. The lowest temperature variation was observed for medium-dose while highest was for the high-dose. These preliminary results indicate a favorable safety profile for t-PBM treatment with NIR in regard to changes in brain temperature.

Keywords: Neuromodulation, Photobiomodulation, Near-Infrared Light, Depression, MR Spectroscopy

Disclosure: Niraxx Light Therapeutics Inc: Advisory Board (Self), Niraxx Light Therapeutics Inc: Board Member (Self), Niraxx Light Therapeutics Inc: Consultant (Self), Niraxx Light Therapeutics Inc: Founder (Self), Niraxx Light Therapeutics Inc: Stock / Equity (Self), Niraxx Light Therapeutics Inc: Patent (Self)

P326. A Novel Cortical Target for Neuromodulation of Reward Network Activity and Affect Regulation

Merage Ghane*, Simona Graur, Michele Bertocci, Henry Chase, Tyler Conrad, Alexander Skeba, Rachel Kaskie, Sabine Janssen, Lisa Bonar, Tyler J. Brady, Fabio Ferrarelli, Mary Phillips

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

Background: Recurring hypo/manic episodes are a characteristic symptom of Bipolar Disorder (BD) and associated with interfering impulsivity, reward sensitivity, and sensation-seeking. Existing treatments come with severe side effects and high relapse rates, highlighting a need for more effective and mechanistically targeted treatments. Elevated left ventrolateral prefrontal (vlPFC) and ventral striatum (VStr) activity during reward expectancy (RE) has shown associations with hypo/mania risk and dysregulated affective states. While noninvasive neuromodulation of subcortical structures is not yet robustly possible, there are cortical targets, such as the vlPFC, that have direct and indirect projections to RE-relevant subcortical circuitry. We aim to further understand the mechanistic role of the vlPFC in RE-related activity, affect regulation, and hypo/mania to identify novel targets for neuromodulatory intervention. To do this, we are examining how inhibitory continuous (c)TBS to the left vlPFC, compared to left somatosensory (control), and sham differentially impacts RE-related activity and acute affect in BD and healthy adults (18-35 years). We hypothesized that one of the three cTBS conditions would be associated with greater decrease in left vlPFC and left ventral striatum (vStr), and that change in activity would be associated with change in affective state.

Methods: To date, 11 adults (73% female) completed all sessions and pre and post-TBS fMRI assessments of RE-related neural activity and affect, using an uncertain probabilistic reward task (Card Guessing Task). We used a priori defined seeds in the left vlPFC and left vStr to test whether one of the three stimulation (stim) conditions showed a greater decrease in RE-related activity from pre-to-post stimulation. Additionally, we used linear regression within stim condition and tested whether change in vlPFC and vStr activity accounted for significant variance in positive and negative affect change from pre-to-post stim. Given that this data is part of an ongoing clinical trial, group and cTBS condition targets are blinded until the study end.

Results: Due to continued data collection, we are primarily focusing results on effect sizes to maintain best practices until sufficiently powered for robust statistical tests. One cTBS condition led to greater decreases in RE-related left vlPFC (partial EtaSq = .11) and left vStr (partial EtaSq = .27) activity vs. the other cTBS conditions. For this condition only, change in left vlPFC activity, but not vStr accounted for a significant portion of the variance in negative (B = .763, p = .048) and positive affect change (B = -.657, p = .01). Change in vlPFC activity accounted for 29.9% of the variance (r2 = .299) in negative affect change (after accounting for positive affect change, full model r2 = .559) and 41.6% of the variance in positive affect change (after accounting for negative affect change; full model r2 = .677).

Conclusions: The left vlPFC is a promising neural target for neuromodulatory intervention in adults with BD. Preliminary results provide proof of concept and suggest that targeted cTBS reduces RE-related activity in both the cortical left vlPFC and subcortical vStr. However, only reductions in RE-related activity in the vlPFC appear to contribute to the regulation of positive and negative affect. Given that data collection is ongoing, we plan to extend these methods to a larger sample and additionally test for a causal role of the vlPFC in hypo/mania related symptoms and reward sensitivity. Future work should test the transdiagnostic relevance of this approach in conditions impacted by affect dysregulation and/or elevated approach motivation/impulsivity more broadly.

Keywords: Theta-Burst Stimulation, Reward Expectancy, Affective Instability, vlPFC, Bipolar Disorder, fMRI

Disclosure: Nothing to disclose.

P327. Imaging Brain SV2A Density Measured With [11 C]UCB-J μPET as a Readout Of Depression and Antidepressant Response in a Mouse Model of Depression

Claire Leroy, Sebastien Goutal, Denis David, Wadad Saba, Maud Goislard, Michel Bottlaender, Fabien Caillé, Phi T Nguyen, Alain Gardier, Rene Hen, Emmanuelle Corruble, Romain Colle, Nicolas Tournier, Indira Mendez-David*

Université Paris-Saclay, UVSQ, Centre de Recherche en Epidémiologie et Santé des Populations (CESP), UMR 1018, CESP-Inserm, ORSAY, France

Background: The high prevalence of major depressive disorder (MDD) and the varying degrees of antidepressant effectiveness justify investigating the molecular determinants of pathogenesis and therapeutics. Several studies provided evidence of reduced synaptic density and cell loss in rodent models of depression. Antidepressant therapies can rapidly induce synaptogenesis and reverse these neuronal deficits. Estimating synaptic density in the living brain is now possible in positron emission tomography (PET) using novel radiotracers targeting the synaptic vesicle glycoprotein-2-A (SV2A). Recently, synaptic density deficiency has been proven in a small number of high severely depressed patients. However, the effect of an antidepressant drug on synaptogenesis in the human brain remains to be evaluated in vivo. This preclinical study aims to profile using SV2A PET imaging the synaptic density with depression-like phenotype and antidepressant drug responses in a well-validated mouse model based on the elevation of glucocorticoids.

Methods: Adult male C57BL/6JRj mice were subjected to chronic corticosterone treatment (CORT, 35 ug/ml) in the drinking water to induce a depression-like phenotype. Mice were thereafter subjected to 4-weeks of selective serotonin reuptake inhibitors (SSRI) antidepressant treatment (fluoxetine, 18 mg/kg/day er) or

Vehicle (VEH). Synaptic density was compared between CORT-Fluoxetine (N = 6), CORT-VEH (N = 6) and VEH-VEH (N = 5) using a 90-min PET acquisition after injection of [11 C]UCB-J (3.32 Mbq ± 1.43), a SV2A radioligand.

Depression score and antidepressant response were assessed by calculating the latency to feed at the Novelty Suppressed Feeding test, before (at 4 weeks) and after (at 8 weeks) antidepressant treatment. Standardized Uptake values ratio between 60-90 min of PET acquisition (SUVr) of [11 C]UCB-J binding were extracted in cortex and hippocampus and normalized to heart blood using Pmod.

Results: [11 C]UCB-J SUVr was decreased in the cortex and hippocampus of CORT-VEH mice compared to VEH-VEH and CORT-Fluoxetine group. [11 C]UCB-J SUV in CORT-Fluoxetine and VEH-VEH groups were not significantly different. Chronic corticosterone (CORT-VEH)-induced increase in depression score was significantly reduced after a 4-week fluoxetine treatment (CORT-Fluoxetine mice) ($p = 0.014$), confirming antidepressant-like efficacy. Interestingly, we observed an inverse correlation between the depression score and [11 C]UCB-J SUVr in the cortex ($p = 0.026$).

Conclusions: This study suggests that the antidepressant effects of SSRIs may be related to presynaptic terminal restoration. Synaptic plasticity might contribute to the neurobiological substrate of depression and may be a target for the management of therapy efficacy. Further investigations using an immunohistochemical study of synaptogenesis is currently ongoing for a better characterization of the biological meaning of this change in [11 C]UCB-J binding.

Keywords: Synaptogenesis, Biomarker, Positron Emission Tomography (PET), Depression, Antidepressant

Disclosure: Scientific/Medical Advisory Board Member: Advisory Board (Spouse)

P328. Variability in Sleep and Activity While Hospitalized Relate to Post-Discharge Outcomes in Youth

Anna Van Meter*, Wei Guo, Sun Jung Kang, Kathleen Merikangas

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

Background: Discharge from inpatient psychiatric care is typically based on observed reductions in patients' externalizing (e.g., manic, psychotic) symptoms and/or resolution of safety concerns related to suicidal or homicidal ideation. However, these markers of discharge readiness are flawed, given high rates of readmission1. Other markers of patients' mental health, including sleep quality and activity patterns, may provide a more valid assessment of stability and discharge readiness. Importantly, measurement of sleep and activity can be made using wearable devices and is not subject to the same biases as self-report (including motivation to feign wellness in order to go home).

We used a wrist-based actigraph, the GENEActiv, to assess stability and quality of sleep and physical activity in adolescent inpatients over the course of hospitalization to determine whether there is an association between these metrics and post-discharge outcomes. We hypothesized that patients who had disturbed sleep and/or inconsistent patterns of physical activity would be more likely to experience clinically significant symptoms one month post-discharge than patients whose sleep and activity are consistent. Additionally, we hypothesized that collecting data with the GENEActiv will prove to be both safe and feasible in the inpatient environment.

Methods: All new admissions to an adolescent inpatient unit who were capable of providing informed consent were invited to participate. Participants completed self-report measures of depression (PHQ-9), mania (GBI-10M), and anxiety (GAD-7) at admission, discharge, and one-month post-discharge. Participants wore a GENEactiv for the duration of their hospitalization. Linear regression, controlling for age and self-reported sex (assessed separately from gender), tested associations between change in symptoms (baseline-to-discharge and baseline-to-follow-up) with actigraph-based sleep and activity metrics while hospitalized.

Results: There were 108 participants with valid actigraph data. Mean age was 15.0 years old (SD = 1.4), 80% were female. Higher average activity during periods of rest were associated with higher average GAD ($\beta = -1.99$, $p = 0.007$; $\beta = -1.59$, $p = 0.044$) and PHQ ($\beta = -3.25$, $p < 0.001$; $\beta = -2.05$, $p = 0.030$) scores at discharge and follow-up, respectively. Variability in activity during rest periods was also associated with less improvement in GAD ($\beta = -2.82$, $p = 0.002$) and PHQ ($\beta = -4.14$, $p < 0.001$) scores at discharge. Similarly, participants who woke up more frequently during their sleep period had higher GAD ($\beta = -0.33$, $p = 0.027$) scores at follow-up and variability in the number of nighttime awakenings was associated with higher GAD ($\beta = -0.65$, $p = 0.019$) and PHQ ($\beta = -0.92$, $p = 0.005$) scores at discharge. Variability in sleep duration was also associated with less improvement in PHQ ($\beta = -2.44$, $p = 0.018$) from baseline to discharge. Participants with high variability in the difference in their level of activity across the day had less improvement in their PHQ scores ($\beta = -39.91$, $p = 0.029$) from baseline to follow-up. Participants who were more sedentary showed less improvement in their GBI-10M scores ($\beta = -0.02$, $p = 0.030$) at follow-up. No other variables were associated with change in manic symptoms. Surprisingly, greater variability in sleep onset time was associated with improvement in GAD ($\beta = 2.32$, $p = 0.027$) and PHQ ($\beta = 6.40$, $p < 0.001$) scores from baseline to discharge. Only 13 participants required urgent care (ED or hospitalization) post-discharge; this outcome was not associated with sleep or activity.

Conclusions: Nighttime agitation and wakefulness were associated with less improvement in depression and anxiety at both discharge and follow-up. Surprisingly, there was no meaningful association between manic symptoms and activity or sleep. The result that sleep onset variability was associated with better outcomes may be related to circadian shifting (i.e., moving from being up all night to sleeping at night) over the course of the hospitalization, rather than day-to-day changes. Future research must evaluate whether stabilization of patients' sleep/activity over the course of hospitalization, rather than averages and standard deviations, relate to outcomes. If so, these metrics have potential to help determine discharge readiness for patients hospitalized for internalizing concerns.

Keywords: Actigraphy, Adolescent, Psychiatric Hospitalization

Disclosure: Nothing to disclose.

P329. Real-Time Assessment of Positive and Negative Affective Fluctuations and Mood Lability in a Transdiagnostic Sample of Youth

Reut Naim*, Shannon Shaughnessy, Ashley Smith, Sarah L. Karalunas, Katharina Kircanski, Melissa A. Brotman

National Institutes of Health, NIMH, Bethesda, Maryland, United States

Background: Emotional lability, or rapid and intense fluctuations in affect, is associated with increased psychopathology and impairment in youth. Although not a diagnostic criterion, emotional lability has been documented in depression, anxiety, and attention-deficit/hyperactivity disorder (ADHD) and is a transdiagnostic risk factor for general psychopathology. However, prior research has primarily relied on retrospective report, focused on negative valanced emotions, and has yet to include participants with Disruptive Mood Dysregulation Disorder

(DMDD), a diagnosis which manifests with emotional lability. The present study leverages real-time ecological momentary assessment (EMA) to examine naturally occurring aberrant shifts in affective states in a transdiagnostic pediatric sample. This is the first EMA study that includes participants with DMDD that also explores both negative and positive affective fluctuations. We had two main aims. First, we wanted to compare emotional lability between different diagnostic groups. We hypothesized that youth with psychopathology would have elevated levels of emotional lability when compared to healthy volunteer (HV) children. We also anticipated that DMDD and ADHD participants would experience greater emotional lability than participants with anxiety disorders (ANX), due to emotional lability being an inherent aspect of these disorders. Our second aim was to compare momentary positive and negative affective fluctuations between diagnostic groups. Based on the previous literature, we hypothesized that patients would have more daily affective fluctuations compared to HV participants. In an exploratory manner, we wanted to assess whether fluctuations in negative and positive fluctuations would be associated with functional impairment across the different diagnostic groups.

Methods: 130 participants (M = 12.55 years, SD = 2.51 years, 70% males, 65.40% White/Caucasian) with primary diagnoses of DMDD, ADHD, ANX, or HV completed a previously validated one-week EMA protocol for irritability (Naim et al., 2021). Participants rated mood change and affective symptoms three times per day for one week. Items of interest included ratings of positive affect, specifically momentary happiness and giddiness, and negative affect, specifically momentary anxiety, anger, and unhappiness. A composite score for each positive and negative component of emotional lability was generated using an unweighted average of all items in each category. Before compositing, we assessed within- and between-person reliability of the EMA-items using multilevel confirmatory factor analyses (MCFA) which yielded overall medium-to high reliability (range: 0.41- 0.89). To capture within-person variations in affective states over time, a computation of root mean successive squared difference scores (RMSSD) for these composite scores was applied. ANOVAs were conducted to compare affect fluctuations among diagnostic groups and linear regressions were conducted to test for their association with functional impairment, which was assessed by clinician-reported measure (Clinical Global Impressions Severity Scale, CGI-S).

Results: Compliance rate with the EMA protocol was high (M = 78.54%, SD = 16.38%), with all groups presenting similar rate. As expected, differences in emotional lability across diagnostic groups were found, with clinical groups exhibiting higher levels of emotional lability compared to HV (all βs > 0.34, ps<0.046). Within the patient groups, DMDD youth demonstrated the most labile and fluctuating negative and positive affect (βs > 0.41, ps<0.049). Emotional lability was associated with global impairment in the whole sample (βs > 0.11, ps< 0.015).

Conclusions: Findings provide evidence that emotional lability is a salient and important mechanism to understand in the context of childhood mood disorders, particularly DMDD. Findings also provide evidence that aberrant fluctuations in positive affect may be just as important as aberrant fluctuations in negative affect, and both are related to psychopathology in youth. Additionally, this study supports added advantages of using real-time in-vivo measures to augment classic, retrospective parent/child or clinician clinical assessments. Targeting labile mood in-vivo may also be a potential treatment for DMDD, which is essential as few treatments for the disorder have been developed. As our sample was largely composed of White/Caucasian males, future work should increase sample diversity, to further examine generalizability.

Keywords: Emotional Dysregulation, Mood Disorders, Ecological Momentary Assessment, Youth, Pediatric

Disclosure: Nothing to disclose.

P330. Predicting the Likelihood of Remission With Antidepressant Medication in Depression: A Practical Patient-Level Machine Learning Approach

Adina Fischer, Scott Fleming, Kelsey Hagan*, Bailey Holt-Gosselin, Alan Schatzberg, Leanne Williams

Columbia University Irving Medical Center, New York, New York, United States

Background: Less than half of patients with major depressive disorder (MDD) experience symptom remission from their first antidepressant medication (ADM) trial. There is limited empirical data to guide ADM selection for an individual patient. As such, clinicians rely on trial-and-error and evidence from what works for the average patient when treating MDD with first-line ADM, which results in symptom persistence and extended morbidity and mortality for those who do not remit. Using state-of-the-art machine-learning methods, we developed a model that identified patient-specific predictors of likelihood remission (and non-remission) with first-line ADM.

Methods: Recursive feature elimination was used to develop a parsimonious (25 feature) gradient-boosted decision-tree model that predicted binary remission (yes/no) from ADM. Features included pre-treatment clinical, demographic, cognitive, and behavioral variables collected from participants (N = 1008) during the baseline visit of the International Study to Predict Optimised Treatment in Depression (iSPOT-D): a randomized, parallel-model, open-label, repeated-measure, longitudinal 8-week trial assessing response to three commonly prescribed first-line ADMs (sertraline, escitalopram, venlafaxine). Remission was defined as a score < 7 on the 17-item Hamilton Depression Rating Scale (HDRS) at a 12-week follow-up. Models were evaluated based on their accuracy and AUROC for predicting remission on a held-out test set using only measurements collected at baseline. Shapley values were used to ascertain which variables most impacted model estimates at the group and individual (patient) levels.

Results: The trained model exhibited performance significantly exceeding random chance on a held-out test set (AUROC = .64, 95% CI = .55 to .71, p < .001; Accuracy = .63, 95% CI = .56 to .70, p = .010). Of participants who were identified as being at high risk of non-remission by our model (i.e., predicted probability of remission < 20%), model accuracy was .71. Features identified by recursive feature elimination to be of greatest predictive value include measures of pre-treatment: depression symptom severity, anxiety symptom severity, impaired cognitive function (verbal memory and information processing speed), impaired identification of facial emotions (fear and disgust), and agitation (facial and motor). This model was compared to models using common linear modeling approaches with the same number of features (25). Elastic net regularization yielded poorer performance on the test set relative to our approach (AUROC = .59, Accuracy = .55), as did use of simpler downstream models, such as canonical logistic regression (AUROC = .54, Accuracy = .55) and logistic regression with elastic net regularization (AUROC = .59, Accuracy = .61).

Conclusions: Using sophisticated machine-learning methods, we developed a model that identified patient-specific predictors of the likelihood of remission from first-line ADM, which could have powerful clinical implications if implemented in outpatient psychiatry or primary care settings. Moreover, findings illustrate the benefit of using sophisticated machine-learning models (gradient-boosted decision trees) over simple logistic regression, as this method improved both model accuracy and interpretability. Results highlight how variables obtained from standard clinical evaluations may be subjected to machine-learning models to assist in developing individually tailored treatment plans, thereby optimizing outcomes and reducing morbidity and mortality for patients with MDD.

Keywords: Antidepressants, Major Depressive Disorder (MDD), Machine Learning, Prediction, Remission

Disclosure: Nothing to disclose.

P331. TMS Doses Based on Motor Threshold Differ Between DLPFC, OFC, and Motor Cortex: A Case for Electric Field Dosimetry in Clinical Studies

Helmet Karim*, Ahmad Mayeli, Francesco Donati, Brian Coffman, Daniel M. Blumberger, Noah Philip, Mary Phillips, Carmen Andreescu, Rebecca Price, Fabio Ferrarelli

University of Pittsburgh Western Psychiatric Institute and Clinic, Pittsburgh, Pennsylvania, United States

Background: Transcranial magnetic stimulation (TMS) is utilized for treating several neuropsychiatric disorders, but outcomes remain variable. The motor threshold (MT) is used to standardize doses but does not account for structural variability in other cortical areas, such as dorsolateral prefrontal cortex (DLPFC) or orbitofrontal cortex (OFC). Electrical field modeling can help estimate those differences. We aimed to compare E-fields of motor cortex, DLPFC, and OFC at MT and 120% of MT, which is the standard dose in most TMS studies.

Methods: We collected T1-weighted images and MT from compulsive behavior disorder patients ($n = 53$) and healthy controls ($n = 12$). We estimated E-fields over motor cortex, DLPFC, and OFC at 100% and 120% of MT. Separate two-way repeated measures ANOVA with post-hoc t-tests were performed.

Results: In the compulsive behavior sample, E-fields at MT in the motor cortex and DLPFC were not statistically different but were greater than OFC. Motor cortex E-fields at MT were less than DLPFC E-fields at 120% MT but was not statistically different when compared to OFC E-fields at 120% MT. A similar pattern was identified in controls.

Conclusions: MT may not be an effective standard for dosimetry in TMS with variable dosing depending on the region of interest. E-field modeling can be used to deliver similar doses of TMS across participants or adjust for differences in dosing, thus allowing personalized TMS treatments.

Keywords: TMS, Electrical Field Modeling, Depression, Neuromodulation, OCD

Disclosure: Nothing to disclose.

P332. Deep Phenotyping in Routine Inpatient Psychiatric Care: Methods, Feasibility, Early Results, Potential Applications

Agustin Yip*, Savannah Layfield, Lucie Duffy, Samantha Wong, Joann Chen, Abdirahman Osman, Fernando Rodriguez-Villa, Steven Gelda, Eliot Gelwan, Philippe Beauchamp, Habiballah Rahimi Eichi, Joshua Salvi, Justin T. Baker, Alisa Busch, Jane Eisen, Kerry Ressler

McLean Hospital, Belmont, Massachusetts, United States

Background: There is increasing acknowledgment of the need for the integration of 'continuous telemetry' and 'deep phenotyping' into routine psychiatric care as a way of generating real-world evidence and quantitative objective measures for the comparative effectiveness and assessing heterogeneity of treatment effects of established and novel interventions. Here we report, to our knowledge, the first instance of concurrently deploying electronic health records (EHR), patient-reported outcome measures (PROMs), actigraphy, ecological momentary assessments (EMA), and biomarker and genomic data collection from individuals admitted for depression/suicidality and anxiety to an academic psychiatric inpatient unit.

Methods: Participants admitted to depression and anxiety inpatient psychiatric units at McLean Hospital completed a battery of self-assessments (depression: QIDS-SR-16, anxiety: GAD-7, functional status: BASIS-24) and screening instruments (borderline personality disorder [BPD]: MSI-BPD, posttraumatic stress disorder [PTSD]: PCL-5, substance use: DAST-10, alcohol use: AUDIT) within 48 hours of admission and within 24 hours of discharge as part of the hospital's Clinical Measurement Initiative (CMI), complementing electronic health records (EHR) data. In addition, patients were offered an opportunity to opt into our genomics (Biobank), wearable device (actigraphy), real-time active and passive assessments of suicidality, and context-adaptive multimodal informatics (CAMI -- audiovisual recording of clinician-patient interactions) initiatives as these modalities were rolled out on to the units. This project received IRB approval.

Results: Over a two-year period, 753 inpatients completed CMI on admission, 552 (73.3%) of whom also reported depression (QIDS-SR-16) and anxiety (GAD-7) symptom severity, and functional status (BASIS-24), upon discharge from the inpatient hospital stay. A large subset ($n = 434$, 57.6%) consented to add-on phenotyping, comprising Biobank ($n = 301$), actigraphy ($n = 134$), CAMI ($n = 24$), and suicidality-related assessments ($n = 28$). Most patients who met inclusion criteria consented to CMI, but uptake was variable for the other phenotyping modalities, reflecting – in part – differences in participant burden and rollout timing.

Participants showed robust improvement in depressive (QIDS-SR-16 mean [SD] admission vs discharge 15.6 [5.6] vs 8.7 [5.0]; $t(530) = 29.27$, $p < 0.001$) and anxiety (GAD-7 mean [SD] admission vs discharge 13.5 [5.9] vs 6.3 [5.1]; $t(529) = 29.46$, $p < 0.001$) symptoms and functional status (BASIS-24 mean [SD] admission vs discharge 37.6 [14.1] vs 21.2 [11.9]; $t(542) = 29.42$, $p < 0.001$) during a relatively brief (mean [SD] = 12.6 days [14.9]) acute inpatient admission. There was a similar improvement in suicidality (BASIS-24 thoughts of ending life $\chi2(10, n = 542) = 300.5$, $p < 0.001$; QIDS-SR-16 thoughts of death and suicide $\chi2(6, n = 545) = 266.0$, $p < 0.001$) and BASIS-24 self-harm thoughts ($\chi2(10, n = 548) = 291.7$, $p < 0.001$).

Screening positive for BPD on the MSI-BPD was associated with worse depressive-symptom and suicidality severity at both admission (18.6 [4.4] vs 14.3 [5.6] $p < 0.001$ and 52.2% vs 32.1% severe to very severe suicidality $p < 0.001$, respectively) and discharge (9.8 [5.4] vs 8.3 [4.8] $p = 0.002$ and 10.0% vs 7.8% severe to very severe suicidality $p < 0.001$, respectively), but not with length of stay (13.4 days [16.1] vs 12.2 days [14.5] $p = 0.4$). Screening positive for PTSD on the PCL-5 was associated with worse depressive-symptom and suicidality severity at admission (19.1 [3.7] vs 13.9 [5.7] $p < 0.001$ and 50.5% vs 33.7% severe to very severe suicidality $p < 0.001$, respectively), length of stay (14.7 days [16.1] vs 11.4 days [13.9] $p = 0.04$), and with depressive symptom severity (10.9 [5.3] vs 7.8 [4.6] $p < 0.001$) but not suicidality severity on discharge (10.1% vs 8.8% severe to very severe suicidality $p = 0.2$).

QIDS-SR-16 composite sleep disturbance at admission was related to increased reports of 'thoughts of ending life' at admission ($p = 0.001$), and 'sleeping too much' on admission was related to 'thoughts of ending life' at discharge. QIDS-SR-16 'sleeping too much' was not associated with actigraphy-derived sleep duration measured up to first 5 nights of wear ($F(3,58)=0.93$, $p = 0.4$). Sleep duration was not associated with QIDS-SR-16 'thoughts of ending life' at admission ($F(4,57)=1.20$, $p = 0.3$) but was inversely associated with 'thoughts of ending life' at discharge ($F(3,58)=6.57$, $p = <0.001$).

Conclusions: The addition of a rotating set of deep phenotyping modalities to EHR data and routine PROM collection is eminently feasible, if challenging, in a well-resourced inpatient setting. Demographic, comorbidity, and actigraphy (sleep) factors modify the effect of inpatient care among those admitted for

depression/suicidality and anxiety. We show robust improvements in a broad range of transdiagnostic, standardized and validated outcome measures during a relatively brief inpatient stay. Early data suggest that quantitative sleep actigraphy may complement self-report measures as a robust predictor of outcomes, in particular suicidal thinking, upon discharge. Overall, these data demonstrate the feasibility of deep phenotyping and quantitative assessments, integrated with routine inpatient psychiatric care, to provide real-world evidence for comparative effectiveness and assessing heterogeneity of treatment outcomes.

Keywords: Digital Phenotyping, Mood Disorders, Suicidality

Disclosure: Nothing to disclose.

P333. High-Throughput Platform for the Discovery of Novel Structural Neuroplasticity Modulators

Pushpa Kumari*, Vasin Dumrongprechachan*, Yevgenia Kozorovitskiy*

Northwestern University, Evanston, Illinois, United States

Background: Structural plasticity at the level of dendrites and dendritic spines, mediated by neuronal activity, critically shapes the wiring of mammalian brain circuits crucial for development and behavior. Deficits in dendritic spine plasticity are central to neurological impairments and mental health disorders. Recent studies characterize role of pharmacological neuroactive agents such as ketamine, psilocybin and entactogens in amelioration of these deficits. Despite the extensive research focused on activity-dependent synaptic plasticity mechanisms, the field lacks screening tools to perform accurate quantitation of synaptic plasticity in a high-throughput format.

Methods: Here we develop, characterize, scale down, and validate a high-throughput screening platform for dynamic measurements of structural plasticity and overcome the bottlenecks in neuroscience drug discovery. The strength of our platform is based on genetically encoded biosensor designs, where the translation of a high-throughput compatible reporter, Luciferase or Nanoluciferase, fused with PSD95(Δ1.2) is driven under the control of synaptic activity-dependent promoters (Arc, cfos). The readout of these sensors—change in luminescence signal—is a direct, quantitative measurement of local translation and potentiation of dendritic spines. The best performing of 3 distinct sensor designs is further developed the study. Cortical and hippocampal mouse neuronal cultures from males and females are used, although the platform is compatible with neurons derived from human pluripotent stem cells. Each experiment generating images, Western Blot data, or neuroplasticity results includes a minimum of 3 biological replicates, with multiple technical replicates. Repeated measures ANOVA are used for most statistical analyses.

Results: Our activity dependent sensors localize in dendritic spine of primary cortical neurons and provide a luciferase readout in response to known positive neuronal activity inducers such as Forskolin (FSK), gabazine and ketamine, in dose dependent manner. Following NIH assay guidelines, the z score (z'> 0.4, range 0.884-0.925) at two different concentrations of FSK and DMSO control ensures the quality and reproducibility of our HTS platform. Screening ~1280 FDA-approved small-molecules followed by unsupervised cut off analysis provided us with a preliminary list of top hits that may positively affect structural plasticity, including previously known and novel modulators. We are validating new hits further using optically-evoked de novo spinogenesis platform to identify novel plasticity enhancers.

Conclusions: Altogether, our platform provides an unbiased high-throughput screening approach that causally associates neuronal activity with an accurate quantitative luciferase readout and allows rapid screening of large numbers of novel neuroactive compounds. This approach should help gain mechanistic insights into signaling pathways for druggable target development for basic neuroscience applications and therapeutics for numerous neurological and mental health disorders.

Keywords: Neuroplasticity, Dendritic Spine, High-Throughput

Disclosure: Nothing to disclose.

P334. Prediction of Antidepressant Response Using Machine Learning and Resting-State EEG

Chao Wang, Wei Wu*, Akshay Ravindran, Vinit Shah, Maimon Rose, Adam Savitz, Amit Etkin

Alto Neuroscience Inc., Los Altos, California, United States

Background: Antidepressant medications such as the selective serotonin reuptake inhibitors (SSRIs) are the first line treatment for major depressive disorder (MDD), yet the response rates remain low, and the current practice still relies on trial-and-error. Here, we sought to develop and prospectively validate a machine-learning (ML) model for predicting individual response to SSRI treatment, using 19-channel resting-state electroencephalography (rsEEG) data from two MDD clinical trials (total $N = 346$).

Methods: Data from 93 MDD patients in the SSRI arm of an open-label clinical trial were used for model development. Features extracted from the pre-treatment rsEEG were used to build an ML model to predict change in Hamilton Depression Rating Scale (HAMD17) scores. The model was evaluated with 10-fold nested cross-validation, and additionally prospectively tested on 42 unseen holdout open-label SSRI patient samples. We also assessed the model's capacity for cross-study prediction by applying the model to the sertraline ($N = 99$) and placebo ($N = 112$) arms of a double-blind randomized clinical trial (RCT), respectively.

Results: We identified a regularized linear regression model that was significantly predictive of the observed HAMD17 score change in the open label study ($r = 0.35$, $p < 0.001$). Applying the model to the holdout samples yielded similar results ($r = 0.32$, $p = 0.019$). The model was also generalizable across studies, being predictive of the outcome in the sertraline arm of the RCT ($r = 0.28$, $p = 0.003$). Importantly, when applying the model to the placebo arm of the RCT, it failed to predict outcome ($r = 0.03$, $p = 0.375$), suggesting that the model was specific to SSRIs (versus placebo).

Conclusions: Our findings demonstrated that individual response to antidepressants can be robustly predicted in a specific manner by using EEG and machine learning. The successful prospective replication on the holdout set, and generalization across studies, demonstrates the promise of precision psychiatry approaches.

Keywords: Depression, EEG Biomarkers, Machine Learning, Treatment Prediction

Disclosure: Alto Neuroscience Inc.: Founder (Self), Alto Neuroscience Inc.: Employee (Self)

P335. Toll-Like Receptor 4 Agonist Elicits an Inflammatory Response After in Vivo Immune Challenge in Major Depressive Disorder

Leandra Figueroa-Hall*, Kaiping Burrows, Jennifer L. Stewart, Ahlam M. Alarm, Chibing Tan, Rayus Kuplicki, T. Kent Teague, Rajagopal Ramesh, Victoria B. Risbrough, Martin P. Paulus, Jonathan Savitz

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

Background: Approximately 1/3 of individuals with major depressive disorder (MDD) display inflammation, but the precise

mechanisms are not understood. While experimental lipopolysaccharide (LPS; endotoxin) studies have shown behavioral, immunological, and physiological changes in healthy individuals, these studies cannot reveal putative, aberrant inflammatory and regulatory mechanisms in MDD, or distinguish between peripheral and central mechanisms. Hence, a mechanistic approach is required to pinpoint which immunoregulatory mechanisms are defective in MDD. One plausible, mechanistic pathway is the toll-like receptor 4 (TLR4) pathway that upon activation with LPS, produces inflammatory cytokines, such as interleukin-6 (IL-6) and tumor necrosis factor alpha (TNF), which are elevated in some individuals with MDD. Here, we examined immune serum markers to identify peripheral changes, and whether these markers are correlated with TLR4-responsive miRNAs isolated from astrocyte-enriched extracellular vesicles (AEEV).

Methods: This preliminary work leveraged a randomized clinical trial (NCT# NCT03142919) involving acute administration of LPS or saline to individuals with MDD and healthy controls (HC). This study was approved by the Western Institutional Review Board, participants' written informed consent obtained, and carried out in accordance with the principles expressed in the Declaration of Helsinki. MDD ($n = 53$) and HC ($n = 18$) were randomized to LPS (0.8 ng/kg) or placebo (saline) and serum samples immunotyped for IL-6 and TNF from baseline (T0), 2 hours (T2), and 24 hours (T24) post LPS/saline infusion using mesoscale discovery. For AEEV experiments, serum was used to isolate total EV, and a GLAST biotin-conjugated antibody used for astrocyte enrichment. AEEV miRNAs from previously published exploratory sequencing data [MDD ($n = 8$); HC ($n = 5$)] were examined for correlations with IL-6 and TNF. Analyses were performed in R Studio, version 1.4.1717.

Results: Preliminary results revealed statistically significant main effects of visit for IL-6 ($F_{(2,203)} = 53.2$, $p < 0.001$, $\eta^2_g = 0.344$) and TNF ($F_{(2,205)} = 44.9$, $p < 0.001$, $\eta^2_g = 0.305$), with pairwise comparisons indicating T2 was significantly different from baseline and T24. There was no significant interaction between the effects of diagnosis and visit for IL-6 and TNF, but there was a significant interaction between the effects of drug and visit for IL-6 ($F_{(2,200)} = 88.0$, $p < 0.001$, $\eta^2_g = 0.468$) and TNF ($F_{(2,202)} = 99.7$, $p < 0.001$, $\eta^2_g = 0.497$). There was also a significant three-way interaction between visit, drug, and diagnosis for TNF ($F_{(2,196)} = 3.28$, $p = 0.04$, $\eta^2_g = 0.032$), but not IL-6. Previously published exploratory sequencing data showed statistically significant changes in several AEEV miRNAs including hsa.let.7 f.5p and hsa.miR.374a.5p, which were decreased at T2 and then resolved back to baseline at T24. Here, for the first time, we show that at T2, hsa.let.7 f.5p has a trending positive relationship with IL-6 ($r^2 = 0.196$) and TNF ($r^2 = 0.183$) and hsa.miR.374a.5p with IL-6 ($r^2 = 0.321$) and TNF ($r^2 = 0.253$).

Statistics: 1. Immune serum markers were analyzed with general linear models with diagnosis [MDD ($n = 53$) vs. HC ($n = 18$)] and drug [LPS ($n = 12$, HC); ($n = 28$, MDD) vs. saline ($n = 6$, HC); ($n = 25$, MDD)] as factors, visit*drug as the primary interaction, and IL-6 as the primary outcome. The Shapiro-Wilk normality test revealed non-normal data for IL-6 and TNF, therefore data were log transformed. A one-way ANOVA revealed significant main effects of visit on IL-6 ($F(2,203) = 53.2$, $p < 0.001$, $\eta^2_g = 0.344$) and TNF ($F_{(2,205)} = 44.9$, $p < 0.001$, $\eta^2_g = 0.305$). A two-way ANOVA indicated significant interaction effects between drug and visit for IL-6 ($F_{(2,200)} = 88.0$, $p < 0.001$, $\eta^2_g = 0.468$) and TNF ($F_{(2,202)} = 99.7$, $p < 0.001$, $\eta^2_g = 0.497$). A three-way ANOVA revealed significant three-way interaction between visit, drug, and diagnosis for TNF ($F_{(2,196)} = 3.28$, $p = 0.04$, $\eta^2_g = 0.032$).

2. Correlations were determined for immune markers and AEEV miRNAs, hsa.let.7 f.5p and hsa.miR.374a.5p; [MDD ($n = 8$); HC ($n = 5$)]. Here, we show that at T2, hsa.let.7 f.5p has a trending positive relationship with IL-6 ($r^2 = 0.196$) and TNF ($r^2 = 0.183$) and hsa.miR.374a.5p with IL-6 ($r^2 = 0.321$) and TNF ($r^2 = 0.253$).

Conclusions: This is the first experimental LPS administration study in depressed individuals. These preliminary findings provide novel insight into LPS-induced inflammatory responses after in vivo immune challenge in MDD and its association to brain-enriched extracellular vesicle miRNAs. In vivo immune challenge elicits temporal-specific inflammatory responses in depressed individuals that trend positively with AEEV miRNAs, which may help elucidate the relationship between peripheral and central mechanisms in MDD.

Keywords: Lipopolysaccharide, miRNAs, Astrocyte-Derived Exosomes (ADE), Major Depressive Disorder (MDD)

Disclosure: Nothing to disclose.

P336. Irritability and Suicidal Ideation in Depressed Individual Across the Lifespan: Clinical Significance and Potential Neurocircuit Mechanisms

Manish Jha*, Cherise Chin-Fatt, Abu Minhajuddin, Madhukar Trivedi

UT Southwestern Medical Center, Dallas, Texas, United States

Background: Recent reports have linked irritability to suicidal ideation (SI) in adults with major depression. Here, we seek to (Study 1) evaluate whether the association between irritability and SI differs based on age, and (Study 2) identify the neurocircuit mechanisms of irritability that mediate its association with SI.

Methods: Study 1: Individuals in VitalSign6 quality improvement project who had irritability [Concise Associated Symptom Tracking irritability domain (CAST-IRR)]and SI [Concise Health Risk Tracking suicidal thoughts factor (CHRT-SUI)] measures were included $N = 2248$). Linear regression analysis with an age-by-CAST-IRR interaction was used to evaluate whether association between irritability and SI differed based on age. In this analysis, CHRT-SUI was the dependent variable, age-by-CAST-IRR was the independent variable of interest, and sex, race, and ethnicity were covariates. To interpret the significant interaction, post hoc analyses were conducted where separate linear regression analysis for pediatric (aged 12 to 17 years) and adult (aged 18 to 64 years) groups were conducted with CHRT-SUI as the dependent variable, CAST-IRR as the independent variable of interest and sex, race, and ethnicity as covariates.

Study 2: Participants of the Establishing Moderators and Biosignatures of Antidepressant Response in Clinical Care (EMBARC) study with magnetic resonance imaging (MRI), CAST-IRR and CHRT-SUI data available were included ($N = 274$). Resting-state functional connectivity (FC) among 121 cortical and subcortical regions were computed ($n = 7260$ FC pairs). Age, sex, race, ethnicity and site were covariates in all regression analyses. Separate linear regression analyses for each of the 7260 FC pair were used to evaluate their association with CAST-IRR, and those with unadjusted p value <0.0005 were then used in mediation analysis. Baron and Kenny approach used to evaluate whether FC pairs accounts for the effect of irritability on SI with age, sex, race, ethnicity and site as covariates. This approach uses three separate linear regression models to evaluate how much of the predictive ability of a variable (irritability) for an outcome (SI) is accounted for by a third variable (FC pair).

Results: Study 1: $N = 1677$ and $N = 571$ individuals were between ages of 12-17 (pediatric) and 18-64 (adult) years, respectively. The age-by-CAST-IRR interaction was significant ($p = 0.0003$) where the association between CAST-IRR and CHRT-SUI was higher in pediatric ($\beta = 0.25$, $SE = 0.03$) versus adult ($\beta = 0.13$, $SE = 0.03$) group.

Study 2: Fifteen FC pairs were associated with irritability at $p < 0.0005$ threshold of which nine FC pairs included the striatum. Functional connectivity of dorsal striatum to lingual and superior temporal regions significantly mediated ($p < 0.05$) the association between symptoms of irritability and SI.

Conclusions: Association between irritability and SI was stronger in youths as compared to adults. Dysfunctions within the striatum may mediate this association, and serve as targets for developing novel circuit-specific treatments. Future studies are needed to replicate and extend these findings, especially in youths with depression.

Keywords: Irritability, Pediatric Irritability, Neurocircuitry, Depressive Disorders, Resting-state fMRI

Disclosure: Acadia Pharmaceuticals: Contracted Research (Self), Janssen Research and Development: Contracted Research (Self), Neurocrine Biosciences: Contracted Research (Self), Navitor/Supernus: Contracted Research (Self), Eleusis: Advisory Board (Self), Eliem/Worldwide Clinical Trials: Consultant (Self), Guidepoint Global: Consultant (Self), Janssen Global Services: Consultant (Self), Janssen Scientific Affairs: Consultant (Self), Medscape/WebMD: Honoraria (Self), Clinical Care Options: Honoraria (Self), NACCME: Honoraria (Self), Global Medical Education: Honoraria (Self)

P337. Hybrid Concept Elicitation and Cognitive Debriefing Patient Interviews to Establish Content Validity of the Dimensional Anhedonia Rating Scale (DARS): Anhedonia is a Core Symptom of MDD; PRO are Important to Assess the Patient Voice

Stephanie Bean, Rahul Dhanda*, Christina Graham, Deborah Hoffman, Mariam Rodriguez, Adrian Ionescu, Stella Karantzoulis, Sidney Kennedy, Sakina Rizvi

Neurocrine Biosciences, Inc., San Diego, California, United States

Background: Anhedonia is a core symptom of major depressive disorder (MDD) and is commonly defined as a loss of interest or pleasure. As a key diagnostic criterion for MDD, anhedonia contributes to patients' health-related quality of life and poor outcomes with current treatments. With this, accurate measurement of anhedonia is crucial to reliably support research efforts and potential pharmacotherapy development in MDD. The 17-item Dimensional Anhedonia Rating Scale (DARS) was developed as a self-reported instrument to address the limitations of existing anhedonia measurements. Notably, the DARS was designed to be generalizable across patient cultures and experiences. To support the available evidence of measurement properties of the DARS, the purpose of this qualitative study was to evaluate and establish content validity of the DARS in adults with anhedonia in MDD through a targeted literature review (TLR), clinician interviews, and patient interviews.

Methods: The TLR was conducted to identify patient-relevant concepts (i.e., signs/symptoms, impacts of signs/symptoms on the patient's day-to-day life and functioning) of anhedonia in MDD and develop a preliminary conceptual model. The model was evaluated and revised through qualitative interviews with clinicians ($N = 6$). The insights derived through the TLR and clinician interviews informed the development of patient interview materials, including a concept elicitation (CE) / cognitive debriefing (CD) discussion guide. The CE exercise was designed to elicit patient-relevant concepts of anhedonia in MDD, while the CD exercise was designed to evaluate patient understanding and use of the DARS via a think-aloud method. One-on-one telephone interviews were conducted with $N = 20$ patients with clinician-confirmed anhedonia in MDD. Interviews were audio-recorded, transcribed, and subsequently coded using qualitative analysis software to identify reported patient-relevant concepts as well as interpretation, clarity, and relevance of the DARS instructions, items, and response options. Reported concepts were analyzed for salience (i.e., most relevant and important to patients), which were then used to finalize the conceptual model. With the final model, an item mapping exercise was conducted, with the goal of evaluating concept coverage of the DARS.

Results: Twelve symptoms and 39 impacts were reported by patients, of which 10 symptoms and 24 impacts were deemed salient and used to finalize the conceptual model. The item mapping exercise revealed that the DARS provided suitable concept coverage in this patient population. CD results demonstrated that the DARS instructions, items, and response options were generally well understood and clear to patients. Additionally, most item concepts were shown to be relevant to patients' experiences; some inconsistencies with specificity or relevance were identified with three items (i.e., two items in the foods / drinks domain, one item in the social activities domain).

Conclusions: Content validity of the DARS was evaluated and confirmed through hybrid CE/CD interviews with patients with anhedonia in MDD. Further performance and other measurement properties of the DARS will be evaluated through planned psychometric analyses.

Keywords: Clinical Outcome Assessments, Patient Reported Outcomes, Content Validity of a Scale, The Dimensional Anhedonia Rating Scale

Disclosure: Neurocrine Biosciences: Employee (Self)

P338. Using Multimodal Neuroimaging to Differentiate Depression in BD From MDD

Ruth Asch*, Ryan Cool, Sophie Holmes, Margaret Davis, Richard Carson, Patrick Worhunsky, Hilary Blumberg, Irina Esterlis

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

Background: Mania/hypomania are a defining diagnostic feature of bipolar disorder (BD), but depression is its most frequent clinical presentation, often leading to misdiagnosis of BD as major depressive disorder (MDD). The metabotropic glutamate receptor 5 (mGluR5) is implicated in cognitive function and mood regulation, thus representing a potential treatment target for disorders associated with glutamate dysfunction, such as BD and MDD. Here we investigate differences in mGluR5 between individuals with MDD, BD, and healthy controls (HC) and examine relationships between mGluR5, brain function, and clinical symptoms.

Methods: Within each study, participants were well matched for demographic characteristics (age, sex, race/ethnicity, smoking status). For Study 1, 62 individuals (37% male, 17 MDD, 27 BD, 18 HC] participated in positron emission tomography (PET) imaging with radiotracer [18 F]FPEB to estimate mGluR5 availability (as estimated by the volume of distribution, VT) in three regions of interest: orbitofrontal (OFC), ventromedial (vmPFC), and dorsolateral prefrontal cortices (dlPFC). Group differences in VT were determined using a multivariate ANOVA with OFC, vmPFC, and dlPFC VT as the dependent variables, and diagnosis (HC, MDD, BD) as the independent variable, followed by Fishers-LSD pairwise comparisons. To evaluate associations between dlPFC mGluR5 availability and clinical variables, we computed Pearson's r. All tests were 2-tailed, and findings were considered significant at $p < 0.05$ level. Statistical tests were conducted in SPSS (v28).

In Study 2, we used functional MR imaging (fMRI) to examine BOLD responses of 48 individuals (48% male, 15 MDD, 14 BD, 19 HC) to fearful displays of facial affect (FEAR) and examined correlations between FEAR responses and dlPFC mGluR5 availability as measured by PET. Processing and analysis of fMRI were

performed in SPM12. The three stimulus conditions (fear, happy, neutral) were modeled separately in an event-related design that included regressors for motion-correction parameters. Group differences in FEAR responses (FEAR > fixation), as well as relationships with dlPFC mGluR5 availability and mood symptoms were examined using standard linear modeling (i.e., one-way ANOVAs with/without covariates of interest). Whole-brain group differences and correlations were investigated at a voxel-level $p < 0.01$ with a cluster-level threshold of puncor<0.05 (kE>600). The mean FEAR BOLD response for significant clusters identified in the whole-brain analyses were extracted (MarsBaR), and ANOVAs with Bonferroni's post hoc tests were used for pair-wise comparisons. Participants in both studies completed clinical scales (MADRS, BDI and POMS) to assess mood symptoms. Cognitive domains of working memory and psychomotor speed were evaluated using the Cogstate testing battery (Groton maze learning test and detection test, respectively).

Results: In Study 1, there was a main effect of group in mGluR5 availability ($F = 2.3$, $p = 0.04$), with lower mGluR5 in BD relative to MDD ($p < 0.01$, -15.6%) and HC ($p < 0.04$, -13.8%). For participants with BD in a depressed mood state ($n = 17$), dlPFC mGluR5 availability was associated with psychomotor speed ($r = -0.6$, $p = 0.03$), and working memory ($r = -0.5$, $p = 0.03$); and depressed mood ($r = -0.7$, $p = 0.005$) and working memory ($r = -0.6$, $p = 0.05$) among subjects with MDD. In Study 2, BD displayed greater FEAR responses in the right temporal-parietal junction ($p = 0.012$, kE=830) relative to HC (padj=0.03) and MDD (padj <0.001). Across all subjects, there was a correlation between dlPFC mGluR5 and the FEAR response in dlPFC/anterior cingulate (ACC) ($p = 0.012$, kE=624). Medial PFC/ACC FEAR response was negatively correlated with mood symptoms in BD ($p < 0.001$, kE=2728), but not MDD.

Conclusions: These data demonstrate differences between BD and MDD in PFC mGluR5 availability and relationships between mGluR5 availability and brain function related to emotion processing that may influence clinically meaningful indices of mood symptoms and cognitive function. Further, mGluR5 PET and fMRI may represent useful tools in the differential diagnosis of BD, and mGluR5 as a potential therapeutic target.

Keywords: Metabotropic Glutamate Receptor 5 (mGluR5), Positron Emission Tomography (PET), Functional Magnetic Resonance Imaging (fMRI), Bipolar Disorder (BD), Major Depressive Disorder (MDD)

Disclosure: Nothing to disclose.

P339. Antidepressants That Increase Mitochondrial Energetics May Elevate Risk of Treatment-Emergent Mania

Mark Frye*, Manuel Gardea Resendez, Brandon Coombs, Marin Veldic, Susannah Tye, Francisco Romo Nava, Aysegul Ozerdem, Miguel Prieto, Alfredo Cuellar-Barboza, Nicolas Nunez, Balwinder Singh, Richard Pendegraft, Alessandro Miola, Susan McElroy, Joanna Biernacka, Eva Morava, Tamas Kozicz

Mayo Clinic, Rochester, Minnesota, United States

Background: Preclinical evidence suggests that antidepressants (ADs) may differentially influence mitochondrial energetics. This study was conducted to investigate the relationship between mitochondrial function and illness vulnerability in bipolar disorder (BD), specifically risk of treatment emergent mania (TEM).

Methods: Participants with BD already clinically phenotyped as TEM + ($n = 176$) or TEM- ($n = 516$) were further classified whether the TEM associated AD, based on preclinical studies, increased (Mito +, $n = 600$) or decreased (Mito-, $n = 289$) mitochondrial electron transport chain (ETC) activity. Comparison of TEM + rates between Mito+ and Mito- ADs was performed using generalized estimating equations to account for participants exposed to multiple ADs.

Results: Adjusting for sex and BD subtype, TEM + was more frequent with antidepressants that increased (24.7%), versus decreased mitochondrial energetics (13.5%, $OR = 2.12$, $p = 0.00002$).

Conclusions: Our preliminary retrospective data suggests there may be merit in reconceptualizing AD classification, not solely based on monoaminergic conventional drug mechanism of action, but additionally based on mitochondrial energetics. Future prospective clinical studies on specific antidepressants and mitochondrial activity are encouraged. Recognizing pharmacogenomic investigation of drug response may extend or overlap to genomics of disease risk, future studies should investigate potential interactions between mitochondrial mechanisms of disease risk and drug response.

Keywords: Bipolar Disorder, Mitochondria, Electron Transport, Mania, Genetic Variation

Disclosure: Myriad: Grant (Self), Assurex Health: Grant (Self), Carnot Laboratories: Other Financial or Material Support (Self), Chymia: Other Financial or Material Support (Self)

P340. The Association Between Cellular Senescence and Clinical Presentation in Late-Life Depression

Johanna Seitz-Holland*, Benoit H. Mulsant, Charles F. Reynolds III, Daniel M. Blumberger, Jordan F. Karp, Meryl A. Butters, Ana Paula Mendes-Silva, Erica L. Vieira, George Tseng, Eric J. Lenze, Breno S. Diniz

Brigham and Women's Hospital, Harvard Medical School, Boston, Massachusetts, United States

Background: Late-life depression (LLD) is common and associated with excessive age-related medical morbidity, cognitive decline, and increased mortality risk. Previous research has suggested that at least a subset of individuals with LLD experience accelerated aging. In addition, previous studies have implicated the role of cellular senescence, a hallmark of biological aging for accelerated aging in LLD. Senescence refers to a state of permanent cell cycle arrest and changes in the cellular secretome, referred to as senescence-associated secretory phenotype (SASP). Previous studies consistently demonstrated an increased expression of SASP proteins in individuals with LLD compared to healthy individuals. However, it is unclear how the increased expression of SASP proteins relates to clinical presentation, including mental and physical health, within individuals with LLD. Therefore, the present study aimed to examine which demographic, physical, and mental health variables are related to peripheral SASP proteins in LLD.

Methods: We examined the relationship between the SASP and demographic, clinical, and cognitive variables in 426 individuals with LLD (mean age 68.9 years, 64% female). We obtained age, self-reported sex, and self-reported race for all individuals. Characteristics of major depressive disorder, including the length of the current depressive episode, age of onset of the first depressive episode, recurrence, and comorbid anxiety disorders, were obtained during the SCID-IV-TR interview. The severity of symptoms was evaluated by the total scores on the Montgomery-Asberg Depression Rating Scale (MADRS), 17-item Hamilton Depression Rating Scale (HAM-D), Brief Symptom Inventory (BSI), Penn State Worry Questionnaire (PSWQ), Anxiety Sensitivity Index (ASI), Scale of Suicidal Ideation (SSI), Medical Outcomes Survey-Mental. We recorded self-reported years of education, Mini-Mental Status Examination (MMSE), Repeatable Battery for the Assessment of Neuropsychological Status (RBANS), and the Delis-Kaplan Executive Function System (DKEFS). In addition, we collected

anthropometric data, pulse and blood pressure, fasting glucose levels, ICD 10 diagnosis of cardiometabolic disorders, Cumulative Illness Rating Scale – Geriatrics (CIRS-G), and the Medical Outcomes Survey- Physical.

We calculated a SASP index based on 22 SASP proteins identified by preclinical studies and our previous publications.

Our statistical analyses consisted of three steps. First, we conducted three factor analyses to group clinical variables into domains related to 1) depression and anxiety characteristics, 2) cognitive functioning, and 3) physical health. Using regression analyses, we then explored the association between the SASP index and age and sex. Last, we examined the association between the SASP index and the factors determined in step one, computing correlation and regression analyses.

Results: Factor analyses revealed two factors related to depression and anxiety (34.69% and 15.10% variance accounted for), one related to cognitive functioning (55.64% variance accounted for), and three related to physical health (27.08%, 17.33%, 13.21% variance accounted for). The physical health 1 comprised the CIRS- G and Medical Outcomes Survey- Physical scales, BMI, glucose, waist/hip ratio, and physical comorbidities.

A regression analysis demonstrated a significant effect of age ($T = 5.24$, $p < .001$, standardized Beta = 0.24) and sex ($T = -4.11$, $p < .001$, standardized Beta = -0.19). Older and male participants presented with a higher SASP index.

A higher SASP index correlated with worse cognitive functioning ($r = -0.18$, $p = 0.001$), and worse physical health 1 ($r = 0.41$, $p < .001$).

Regression analyses with the SASP index as the dependent variable and age, sex, and the six factors as independent variables showed a significant effect of age ($p < .001$), sex ($p = 0.0050$), and physical health 1 ($p < .001$). Physical health 1 was also the most significant independent variable when splitting our sample into males ($p < .001$) and females ($p < .001$).

Conclusions: Our findings highlighted the role of physical health in accelerated aging in LLD. In addition, the SASP index was associated with age and sex, and cognitive functioning. In contrast, the SASP index was not related to the severity or chronicity of depression and anxiety characteristics. This finding does not contradict our previous studies that have consistently demonstrated an increased SASP index in individuals with a major depressive disorder compared to non-depressed older adults. However, it suggests that the SASP index is more closely associated with physical health and cognitive functioning within individuals with LLD. Therefore, our findings are consonant with a viewpoint that depression is an unwanted co-traveler with co-occurring medical burden and associated disability. They further suggest that future treatment efforts should include interventions and lifestyle modifications that target general health, such as weight loss and exercise programs and optimized control of chronic medical conditions such as diabetes, hypertension, and hypercholesterolemia. Furthermore, longitudinal studies are needed to understand if the inhibition of cellular senescence is a promising treatment target in LLD.

Keywords: Depression, Accelerated aging, Senescence

Disclosure: Nothing to disclose.

P341. An Exploratory Machine Learning-Based Approach to Predicting Outcomes With Intravenous Racemic Ketamine Treatment of Major Depressive Disorder

Albert Arias*, Richard Feinn, Patrick Oliver

Virginia Commonwealth University, Richmond, Virginia, United States

Background: We recently reported a large (424 patients) retrospective case series of depressed subjects (most of which had treatment-resistant depression) treated with intravenous ketamine in regular clinical practice (Oliver et al., in press). Good remission (38%) and response (72%) rates were achieved but logistic regression did not identify any convincing significant predictors of response based on available demographic and phenotypic variables. Similar to other recent efforts to predict treatment response in depression (Lin et al., 2021, Athreyu et al., 2019) we sought to use machine learning on our extensive database to identify a model for predicting ketamine response and important variables for the prediction.

Methods: The patients were adults with Major Depression $N = 400$ (those of the 424 with complete outcomes data) and were recruited from the community as part of regular practice at private clinics. Ketamine infusions were administered at a starting dose of 0.5 mg/kg/40 minutes for six infusions within 21 days and continued as needed with titration for the effect of partial disassociation as per the clinical course, further details are available in the forthcoming publication. The PHQ-9 was the primary outcome measure and was completed at intake, prior to each infusion, and then every two weeks via electronic form. Response (>50% improvement from baseline) and remission (PHQ-9 score <5) were used as the categorical outcomes. The Random Forest (RF) package in R Studio was used to generate models of outcomes based on 25 available demographic and other phenotypic variables (e.g., sex, age, suicidal ideation, BMI, etc.).

Results: The RF model for remission was fairly accurate in predicting outcomes with an area under the curve (AUC) = 75.1%, and accuracy of 73.5%, with the model for response somewhat less accurate; AUC = 71%, accuracy 68%. For remission, the most important variables in the model (most with > 5% mean decrease in Gini index) were; baseline PHQ-9, total number of infusions, age, dosage per kg average, BMI, baseline anxiety, current use of benzodiazepine, a history of an alcohol problem, and suicidal ideation, amongst others). The most important variables in the response model were quite similar.

Conclusions: This preliminary exploratory analysis suggests that a set of variables that are easily and quickly obtained at intake can provide a good estimate of the probability of response and remission with ketamine treatment for prospective patients with MDD. It could also potentially be used to help estimate the ideal number of infusions to optimize the probability of response in a patient. These models could also potentially be improved upon by discovering additional important variables (e.g., neurobehavioral traits, transdiagnostic phenotypic features, genetic markers) in future studies. Though there is clear room for improvement, these findings suggest further study of these precision medicine methods may advance the field.

Keywords: IV- Ketamine, Ketamine, Treatment Resistant Depression, Machine Learning, Precision Medicine for Depression

Disclosure: Nothing to disclose.

P342. Efficacy of Adjunctive Cariprazine in Patients With Major Depressive Disorder and Anxiety Symptoms: A Post Hoc Analysis

Maurizio Fava, Vladimir Maletic, Chen Chen, Julie Adams, Ken Kramer, Majid Kerolous*

AbbVie, Madison, New Jersey, United States

Background: Anxiety symptoms are common in patients with major depressive disorder (MDD). The presence of these symptoms makes diagnosis and treatment of MDD more difficult and can negatively impact treatment outcomes. Cariprazine is a dopamine D3-preferring D3/D2 and serotonin 5-HT1A receptor

partial agonist that is approved to treat adults with schizophrenia or manic, mixed, and depressive episodes associated with bipolar I disorder. The efficacy of cariprazine as adjunctive treatment for patients with MDD and inadequate response to antidepressant treatment (ADT) alone has been evaluated in late-stage clinical studies. This post hoc analysis evaluated the effects of adjunctive cariprazine on symptoms of depression in patients with MDD and anxiety symptoms using data from a positive phase 3 fixed-dose, randomized, double-blind, placebo-controlled trial (NCT03738215).

Methods: Patients with MDD and inadequate response to ongoing ADT were randomized to placebo + ADT, cariprazine 1.5 mg/d + ADT, or cariprazine 3 mg/d + ADT for 6 weeks of double-blind treatment; the primary efficacy outcome was change from baseline to week 6 in Montgomery-Åsberg Depression Rating Scale (MADRS) total score. Post hoc analyses evaluated change from baseline in MADRS total score in subgroups of patients with and without baseline anxiety symptoms. The anxiety subgroup included patients with a baseline score ≥7 on the Hamilton Depression Rating Scale (HAM-D) Anxiety/Somatization factor (sum of 6 items: psychic anxiety, somatic anxiety, gastrointestinal somatic symptoms, general somatic symptoms, hypochondriasis, and insight). Analyses used a mixed-effects model for repeated measures; statistical significance was determined using the 95% confidence interval (95% CI) associated with the least squares mean difference (LSMD) versus placebo.

Results: A total of 751 patients were included in the intent-to-treat (ITT) population and post hoc analysis. In the overall ITT population, the difference in MADRS total score change from baseline to week 6 versus placebo + ADT was statistically significant after multiplicity adjustment for cariprazine 1.5 mg/d + ADT (-14.1 vs -11.5; adjusted P = .0050), but not for cariprazine 3 mg/d + ADT (-13.1; P = .0727). In post hoc analysis, 83% of patients (627/751) met criteria for anxiety at baseline. In patients with baseline anxiety, least squares (LS) mean change from baseline to week 6 in MADRS total score was significantly greater for cariprazine 1.5 mg/d + ADT versus placebo + ADT (-14.1 vs -11.7; LSMD [95% CI] = -2.4 [-4.2, -0.7]); for cariprazine 3 mg/d + ADT, LS mean change from baseline in MADRS total score was -13.1 (LSMD [95% CI] vs placebo + ADT = -1.5 [-3.2, 0.3]). In the smaller subgroup of patients without baseline anxiety (n = 124), LS mean changes from baseline in MADRS total score were numerically greater for cariprazine 1.5 mg/d + ADT (-12.8) and 3 mg/d + ADT (-10.1) compared with placebo + ADT (-9.8), but LSMDs (95% CIs) versus placebo did not reach statistical significance for either cariprazine dose (1.5 mg/d = -3.0 [-7.3, 1.3], 3 mg/d = -0.4 [-4.5, 3.7]).

Conclusions: The high percentage of patients with anxiety symptoms in the positive fixed-dose study of cariprazine as adjunctive treatment in MDD supports previous findings that anxiety is a common feature of MDD. In post hoc analysis, cariprazine 1.5 mg/d + ADT compared with placebo + ADT was associated with significantly greater improvement in depressive symptoms in the subgroup of patients with MDD and baseline anxiety. In patients without baseline anxiety, the effect size versus placebo for cariprazine 1.5 mg/d + ADT was comparable to that observed in patients with baseline anxiety, though differences were not statistically significant, which was likely due to small sample size in this group. These outcomes suggest that adjunctive cariprazine was efficacious in reducing depressive symptoms in patients with MDD whether or not they had anxiety symptoms at baseline.

Keywords: Cariprazine, Dopamine, Major Depressive Disorder, Anxiety

Disclosure: AbbVie: Employee (Self)

P343. Comparative Effectiveness of Medications in Bipolar Disorder in Real-World Settings Based on 60,045 Patients

Markku Lähteenvuo*, Tapio Paljärvi, Antti Tanskanen, Heidi Taipale, Jari Tiihonen

Niuvanniemi Hospital, Kuopio, Finland

Background: Bipolar disorder is a severe psychiatric disorder with a prevalence of 2-3% for the bipolar spectrum. The treatment patterns of bipolar disorder have changed during the recent years, with mood stabilizer usage decreasing and the usage of antipsychotics increasing. However, information of the comparative effectiveness of these different treatment options especially in real-world settings is still sparse.

Methods: We identified anyone between the age of 16-65 years diagnosed with bipolar disorder in Finland (ICD-10: F30-F31) during the years 1987-2018 using Finnish nationwide registries and excluded anyone with a diagnosis of schizophrenia spectrum disorder (ICD-10: F2X) or dementia (F00-F03, G30) before their diagnosis of bipolar disorder (resulting cohort size: n = 60,045, 56.4% female, mean age 41.7 years (Standard Deviation (SD) 15.8 years). A sub-cohort of patients newly diagnosed with bipolar disorder and without use of antipsychotics or mood stabilizers one year prior to their first diagnosis of bipolar disorder was also identified (incident cohort, n = 26,395, 54.9% female, mean age 38.2 years (SD 13.0 years). Hospitalizations for psychiatric (ICD-10: FXX) and non-psychiatric (ICD-10: any other than F-diagnosis) reasons were used as proxy outcome measures for relapse and safety, respectively. Medication use was modelled using the Finnish Prescription registry and the established PRE2DUP method on a day-by-day basis. Exposures to medications were compared against non-exposure to the same medication group (non-exposure served as reference, for example: exposure to long-acting injectable antipsychotics (LAI) vs. non-use of antipsychotics or exposure to lithium vs. non-exposure to mood stabilizers). The hazard ratios (HR) with 95% confidence intervals (95% CI) for the outcome measures as per medication exposures were than calculated by using Cox hazard models during the follow-up years 1996-2018 using advanced within-individual models to eliminate selection bias. Exposures with less than 50 person-years of use were not reported. Separate analyses were run for the whole cohort and the incident cohort. The hazard ratios were adjusted (aHR) for the following covariates: other psychotropic medications used, order of treatment, time since diagnosis of bipolar disorder.

Results: A total of 104,093 psychiatric hospitalization events were recorded during the follow-up dispersed between 26,159 individuals. The medications associated with lower risk of psychiatric hospitalizations were olanzapine LAI (aHR 0.54, 95% CI 0.37-0.80), haloperidol LAI (aHR 0.62, 95% CI 0.47-0.81), zuclopenthixol LAI (aHR 0.66, 95% CI 0.52-0.85), lithium (aHR 0.74, 95% CI 0.71-0.76), clozapine (aHR 0.75, 95% CI 0.64 – 0.87), carbamazepine (aHR 0.81, 95% CI 0.75-0.87), levomepromazine (aHR 0.88, 95% CI 0.83-0.93), lamotrigine (aHR 0.88, 95% CI 0.85-0.92), valproic acid (aHR 0.89, 95% CI 0.87-0.92), pregabalin (aHR 0.92, 95% CI 0.86-0.98) and chlorprothixene (aHR 0.93, 95% CI 0.86-0.99). Medications associated with a statistically higher risk were quetiapine (aHR 1.03, 95% CI 1.00 – 1.06) and ziprasidone (aHR 1.26, 95% CI 1.07-1.49). The results were mostly similar for the incident cohort.

A total of 144,434 non-psychiatric (somatic) hospitalization events were recorded during the follow-up dispersed between 33,380 individuals. Of the studied medications, only lithium (aHR 0.77, 95% CI 0.74-0.81) and carbamazepine (aHR 0.91, 95% CI 0.85-0.97) were associated with significantly reduced risk of non-psychiatric hospitalizations, whereas risperidone (aHR 1.07, 95% CI

1.02-1.13), olanzapine (aHR 1.10, 95% CI 1.05-1.15), quetiapine (aHR 1.10, 95% CI 1.07-1.13), haloperidol (aHR 1.12, 95% CI 1.03-1.22), melperone (aHR 1.19, 95% CI 1.03-1.35), pregabalin (aHR 1.25, 95% CI 1.19-1.31), gabapentin (aHR 1.28, 95% CI 1.20-1.38), clozapine (aHR 1.29, 95% CI 1.07-1.55), ziprasidone (aHR 1.36, 95% CI 1.06-1.75) and haloperidol LAI (aHR 1.43, 95% CI 1.01-2.03) were associated with significantly increased risk of non-psychiatric hospitalizations. Results were similar for the incident cohort.

Conclusions: Lithium and certain long-acting injectable antipsychotics were associated with best outcomes and should be favoured. Quetiapine and ziprasidone were associated with significantly increased risk for both psychiatric and non-psychiatric hospitalizations.

Keywords: Bipolar Disorder, Antipsychotic Treatment Practice, Mood Stabilizers, Pharmacoepidemiology

Disclosure: Janssen: Honoraria (Self), Janssen-Cilag: Honoraria (Self), Lundbeck: Honoraria (Self), Otsuka: Honoraria (Self), Recordati: Honoraria (Self)

P344. Pharmacogenetic and Pharmacokinetic Modelling for Precision Prescribing of Venlafaxine in Late-Life Depression

Daniel Mueller*, Xiaoyu (Betsy) Men, Zachary Taylor, Victoria Marshe, James Kennedy, Eric Lenze, Benoit Mulsant, Charles Reynolds, Laura Ramsey

University of Toronto, Toronto, Canada

Background: Late-life depression (LLD) is characterized by major depressive disorder (MDD) in adults older than 50-65 years old. LLD is frequently associated with medical comorbidities (e.g., cardiovascular and cerebrovascular disease) and cognitive decline. More than 50% LLD patients fail to achieve remission or have a high risk of relapse once remitted. Therefore, predictive models using genetic biomarkers will be helpful in guiding and optimizing LLD treatment. Venlafaxine (VEN) is a commonly prescribed antidepressant for LLD treatment. VEN is primarily metabolized to its active metabolite o-desmethylvenlafaxine (ODV) by the cytochrome P450 (CYP) 2D6 enzyme. Given that both VEN and ODV are pharmacologically active, the sum of VEN and ODV is frequently referred to as the active moiety (AM) and analyzed as a single pharmacological entity. In this study, we aim to utilize a pharmacokinetic (PK) modelling method, which can estimate key PK parameters using sparse data across multiple time points to account for interindividual variability, to assess 1) whether CYP2D6 genotypes contribute to different VEN PK parameters, and 2) the relationship between VEN treatment responses and drug exposure.

Methods: We investigated individuals that participated in the Incomplete Response in Late-Life Depression: Getting to Remission (IRL-GRey, NCT00892047). During the IRL-GRey study phase 1, participants received open-labeled VEN for 12 weeks, starting from 37.5 mg/d and titrated to 300 mg/d if tolerated. Remission was defined as a MADRS score ≤10 at both of the final two consecutive visits. The presence of any adverse effect was also recorded using UKU rating scale at the end of phase 1. We used the existing guideline by Caudle et al.to transfer CYP2D6 genotype to metabolizer status by summing the activity values of haplotypes. Individuals were identified as CYP2D6 poor metabolizers (PM), intermediate metabolizers (IM), normal metabolizers (NM), and ultra-rapid metabolizers (UM). We adapted and modified a pharmacokinetic (PK) model previously described by Lindauer et al. in NONMEM. Our PK model was adjusted for body weight and CYP2D6 metabolizer status. We analyzed whether different CYP2D6 metabolizers had different VEN clearance and exposure of VEN, ODV, and AM using one-way ANOVA. Further, we built regression models to assess the association between treatment responses and drug exposure. The significance level was p-value < 0.05.

Results: The PK model ($n = 304$) demonstrated good predictive performance for VEN and ODV after adjusting for CYP2D6 metabolizer status and body weight. The four metabolizer groups had significantly different model-estimated VEN clearance, VEN exposure, and ODV exposure, while there was no difference across metabolizer status in AM exposure.

Before dose adjustment, higher VEN exposure, higher ODV exposure, and higher AM exposure were all associated with poorer treatment efficacy ($n = 295$). After dose-adjustment, no association was found between treatment efficacy and dose-adjusted VEN exposure, or dose-adjusted AM exposure. The overall presence of adverse effect was associated with higher VEN exposure, higher ODV exposure, and higher AM exposure ($n = 287$). For specific adverse effects, orthostatic dizziness was associated with higher VEN exposure and higher AM exposure. Notably, PMs had the highest rate of orthostatic dizziness. Higher AM exposure was associated with the presence of nausea/vomiting.

Conclusions: We adapted a transit PK model which fitted our data well. Our results showed significant impacts of CYP2D6 on estimated VEN PK parameters. By predicting PK parameters prior to treatment, extreme CYP2D6 metabolizers, especially CYP2D6 PMs, can benefit from genetic testing to avoid treatment-induced adverse effects.

Keywords: Late-Life Depression, Pharmacogenetics, Pharmacokinetics, Venlafaxine

Disclosure: Nothing to disclose.

P345. Categorical Improvement Across Bipolar Depression Symptoms: Pooled Analyses of Cariprazine Randomized Phase II/III Trials

Roger McIntyre, Lakshmi Yatham, Eduard Vieta, Lauren Aronin, Binh Nguyen*

AbbVie, Madison, New Jersey, United States

Background: Bipolar I disorder (BP-I) is a chronic mood disorder characterized by an admixture of manic, hypomanic, and depressive symptoms; depressive symptoms are the leading cause of morbidity and time spent unwell. Cariprazine is a dopamine D3-preferring D3/D2 and serotonin 5-HT1A receptor partial agonist that is approved to treat manic, mixed, and depressive episodes associated with BP-I. The efficacy of cariprazine in BP-I depression was established in 3 fixed-dose, randomized, double-blind, placebo-controlled phase 2/3 trials (NCT01396447, NCT02670538, NCT02670551). Data from these trials were pooled for post hoc analyses to evaluate clinically relevant symptom improvement using the 10 individual items of the Montgomery-Åsberg Depression Rating Scale (MADRS).

Methods: Patients were randomized to fixed-dose cariprazine 0.75 mg/d (1 study only), 1.5 mg/d, or 3 mg/d; the primary endpoint in each study was MADRS total score change from baseline to week 6. For each individual MADRS item, post hoc analysis was conducted to determine the percentage of patients that shifted from mild or worse baseline symptoms (MADRS item score ≥2) to minimal or no symptoms (MADRS Item Score <2) at week 6; in a more rigorous analysis, the percentage of patients that shifted from moderate or worse baseline symptoms (MADRS Item score ≥4) to mild or better symptoms (MADRS Item Score ≤2) at week 6 was also evaluated. Individual MADRS item scores range from 0-6, with higher scores indicating greater symptom severity. Cariprazine 1.5 mg/d and 3 mg/d were pooled for post hoc analysis; cariprazine 0.75 mg/d was not included because it is not within the recommended dose range for cariprazine. Odds ratios

(ORs) and 95% confidence intervals (95% CI) were calculated for each comparison of cariprazine versus placebo.

Results: The pooled intent-to-treat population included a total of 1383 patients (placebo=460; cariprazine=923 [1.5 mg=461, 3 mg/d = 462]). Across 9 of 10 MADRS items, 67.5% to 100% of patients had mild to severe baseline symptoms (score ≥2), and 25.2% to 82.3% of patients had moderate to severe baseline symptoms (score ≥4); a lower percentage of patients had baseline Suicidal Thoughts (score ≥2 = 14.2%; score ≥4 = 0%), which was likely due to exclusion criteria in the original studies. On 8 of 10 MADRS items, a significantly greater percentage of cariprazine- versus placebo-treated patients shifted from mild or worse severity to minimal or no symptoms at week 6 (OR [95% CI]): Apparent Sadness (1.6 [1.2, 2.0]; P = .0005]), Reported Sadness (1.6 [1.2, 2.0]; P = .0005), Reduced Sleep (1.5 [1.1,1.9]; P = .004), Reduced Appetite (1.4 [1.1, 1.9; P = .011), Concentration Difficulties (1.5 [1.1, 2.0]; P = .005), Lassitude (1.4 [1.1, 1.8]; P = .006), Inability to Feel (1.5 [1.2, 1.9]; P = .001), and Pessimistic Thoughts (1.5 [1.2, 2.0]; P = .001); no significant differences were observed on the Inner Tension and Suicidal Thoughts items. In patients with moderate or worse baseline symptoms, a significantly greater percentage of cariprazine- versus placebo-treated patients shifted to mild or better symptoms on 7 of 10 MADRS items (OR [95% CI]): Apparent Sadness (1.8 [1.4, 2.3]; P < .0001), Reported Sadness (1.5 [1.2, 2.0]; P = .008), Reduced Sleep (1.3 [1.0, 1.7]; P = .005), Reduced Appetite (1.8 [1.1, 2.9]; P = .002), Concentration Difficulties (1.6 [1.2, 2.1]; P = .002), Lassitude (1.6 [1.2, 2.1]; P = .001), and Inability to Feel (1.5 [1.1, 2.0]; P = .006); no significant differences were observed on the Inner Tension and Pessimistic Thoughts items and no patients had a score ≥4 on the baseline Suicidal Thoughts item.

Conclusions: A significantly greater proportion of patients treated with cariprazine versus placebo shifted to a lower category of severity symptom across most individual MADRS items. This suggests that cariprazine was associated with clinically meaningful improvement across a broad range of depressive symptoms in patients with BP-I depression.

Keywords: Bipolar I Depression, Bipolar I Disorder, Cariprazine, Depression, MADRS

Disclosure: AbbVie: Employee (Self)

P346. Precision Functional Brain Mapping to Understand the Mechanisms of Psilocybin

Joshua Siegel*, Dean Wong, Demetrius Perry, Timothy Laumann, Abraham Snyder, Marcus Raichle, Eric Lenze, Nico Dosenbach, Ginger Nicol

Washington University, Saint Louis, Missouri, United States

Background: Animal models suggest that limbic plasticity induced by psilocybin may be key to its antidepressant effects. To understand how the neurotrophic and psychological effects of psychedelics relate, it is necessary to measure psilocybin-induced plasticity in humans.

Methods: In this cross-over study, we employed precision functional mapping (PFM) and diffusion basis spectrum imaging (DBSI) to measure changes in functional connectivity (FC) and inflammation, respectively, during and after psilocybin exposure in healthy adults (18-45 years). Participants received 25 mg of psilocybin (PSIL) and 40 mg of methylphenidate (MTH) 1-2 weeks apart. Participants underwent numerous imaging sessions prior to, during (60 minutes following injections), and in the weeks after drug sessions. Primary outcomes were imaging tolerability (defined as scan completion below a strict motion cutoff), subjective experience (e.g., Mystical Experiences Questionnaire), and limbic DBSI and FC.

Results: Six adults (3 female) completed the study. Precision resting fMRI was acquired roughly every other day over approximately 1 month, for a mean of 34 scans (SD 5) per subject. All participants completed scans conducted during PSIL sessions, with high-quality obtained in 5/6 participants. Measurable differences between groups on the MEQ were observed across all four factors (t-test; $p < 0.05$).

In the cortex, PSIL produced a decrease in BOLD signal power across regions that was reflected in reduced neurovascular response during task fMRI. In the limbic system, PSIL produced in increase in FC between structures (hippocampus, amygdala, Nacc, subgenual cingulate) that could not be attributed to neurovascular or respiratory changes. Persisting changes in hippocampal-frontal FC were observed 1 day after dosing. No persisting changes (after vs before) were observed in network modularity or signal power. Participants showed a trend towards decreased hippocampal DBSI restriction fraction (marker or inflammation) following PSIL exposure (linear mixed effects model, main effect of timexdrug; $p = 0.08$).

Conclusions: These results demonstrate that treatment-related changes in FC and inflammation, measured via PFM and DBSI methods, respectively, occur following PSIL exposure and are observable and distinguishable from MTP in healthy younger adults. This study establishes the feasibility of precision imaging during acute PSIL exposure.

Keywords: Psilocybin, Resting State Functional Connectivity, Precision Psychiatry, Hippocampus, Inflammation

Disclosure: Nothing to disclose.

P347. Both Levomilnacipran and Duloxetine Potently Inhibit Serotonin Reuptake Throughout Their Therapeutic Dose Range in Healthy Male Volunteers

Stephen Daniels, Katerina Nikolitch, Jonathan S. James, Jennifer L. Phillips, Pierre Blier*

University of Ottawa Institute of Mental Health Research, Ottawa, Canada

Background: Medications that potently inhibit serotonin (5-HT) reuptake and/or norepinephrine (NE) reuptake are effective in the treatment of major depressive disorder (MDD). Serotonin/NE reuptake inhibitors (SNRI) are first-line treatment for MDD, but for both venlafaxine and duloxetine, higher doses are required to inhibit the NE reuptake process than those necessary to potently inhibit 5-HT reuptake. In a previous analysis, it was shown that levomilnacipran, but not duloxetine, inhibits NE reuptake throughout its therapeutic range. Using positron emission tomography (PET), racemic milnacipran occupies only 60% of 5-HT transporters at its minimal effective dose in MDD (100 mg/day), whereas duloxetine occupies 84% administered in similar conditions. There is no such data for the occupancy of 5-HT transporters by levomilnacipran. An alternative approach to evaluate 5-HT reuptake activity has been to assess the depletion of whole blood 5-HT as 90% of 5-HT in the blood is in the platelets, which do not synthetize 5-HT, but have a 5-HT transporter that is identical to those on 5-HT neurons.

Methods: Healthy male participants (18-40 years of age) were initially randomized to take either placebo for 21 days, levomilnacipran (40 mg/day for 7 days, increased to 80 mg/day for 7 days, and then 120 mg/day for 7 days), or duloxetine (60 mg/day for 7 days, 90 mg/day for 7 days, and 120 mg/day for 7 days). Participants could prolong administration periods to allow for adaptation to side effects. The concentration of whole blood 5-HT was determined using high performance liquid chromatography from samples collected at steady state levels of the drugs.

Results: There were 10 completers in the placebo group, 10 for levomilnacipran, and 9 for duloxetine. Two participants withdrew due to side effects, one in each of the active treatment arms. Baseline levels of 5-HT ranged between 400 and 500 picomol/ml and remained unaltered in the subjects who received placebo. There was a robust depletion of 5-HT after 7 days with both duloxetine (75%) and levomilnacipran (85%), which were not significantly different from each other. Higher regimens of both drugs lowered 5-HT levels further and were below the detection level of our assay (F2, 6: 6.13; $P < 0.001$).

Conclusions: These findings replicate our previous results using whole blood 5-HT depletion to assess the degree of 5-HT reuptake inhibition that duloxetine produces at 60 mg/day. Such results are also consistent being PET imaging in the brain. Levomilnacipran was equipotent with duloxetine in blocking 5-HT reuptake also at its minimal effective dose in MDD. Higher regimens of levomilnacipran and duloxetine led to further decreases of 5-HT levels consistent with prior results using larger doses of the SNRI venlafaxine. Taken together with prior results assessing NE reuptake inhibition in this group of healthy participants, these assays confirm that duloxetine is a SNRI, potently inhibiting 5-HT reuptake first and requiring a titration to 120 mg/day to inhibit NE reuptake. In contrast, levomilnacipran acts as a dual reuptake inhibitor from its minimal effective regimen in MDD.

Keywords: Serotonin Transporter, Peripheral Blood Marker, Serotonin and Norepinephrine Reuptake Inhibitor

Disclosure: Abbvie: Advisory Board, Grant, Honoraria (Self)

P348. Brexanolone Therapeutics in Post-Partum Depression Involves Inhibition of Systemic Inflammatory Pathways

A. Leslie Morrow*, Irina Balan, Riah Patterson, Giorgia Boero, Holly Krohn, Todd O'Buckley, Samantha Meltzer-Brody

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

Background: Brexananolone (FDA approval, Feb 2019) has fast, long-lasting and remarkable efficacy (>60% HAM-D score improvement) in the treatment of post-partum depression (PPD). Treatment requires hospitalization and substantial patient expense. Understanding the therapeutic mechanism(s) offer insight to address the core pathology in PPD. We test the hypothesis that Brexanolone inhibits proinflammatory immune signaling and macrophage sensitivity in PPD patients to promote clinical recovery.

Methods: PPD patients under treatment at UNC hospitals provided blood samples before and after Brexanolone infusion according to the FDA-approved protocol. All patients were under concurrent treatment with other psychotropic medications, but unresponsive prior to Brexanolone therapy. Serum was collected to determine neurosteroid levels and isolated cell lysates were examined for inflammatory markers at baseline as well as sensitivity to the inflammatory activators lipopolysacharide (LPS) and imiquimod (IMQ).

Results: Brexanolone infusion alters multiple neuroactive steroid levels, inhibits inflammatory mediators, and desensitizes inflammatory response to promote clinical improvement. Brexanolone infusion increased allopregnanolone (+2443 pg/ml, 95% CI 2009-2711, Bonferroni's $p = 0.0001$) and allotetrahydrodeoxycorticosterone (+63.4 pg/ml, 95%CI 48.3-78.4, Bonferroni's $p < 0.001$), while decreasing 3α,5α-androsterone (-738 pg/ml, 95%CI -1243 to -232, Bonferroni's $p = 0.03$) and 3α,5α-androstandiol (-23.1 pg/ml, 95%CI -33.9 to -12.4, Bonferroni's $p < 0.001$) in a manner that was highly correlated with HAM-D score. Brexanolone infusion inhibited tumor necrosis factor-α (TNF-α, 58%, $p = 0.03$), and interleukin-6 (IL6, 61% $p < 0.04$), but not interferon gamma inducible protein-10, and these effects were correlated with HAM-D score improvement (TNF-α: Pearson $r = 0.61$, $p < 0.06$; IL6: Pearson $r = 0.68$, Bonferroni's $p < 0.03$). Furthermore, Brexanolone infusion prevented the activation of TNF-α, IL1β and IL6 by both LPS and IMQ, suggesting a desensitized inflammatory response. Brexanolone infusion partially inhibited the LPS-induced elevation of IL-1β (-53.6%; Bonferroni's $p = 0.03$), IL-6 (-50.4%; Bonferroni's $p = 0.03$), and TNF-α (-31.3%; Bonferroni's $p = 0.02$). Brexanolone infusion also partially inhibited the IMQ-induced elevation of IL-1β (-48.9%; Bonferroni's $p = 0.02$), IL-6 (-47.0%; Bonferroni's $p = 0.02$), and TNF-α (-32.4%; Bonferroni's $p = 0.048$. Pearson correlation indicated that the inhibition of LPS- or IMQ-induced elevation of IL-1β, IL-6 or TNF-α was positively correlated with HAM-D score improvement.

Conclusions: The data suggest that Brexanolone therapeutic actions involve the inhibition of inflammatory mediator production through desensitization of immune cells and changes in endogenous neurosteroid levels in blood. These data support the idea that inflammation plays a key role in post-partum depression and inhibition of specific inflammatory pathways underlies its therapeutic efficacy.

Keywords: Allopregnanolone, Inflammatory Markers, Depression

Disclosure: Sage Therapeutics: Grant (Self), UNC Chapel Hill: Patent (Self)

P349. Investigation of the Pharmacological Properties of 2-Br-LSD, a Non-Hallucinogenic LSD Analog

Adam Halberstadt*, Yasmin Schmid, Emma Bonniwell, Janelle Lanham, Abdi Ghaffari, Andrew Cao, Hooshmand Sheshbaradaran, John McCorvy

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

Background: Psychedelic drugs such as lysergic acid diethylamide (LSD) and psilocybin induce hallucinogenic effects via 5-HT2A receptor activation. Over the last two decades, considerable clinical evidence has emerged indicating that LSD and psilocybin have therapeutic efficacy against a range of psychiatric disorders, including depression, anxiety, headache, pain, and substance abuse. Similar to LSD, the closely related derivative 2-bromo-LSD (2-Br-LSD, TD-0418A) is also being investigated as a potential treatment for depression and headache. The therapeutic efficacy of 2-Br-LSD is somewhat surprising, however, because 2-Br-LSD does not produce hallucinogenic effects in humans and reportedly acts as a 5-HT2A receptor antagonist.

Methods: A combination of in vitro and in vivo pharmacological studies were conducted with 2-Br-LSD to assess its receptor interactions and potential mechanism-of-action. The interaction of 2-Br-LSD with 33 monoaminergic GPCR targets was assessed using G protein-dissociation and β-arrestin2 recruitment BRET assays. Head-twitch response (HTR) studies were conducted in male C57BL/6 J mice to determine whether 2-Br-LSD has an LSD-like behavioral profile ($n = 5$-8 mice/group). The HTR is rapid rotational head movement induced by psychedelic drugs in mice via 5-HT2A receptor activation. The HTR was detected in mice using an automated method based on artificial intelligence.

Results: Compared to LSD, 2-Br-LSD shows considerably less off-target activity at GPCRs. The presence of a single bromine atom at the 2-position of LSD leads to partial agonist activity at

several GPCRs, including the 5-HT2A subtype (Emax = 59% relative to serotonin); LSD, by contrast, has much higher agonist efficacy at 5-HT2A (Emax = 92%). Unlike LSD, 2-Br-LSD lacks activity at the 5-HT2B receptor, an interaction linked to valvulopathy and pulmonary hypertension. While LSD (0.1 mg/kg IP) induced the HTR in mice, administration of 2-Br-LSD at IP doses ranging from 0.1–10 mg/kg failed to induce the behavior. Furthermore, pretreatment with 2-Br-LSD (0.1–3 mg/kg IP) blocked the HTR induced by the 5-HT2A agonist/psychedelic drug 2,5-dimethoxy-4-iodoamphetamine (DOI) ($F(4,26) = 17.96$, $p < 0.0001$), indicating 2-Br-LSD is brain penetrant and interacts with the 5-HT2A receptor after in vivo administration. We also found that 2-Br-LSD produces weak recruitment of β-arrestin via 5-HT2A in vitro and has reduced potential to induce tolerance in vivo in the HTR paradigm.

Conclusions: Overall, 2-Br-LSD has an improved pharmacological profile compared to LSD. 2-Br-LSD appears to act as a non-hallucinogenic 5-HT2A agonist, similar to the isolysergic acid derivative lisuride. Our studies provide insight into the non-hallucinogenic activity of 2-Br-LSD, which is probably a consequence of its weak partial agonist activity at the 5-HT2A receptor. Follow-up studies are necessary to assess whether 2-Br-LSD can mimic the therapeutic effects of LSD but with less potential to produce hallucinogenic side-effects.

Keywords: Psychedelics, Psychedelic medicine, LSD, 5-HT2A Receptor, Head Twitch Response

Disclosure: BetterLife Pharma Inc.: Contracted Research (Self)

P350. GluN2B-NMDARs on Gabaergic Interneurons Mediate the Antidepressant-like, but not Psychotomimetic, Effects of Ketamine in Mice

Santosh Pothula*, Alexa-Nicole Sliby, Min Wu, Rong-Jian Liu, Ralph DiLeone, Ronald Duman

Broad Institute of MIT and Harvard, Cambridge, Massachusetts, United States

Background: A single subanesthetic dose of ketamine exerts rapid antidepressant-like effects via rapid glutamate efflux, activation of mTORC1 signaling, and enhanced synaptic transmission in the medial prefrontal cortex (mPFC); however, the initial cellular trigger for these synaptic and behavioral actions of ketamine remains unclear.

Methods: Here, we used electrophysiology, biochemistry, molecular biology, genetic and behavioral approaches to determine the baseline sex differences in behavior after GluN2B conditional deletion from Sst- and Pvalb-interneurons, and effect of ketamine on chronic unpredictable stress (CUS)-induced behavioral deficits, activation of mTOR signaling cascade and changes in synaptic transmission in adult male and female GluN2Bfl/fl (WT) and Sst-creGluN2Bfl/- and Pvalb-creGluN2Bfl/- conditional knockout (cKO) mice. One-way or Two-way ANOVA post hoc Bonferroni or t-tests were used for data analysis and $p < 0.05$ was considered as statistically significant.

Results: Acute treatment of ketamine significantly decreased NMDA-induced burst firing of Sst- and Pvalb-interneurons ex vivo in mPFC ($p < 0.05$). Deletion of GluN2B from Sst- or Pvalb-interneurons in the cKO produced changes in synaptic transmission in mPFC. Consistent with the disinhibition hypothesis, ketamine significantly decreased inhibitory, but enhanced excitatory transmission of layer II/III and V pyramidal neurons of mPFC ($p < 0.05$) and reverses CUS-induced behavioral deficits only in WT, but not Sst-creGluN2Bfl/- and Pvalb-creGluN2Bfl/- cKO, mice. In contrast, ketamine treatment-induced psychotomimetic effects (increased locomotion and sensory motor gating deficits) in both WT and Sst-creGluN2Bfl/- or Pvalb-creGluN2Bfl/- cKO mice in locomotor and prepulse inhibition tests. Preliminary data demonstrate that deletion of GluN2B from Sst-interneurons blocks ketamine-induced activation of mTOR signaling cascade. Ongoing studies are evaluating the effect of ketamine on mTOR signaling in Pvalb-creGluN2Bfl/- cKO mice.

Conclusions: Our results demonstrate that Gabaergic interneurons are an initial cellular trigger for synaptic and antidepressant-like behavioral actions of ketamine. Consistent with the disinhibition hypothesis, ketamine actions are mediated by GluN2B-NMDARs on Gabaergic interneurons via disinhibition of pyramidal neurons, activation of mTOR signaling, and enhanced synaptic function in mPFC.

Keywords: (R,S)-ketamine, NMDAR, GABAergic Interneurons, Antidepressant, Psychotomimetic Effects

Disclosure: Nothing to disclose.

P351. Anti-Anhedonic Profile of ENX-104, a Novel and Highly Potent Dopamine D2/3 Receptor Antagonist

Krishna Vadodaria*, Brian D. Kangas, David S. Garvey, William Brubaker, Diego A. Pizzagalli, Vikram Sudarsan, Kimberly E. Vanover, Jordi Serrats

Engrail Therapeutics, San Diego, California, United States

Background: Anhedonia, defined as the loss of pleasure derived from previously rewarding activities, is a behavioral phenotype present in many neuropsychiatric conditions, including major depression. Alterations in dopaminergic (DA) corticostriatal circuitry and dysfunction of mesolimbic DA pathways underlie reward processing deficits. At low doses, amisulpride, a D2/3 and 5-HT7 receptor antagonist, shows antidepressant effects and enhances reward-related neural responses, whereas at high doses it has been used as an antipsychotic. We hypothesized that pro-hedonic effects of amisulpride are mediated by its dopaminergic effects, specifically via blockade of presynaptic inhibitory D2/3 receptors at low receptor occupancy (RO), which in turn boosts DA neurotransmission. Based on this evidence, we developed a novel, selective D2/3 receptor antagonist, ENX-104 and predicted that it would enhance reward responsiveness at low doses (~40% D2/3 RO) without impairing motor function. We characterized the in vitro pharmacological profile, the pharmacokinetic (PK) profile, the corresponding receptor occupancies, and the behavioral impact of this investigational new drug, ENX-104.

Methods: ENX-104 pharmacology was characterized using binding (radioligand competition) and functional (agonist or antagonist mode) assays in vitro with separate cell lines overexpressing recombinant human receptors: D2L, D3, D4.4, 5-HT1A, 5-HT1B, 5-HT2A, 5-HT7.

For PK characterization of ENX-104, rats ($n = 3$) were orally dosed (0.5 mg/kg [mpk] and 5 mpk) with plasma collection at eight timepoints post dose (5 min-24 h), with a group of rats ($n = 5$) for plasma and brain collection 1, 4, 8, 24 h post dose.

In the D2/3 RO study, rats ($n = 5$) were orally dosed ENX-104 at 2.5 mpk and plasma and brains were collected at 1, 2, 4, 8, and 24 h post dose. In an ex-vivo competitive tracer binding assay, striatal tissue was incubated with radiolabled [3H]raclopride. Binding of labeled tracer was inversely proportional to ENX-104 occupancy at D2/3 receptors. Comparisons to vehicle were by Dunnetts's test. PK and RO data were further utilized to build PK:pharmacodynamic (PD) models enabling the prediction of D2/3 RO at different doses and time points following ENX-104 treatment.

To evaluate reward responsiveness, rats ($n = 8$) were trained on a touchscreen-based probabilistic reward task (PRT) analog using parameters based on the human task. To back-translate this into a

preclinical model predictive of anti-anhedonic efficacy, we validated the PRT with amisulpride. Vehicle, amisulpride (1, 5, 50 mpk) or ENX-104 (0.5, 1, 2.5 mpk) were orally administered 4-5 h before the PRT session in separate studies with a Latin square design.

For motor function, rats (n = 8) were orally dosed ENX-104 (1, 2.5, 10 mpk) and catalepsy was scored by an observer at 1.5, 3 and 4 h post dose. Rat paws were gently placed on a rubber bung and a score of 1 was given for each paw that remained in position for at least 15 seconds, and latency for the time each paw remained on the bung was calculated. Comparisons to vehicle were done by exact Wilcoxon rank sum tests.

All animal study protocols were approved by IACUC and conduced in accordance with NIH guidelines.

Results: ENX-104 is a potent dopaminergic antagonist, with Ki of 0.01 nM for D2L, 0.2 nM for D3, and 1.6 nM for D4 receptor subtypes. Among serotonergic receptors ENX-104 displayed binding affinity of 3.7 nM at the 5-HT2A receptor, acting as a partial antagonist (EC50 of 14 nM, Emax of ~40%). In comparison, ENX-104 displayed little or no functional activity at the 5-HT1A and 5-HT7 receptors.

ENX-104 exhibited a short plasma half-life with significant brain enrichment and retention over the course of 24 h. The plasma half-life of ENX-104 (0.5mpk oral dose) is nearly 2 h with a tmax of ~30 mins. Although plasma concentrations reduced after 2 h, the maximum brain concentration is reached at ~4 h post dose. These data correspond with the D2/3 RO time course data, showing a maximum receptor occupancy at 4 h post dose. Based on these data, 4 h was chosen as a key time point for behavioral analyses.

In the PRT, doses of amisulpride and ENX-104 targeting low but not high D2/3 RO significantly increased reward responsiveness. This was revealed by substantive increases in response bias (log b) without reductions in task discriminability (log d). General linear model tests of within-subjects contrast (quadradic term) yielded statistical significance across the dose-response function for ENX-104 ($p = 0.048$) and a trend for amisulpride ($p = 0.065$). Given the a priori hypotheses, we conducted paired t-tests of log b values. Low amisulpride doses were significantly different than vehicle (1 mpk, $p = 0.03$; 5 mpk, $p = 0.04$), and ENX-104 low doses were significantly different than vehicle (0.5 mpk, $p = 0.006$ and 1 mpk, $p = 0.04$).

Notably, ENX-104 did not induce catalepsy at lower doses (1 and 2.5 mpk). Catalepsy was observed only in the 10 mg/kg group (D2/3 RO > 80%).

Together with the PK:PD model, these data suggest that D2/3 RO of ~40-60% is the target range for anti-anhedonic effects of ENX-104, with catalepsy emerging only at RO greater than ~80%.

Conclusions: These data predict ENX-104 could reduce anhedonia in humans at low doses without inducing extrapyramidal side effects. Our data support further investigation of ENX-104, a potent and novel D2/3 receptor antagonist, for clinical use in treating anhedonia in psychiatric disorders.

Keywords: Anhedonia, Antidepressant, D2 Dopamine Antagonists, Reward, Depression

Disclosure: Engrail Therapeutics: Employee (Self)

P352. A Single Administration of a Psychedelic Can Persistently Change Synaptic Function Through 5-HT2A Receptor Activation in Rats

Charles Nichols*, Meghan Hibicke, Jason Middleton, Hannah Kramer, Jesper Kristensen

LSU Health Sciences Center, New Orleans, Louisiana, United States

Background: Certain psychedelics such as psilocybin have shown profound and long-lasting therapeutic benefit in several neuropsychiatric disorders including depression and substance use disorder. Clinical trials have shown that a single treatment can have therapeutic benefits lasting months to years. The molecular mechanisms underlying these effects remain unknown but are thought to involve changes in synaptic plasticity. Previously, we demonstrated that a single administration of psilocybin or lysergic acid diethylamide (LSD) to Wistar Kyoto (WKY) rats had long-lasting antidepressant-like effects. In the forced swim test (FST), immobility was dramatically reduced at five weeks (35 days) post treatment with a large effect size. Further, we also have demonstrated that a single treatment with psilocybin rescued deficits in object pattern recognition and decreased immobility in the FST when measured at five weeks in female Sprague-Dawley rats subject to adolescent chronic restraint stress. In these systems, we only tested the effects of psilocybin out to five weeks. In order to assess the durability of effect of a single administration of psychedelic, and to investigate the role of 5-HT2A receptors, we assessed the effects of a single treatment with psilocybin and 25CN-NBOH (a selective 5-HT2A receptor agonist) in the FST at 5 weeks, and 12 weeks in WKY rats. Further, we assessed synaptic function at 12+ weeks in the vmPFC.

Methods: Male adult WKY rats were injected with either psilocybin or 25CN-NBOH (i.p., HCl salt, 1.5 mg/kg), or sterile saline. At five weeks, all rats were assessed in the FST. This involved placing rats into a plastic cylindrical tank (114 cm×30.5 cm) that contained 30 cm of water at 28-30°C for a five-minute swim, and video recording the sessions. Videos were scored for immobility, swimming, and climbing. 12 weeks after treatment, rats were again assessed in the FST to examine persistence of effect. After FST testing was complete, rats were transcardially perfused with a neuroprotective artificial cerebrospinal fluid (aCSF), and brains removed, sliced, and electrophysiological whole cell recordings were made from layer V pyramidal neurons of the vmPFC (~3-5 neurons per animal) to characterize synaptic and intrinsic properties of these neurons.

Results: Both psilocybin and 25CN-NBOH produced a significant decrease in immobility and increase in swimming behaviors to an equivalent extent when measured at five weeks. These changes were stable, and present at the same effect size when measured at 12 weeks. Slice recordings showed that drug treatment depolarized resting membrane potential and decreased action potential firing thresholds in regular spiking neurons and had bidirectional effects in different classes of bursting neurons: psilocybin increased sEPSC amplitude in transient bursting neurons and decreased sEPSC frequency in rhythmic oscillatory bursting neurons.

Conclusions: The effects of a single treatment with psilocybin or 25-CN-NBOH appear to be persistent, as no changes in effect sizes in the FST were measured between 5 weeks and 12 weeks. FST behaviors are mediated by vmPFC projections to the dorsal raphe nucleus. Our results indicate that both drugs have very long-lasting effects on synaptic function that facilitates the activity of neurons in the vmPFC. Because the dose of 25CN-NBOH used is predicted to be selective for 5-HT2A receptor activation, we conclude that 5-HT2A receptor activation is necessary and sufficient for the long-term effects of psychedelics.

Keywords: Serotonin 5-HT2A Receptor, Psychedelics, Psilocybin, Depression, Model Systems

Disclosure: Eleusis Therapeutics: Advisory Board (Self), Eleusis Therapeutics: Stock / Equity (Self), Eleusis Therapeutics: Royalties (Self), Palo Santo Ventures: Advisory Board (Self)

P353. The NMDA Receptor Subunit GluN2D is a Target for Rapid Antidepressant Action

Claus Normann*, Stefan Vestring, Katharina Domschke, Josef Bischofberger, Tsvetan Serchov

University of Freiburg, Freiburg, Germany

Background: Major depressive disorder is a widespread and devastating condition that leads to enormous individual suffering and imposes a serious socioeconomic burden. Despite its vast impact, current medical treatment options are limited, and conventional antidepressants take weeks or even months to alleviate patient symptoms. Subanesthetic doses of ketamine effectively reduce depressive symptoms within hours but are often accompanied by distressing adverse effects limiting its broad clinical use. The identification of ketamine's primary target and subsequent mechanisms further downstream relevant for its antidepressant efficacy could lead to more specific interventions with fewer side effects and better treatment outcomes.

Methods: Young adult mice of both sexes were used for all experiments. Hippocampal brain slices were obtained after decapitation and patch-clamp recordings were performed according to previously published protocols to measure long-term synaptic plasticity (LTP), NMDAR currents and other cell parameters. Slices from mice expressing tdTomato in SOM-INs (SOM-Cre (SST tm2.1(cre)Zjh/J) were used to assess SOM-INs. Protein- and RNA levels were assessed by Western Blot and RT-PCR. For behavioral assments, the chronic despair model of depression, the Nosepoke Sucrose Preference Test in the IntelliCage and an Open Field Test were used. GRIN2D siRNA was applied by intrathecal injection together with in-vivo-jet-PEI solution.

Results: We started by investigating the behavioral antidepressant properties of ketamine in an animal chronic despair model (CDM) of depression. In CDM, animals were subjected to ten-minute forced swim sessions on five consecutive days to induce a depression-like state, followed by a rest period of two days, and different readout assessments were performed thereafter. After a single intraperitoneal injection of ketamine (10 mg/kg body weight), immobility time was significantly reduced to baseline levels and sucrose preference was normalized. In ex-vivo hippocampal brain slices, LTP induction was completely abolished after swim stress and fully restored by ketamine treatment. In brain slices from naïve animals, ketamine facilitated LTP induction in a protocol designed to avoid ceiling effects. However, the underlying mechanism of this effect is not obvious, as hippocampal LTP is N-methyl-D-aspartate (NMDA)-dependent and ketamine is a noncompetitive NMDA receptor antagonist. The NMDAR is a heterotetramer composed of two GluN1 and two GluN2 subunits with widespread synaptic and extrasynaptic distribution on different neurons in the brain. A potential mediator of o positive modulation of LTP could be somatostatin-expressing interneurons (SOM-INs), which constitute a major subpopulation of GABAergic interneurons and provide feedback inhibition to distal dendrites of PCs in the hippocampus and the frontal cortex. As part of a feedback loop, these interneurons effectively control postsynaptic NMDA receptor activation and synaptic plasticity. In brain slices, NMDAR currents were inhibited to a larger degree in SOM-INs than in pyramidal cells. GluN2D subunits are preferentially expressed in SOM-INs, and ketamine has a higher affinity to NMDARs containing this subunit. Selective inhibition of GluN2D-containing NMDA receptors recovered stress-induced blockade of long-term synaptic potentiation, which is consolidated in later stages by structural synaptic modifications. The cellular and behavioral actions of ketamine could be fully mimicked in-vitro and in-vivo by the selective GluN2D antagonist NAB-14 and by small interfering RNA (siRNA)-mediated posttranscriptional silencing of its encoding gene GRIN2D in vivo.

Conclusions: These findings identify the GluN2D subunit of the NMDA receptor on interneurons as novel and highly specific targets for drug treatment of major depression, including RNA-based therapies.

Keywords: Depression, Synaptic Plasticity, NMDA Receptor, In-Vivo siRNA Treatment, Ketamine

Disclosure: Janssen Pharmaceuticals: Honoraria (Self), GluN2D Antagonists to Treat Depression: Patent (Self)

P354. Activation of the Rostral Agranular Insular Cortex is Involved in the Antidepressant Actions of Arketamine

Yukio Ago*, Rei Yokoyama, Atsushi Kasai, Masato Tanuma, Misuzu Hayashida, Yuto Shimazaki, Momoko Higuchi, Hisato Igarashi, Kaoru Seiriki, Shun Yamaguchi, Takanobu Nakazawa, Kenji Hashimoto, Hitoshi Hashimoto

Hiroshima University, Hiroshima, Japan

Background: Esketamine and arketamine are the S(+) and R(-) enantiomers of ketamine, respectively, and esketamine nasal spray has been developed and recently approved for treatment-resistant depression. Accumulating evidence from rodent studies suggests that arketamine may show a more potent antidepressant-like effect and affect fewer perceptual disturbances than esketamine, but the antidepressant mechanism of ketamine enantiomers is not fully understood. In the present study, we aimed to explore and identify brain regions that contribute to the difference in antidepressant action between ketamine enantiomers by brain-wide mapping of immediate early gene expression.

Methods: Experimental procedures involving animals and their care were conducted in compliance with the guidelines of the Guide for the Care and Use of Laboratory Animals, and all animal studies were approved by the Animal Care and Use Committees at Osaka University. We used post-weaning social isolation of mice as a model of depression. Male C57BL/6 J mice were weaned at postnatal day 21 and assigned to either group housing or isolation. At day 63, mice were subjected to a 6-min session of a forced swim test (FST). To visualize neuronal activation on a brain-wide scale with subcellular resolution, we used Arc-dVenus mice, which expresses a destabilized fluorescent reporter protein dVenus under the promoter of immediate-early gene Arc, and analyzed dVenus expression in the whole brains of mice in an unbiased manner by an automated imaging system FAST (block-face serial microscopy tomography), which we recently developed (Seiriki et al., Neuron 2017; Nat Protoc 2019).

Results: Social isolation stress increased the immobility time of mice in the FST, and arketamine showed a greater potency of antidepressant-like effect than esketamine in isolation-reared mice. Both arketamine and esketamine increased neuronal activity in cortical and subcortical regions in isolation-reared Arc-dVenus mice, and then the machine learning classifiers with brain-wide activation mapping identified several candidates of brain areas including the rostral agranular insular cortex that may contribute to the antidepressant-like effect of arketamine as well as discrimination between the effects of arketamine and esketamine. Additionally, chemogenetics-mediated acute inhibition of excitatory neuronal activity in the rostral agranular insular cortex blocked the antidepressant-like effects of arketamine, but not of esketamine, in isolation-reared mice. Conversely, chemogenetic activation of the rostral agranular insular cortex neurons induced antidepressant-like effects in isolation-reared mice.

Conclusions: These findings suggest that esketamine and arketamine have different neural mechanisms underlying their

antidepressant actions. This study also implies that activation of the rostral agranular insular cortex is essential for exerting the antidepressant-like effects by arketamine.

Keywords: Arketamine, Esketamine, Insular Cortex, Antidepressant Mechanisms, Neural Activity

Disclosure: Nothing to disclose.

P355. Ketamine Preservative Benzethonium Chloride Potentiates Hippocampal Synaptic Transmission and Inhibits Neurotransmitter Receptors and Transporters in Vitro

Kyle Brown*, Panos Zanos, Ruin Moaddel, Xi-Ping Huang, Chris Powels, Mike Michaelides, Edna Pereira, Todd Gould

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

Background: Benzethonium chloride (BZT) is an excipient antimicrobial used in numerous products including (R,S)-ketamine (ketamine) drug formulations. A 10 uM ketamine formulation, a concentration commonly used in preclinical studies, contains up to 12 nM BZT. Emerging evidence indicates BZT is functionally active. BZT may therefore contribute to some of the clinical or preclinical effects observed with ketamine.

Methods: We evaluated affinity and functional effects of BZT at neurotransmitter receptors and transporters. Radioligand binding assays determined the affinity of BZT for numerous targets whereas functional effects were established via assessment of neurotransmitter uptake, β-arrestin translocation, cAMP production, and/or Ca2+ mobilization. Plasma and brain distribution of BZT up to eight hours after systemic administration (i.e., intraperitoneal administration) was determined in male mice. The effect of BZT on in vitro synaptic transmission was assessed in the hippocampus of male mice via electrical stimulation of Shaffer collateral afferents and recording field excitatory postsynaptic potentials (fEPSPs) in the CA1 subfield.

Results: BZT potently binds to numerous receptors at high to moderate affinity (e.g., σ2 Ki=7 nM; n = 3-5 independent experiments per receptor), inhibited transporter activity (e.g., dopamine transporter Ki=545 nM; n = 3 independent experiments per transporter), and did not exhibit agonist activity at any assessed metabotropic receptor (n = ≥3 independent experiments per receptor). BZT was detected in the periphery, but not in the brain, of mice following systemic administration (n = 4 at 10, 30, 60, 120, 240, and 480 min post-BZT injection). Bath application of BZT potentiated hippocampal Schaffer collateral-CA1 fEPSPs in mouse hippocampal slices ($F(6,142)=6.60$, $p < 0.0001$; EC50 = 2.03 nM). Specifically, bath application of 10 nM BZT ($p < 0.0001$; n = 14), 50 nM BZT ($p = 0.028$; n = 17), and 300 nM BZT ($p = 0.021$; n = 18), but not VEH, (n = 48), 0.3 nM BZT ($p = 1.00$; n = 19), 2 nM BZT ($p = 0.72$; n = 19), or 10000 nM BZT ($p = 0.067$; n = 14), significantly increased the fEPSP slope value. Separate experiments showed that the potentiating effect of 10 nM BZT persisted following BZT washout ($t(5)=0.987$, $p = 0.37$).

Conclusions: BZT interacts with numerous membrane receptors and can induce synaptic potentiation measured in hippocampal slices. However, it does not readily cross the blood-brain barrier. Studies measuring peripheral endpoints following BZT-containing ketamine treatment or directly exposing systems to BZT-containing ketamine formulations (e.g., intracortical administration) should account for the effects of BZT. Our findings suggest that earlier data attributing physiological effects to ketamine may be impacted by BZT and that additional investigation into the functional impact of BZT is warranted.

Keywords: (R,S)-ketamine, Synaptic Plasticity, Hippocampus, Slice Electrophysiology, Neurotransmission

Disclosure: Nothing to disclose.

P356. Transcriptomic Analysis of Rats Exposed to Chronic Mild Stress and Modulation by Prolonged Treatment With the Antipsychotic Drug Lurasidone

Veronica Begni, Moira Marizzoni, Diana Morena Silipo, Kerstin Camile Creutzberg, Mariusz Papp, Annamaria Cattaneo, Marco Andrea Riva*

University of Milan, Milan, Italy

Background: Exposure of adult rats to chronic stress represents a valuable experimental paradigm to investigate and characterize the mechanisms that may contribute to the development of a pathologic phenotype. One consistent finding following stress exposure is the development of an anhedonic phenotype, which may be normalized through pharmacological intervention with different psychotropic drugs. Anhedonia is a key domain in mood disorder and originates from an array of alterations in different brain regions. In this respect, the prefrontal cortex may play a crucial role considering that it receives a number of functional inputs, and it has an important modulatory role on other structures, including the amygdala and nucleus accumbens.

Here, we performed a transcriptomic analysis on the prefrontal cortex of rats exposed to the chronic mild stress (CMS) paradigm and we investigated the potential impact of chronic treatment with the antipsychotic drug lurasidone on such transcriptional alterations in order to identify genes that may contribute to stress susceptibility as well as those that could play a role in the therapeutic intervention with lurasidone.

Methods: Adult male rats were initially exposed to CMS for 2-weeks and sucrose intake was measured as a proxy for anhedonia. Animals that showed reduced intake (vulnerable) were randomized to receive vehicle or the antipsychotic drug lurasidone (3.0 mg/kg, per os) for 5 more weeks while continuing CMS exposure. Blood and brain tissues were collected 24 h after the last manipulation for the molecular analyses.

Whole-genome RNA sequencing analysis was conducted using the Illumina NextSeq500 platform in the Prefrontal cortex (PFC) of Control rats treated with Vehicle (CT/Veh), CMS rats treated with Vehicle (CMS/Veh), and CMS rats treated with Lurasidone (CMS/Lur). Differentially expressed genes (DEGs) identified by the Bioconductor package Deseq2 with FDR-corrected p-values ($q < 0.05$) were used to run pathway analyses and functional network prediction through Ingenuity Pathway Analysis (IPA).

Results: In line with our previous studies, CMS exposure caused a significant reduction in sucrose intake, which was progressively normalized by lurasidone treatment ($p < 0.001$).

Transcriptomic analysis in the prefrontal cortex allowed us to identify a high number of genes (>500) that were differentially expressed in CMS/Veh, as compared to CT/Veh, but that were also differentially expressed in CMS/Lur when compared to their vehicle-treated counterpart (CMS/Veh).

Using Venn diagram analysis, we found that 107 genes in common were altered in CMS/Veh and CMS/Lur groups. By running pathway analysis, we found that most of these pathways show opposite modulation. Indeed, among the most significant changes, we found that netrin, CREB, and oxytocin signaling pathways were reduced in CMS/Veh and up regulated in CMS/Lur. Similarly, based on disease and function analysis, we observed an increase in cell and neuronal death in CMS/Veh, with opposite changes in rats treated with lurasidone.

Conclusions: In summary, we provide new insights on how chronic lurasidone treatment may counteract the alterations produced upon prolonged exposure to stress, a key vulnerability factor for mental disorders. In particular, the prefrontal cortex appears to be particularly sensitive to stress exposure with a down-regulation of different pathways associated with neuronal

guidance and synaptic plasticity. Our results suggest that pharmacological intervention with lurasidone can counteract such changes and may ultimately promote resilience.

Keywords: Chronic Stress, Transcriptomics, Lurasidone, Plasticity

Disclosure: Angelini: Speakers Bureau (Self), Lundbeck: Speakers Bureau (Self), Otzuka: Speakers Bureau (Self), Sumitomo Pharma: Honoraria (Self), Sumitomo Pharma: Grant (Self), Sunonion: Grant (Self)

P357. The Formation of Stress Vulnerability is Controlled by 5-HTergic System in the Raphe Nucleus Through Striatal N-Acetyltransferase, Shati/Nat8l, in Mice

Hajime Miyanishi*, Shin-ichi Muramatsu, Atsumi Nitta

Graduate School of Medicine and Pharmaceutical Sciences, University of Toyama, Toyama-shi, Japan

Background: Depression is one of the most common mental diseases, with increasing numbers of patients in all over the world. Stress is closely related to the depression pathology. While some individuals show the vulnerable to stress, however, others are resilient. Revealing the underlying regulatory mechanisms of stress sensitivity could offer novel insights for understanding the pathogenesis of depression. Serotonin [5-hydroxytryptamine (5-HT)] is involved in depression pathogenesis, and 5-HT has been targeted for medical treatments of depression. The hereditary reduction of 5-HT in the brain induced the vulnerability to social stress in mice. Shati/Nat8l was identified from the brain of methamphetamine-induced psychosis model mice. Shati/Nat8l has N-acetyltransferase activity, and predominantly synthesizes N-acetyl-aspartate (NAA) from L-aspartate and acetyl-coenzyme. NAA expression in the brain was reported to be changed in the patients with depression. We previously found that Shati/Nat8l mRNA levels in the dorsal striatum were increased in the mice exposed repeated forced swimming stress. In the present study, we demonstrated the role of striatal Shati/Nat8l in the depression pathology and stress sensitivity.

Methods: Male C57BL/6 J mice were exposed repeated social defeat stress (RSDS) using male ICR mice, and the stress susceptible or resilient group were classified by social interaction test. We generated dorsal striatal Shati/Nat8l overexpression (dSTR-Shati OE) mice using adeno associated virus vectors. Depression-like behaviors were assessed using these mice after RSDS or sub-threshold social stress (micro social defeat stress; MSDS). 5-HT content in the dorsal striatum were measured using in vivo microdialysis. The effects of 5-HT in the dorsal striatum to stress sensitivity were investigated by microinfusion of selective serotonin reuptake inhibitors, fluvoxamine (FLX), into the dorsal striatum. We also investigated the contribution of 5-HTergic neuron projected from raphe nucleus to dorsal striatum to stress sensitivity using DREADD system.

Results: We found that Shati/Nat8l mRNA levels were increased in the dorsal striatum of stress susceptible, but not resilient, mice exposed RSDS. We also found that only stress susceptible mice showed the downregulation of 5-HT content in the dorsal striatum. The proportion of stress resilient group after RSDS decreased to 50% in dSTR-Shati OE mice compared to control mice, and these mice showed the vulnerability to even MSDS. The reduction of 5-HT content in the dorsal striatum was observed in dSTR-Shati OE mice. The vulnerability to stress in dSTR-Shati OE mice was recovered by microinfusion of FLX into the dorsal stratum or activation of 5-HTergic neuron projected from raphe nucleus to dorsal striatum by DREADD system.

Conclusions: In the present study, we have demonstrated that striatal Shati/Nat8l-induced stress vulnerability is mediated by the reduction of striatal 5-HT content through 5-HTergic system in the raphe nucleus. Our study suggests that dorsal striatum-raphe nucleus circuit is involved with the stress sensitivity in the depression pathogenesis, and that striatal Shati/Nat8l could be a novel therapeutic target for depression.

Keywords: Depression, Social Defeat Stress, Dorsal Striatum, Dorsal Raphe, Serotonin

Disclosure: Nothing to disclose.

P358. A Non-Hallucinogenic LSD Analog With Therapeutic Potential for Mood Disorders

Argel Aguilar Valles*, Vern Lewis, Andre Telfer, Emily Arsenault, Fatimeh-Frouh Taghavi-Abkuh, Fatema El Sayegh, Alfonso Abizaid

Carleton University, Ottawa, Canada

Background: Major depressive disorder (MDD) is the leading cause of disability worldwide. Current pharmacotherapy treatments, such as SSRIs and SNRIs, have long effective latencies, require chronic administration, and show an estimated 30-40% treatment resistance rate, necessitating the search for more effective, alternate treatments. Interest in psychedelic hallucinogens (e.g., lysergic acid diethylamide [LSD], psilocybin) has seen a resurgence due to their potential for treating neuropsychiatric diseases, including anxiety and depressive disorders. Unlike first line MDD treatments, hallucinogens target serotonin receptors directly and show rapid and persistent antidepressant effects after only one or two treatments, potentially due to activation of synaptic and structural plasticity mechanisms. However, psychedelics induce hallucinations that can last for hours, making efficient and cost-effective treatment difficult. Therefore, discovering non-hallucinogenic derivatives with antidepressant properties is of paramount importance. 2-Bromo-LSD (2-Br-LSD, BETR-001) is an LSD derivative with 5-HT2A partial agonist activity that lacks hallucinogenic effects and has been safely used for cluster headache treatment. Thus, 2-Br-LSD may represent a possible novel therapeutic for treating neuropsychiatric diseases whose pathologies involve dysfunction in cortical serotonergic pathways.

Methods: We investigated the effects of 2-Br-LSD on dendritogenesis in primary rat cortical neuron cell cultures. Cortices from embryonic day 18 rat embryos were enzymatically and mechanically disaggregated; cells were then plated at 50 000 cells/well and treated with different concentrations of 2-Br-LSD (1, 10 and 100 nM, 1 and 10 uM), 10 uM ketamine, or vehicle for 3 hours on days in vitro 3 (DIV3). On DIV6, cells were either processed for immunofluorescence to detect MAP2 or for cell survival using the Neurite Outgrowth Staining Kit (Thermo Fisher). Sholl analysis was used to quantify changes in arbor complexity in imaged cells. To assess spinogenesis, neurons were kept in culture until DIV18, treated as above and stained for MAP2 and F-actin. To assess the contribution of the serotonin 5-HT2A receptor on 2-BR-LSD dendritogenesis, DIV3 neurons were pretreated with the 5-HT2A selective antagonist volinanserin (VOL) at 100 nM, 500 nM, or 1 uM for 1 hour, followed by vehicle or 2-Br-LSD (1 uM). Neurons from 6 different wells per treatment and from at least 2 independent experiments were used for analyses.

To determine the effect of 2-Br-LSD on stress-coping behaviour, adult male and female mice ($n = 12$ group/sex) were treated with vehicle or 2-Br-LSD at 0.3, 1 or 3 mg/kg (IP). 24 hours after treatment, mice were tested in the open field (OF, 10 min) and 1 hour later in the forced-swim test (FST, 6 min). Immediately after, brains were harvested and processed for Golgi staining (FD Rapid GolgiStain Kit, FD NeuroTechnologies). A separate cohort of mice ($n = 11$/group) was treated with VOL (IP, 0.125 mg/kg) or vehicle 15 minutes before 2-Br-LSD (IP, 1 mg/kg), then tested in the OF

and FST the following day. Finally, a group of female mice were subjected to chronic variable stress (CVS; 2 stressors per day in variable order) for 5 weeks (n = 12/group). Following the last stress day, mice were treated with vehicle or 2-Br-LSD (IP) once at 3 mg/kg or 3 times, every 48 h, at a 1 mg/kg dose. Mice were tested in the open field and splash test at two time points post-treatment (3-4 h and 4 weeks).

Results: 2-Br-LSD induced a dose-dependent increase in dendritic arbour complexity, as reflected by the increases in the number of process crossings in the Sholl analysis and the increased length of the dendritic arbour. The maximal effect was achieved with the 1 uM dose comparable to the 10 uM ketamine treatment. 2-Br-LSD also increased spine density in DIV18 neurons, but only at the 10 uM dose. Neither of the treatments decreased cell viability in cultured neurons.

In vivo, 2-Br-LSD decreased immobility in the FST at the 1 mg/kg dose, in both male and female mice, without affecting general locomotion in the OF. Furthermore, spine density in the prefrontal cortex was increased by this treatment. 2-Br-LSD treatment also reversed the effects of CVS in female mice, increasing exploration of the centre of the OF and increasing self-grooming in the splash test.

Promotion of dendritogenesis in vitro and active stress-coping behaviour by 2-Br-LSD depended on 5-HT2A receptors, as VOL blocked both effects.

Conclusions: Our findings demonstrate that 2-Br-LSD induces structural plasticity in cortical neurons in vitro and in vivo, promotes active stress-coping behaviours and reverses the behavioural effects of CVS. These results show that 2-Br-LSD may possess a therapeutic potential and represents a promising alternative to psychedelics in treating MDD.

Keywords: LSD, Dendritic Arborization, Spine Morphogenesis, Stress Coping, 5-HT2A Receptor

Disclosure: BetterLife Pharma: Contracted Research (Self)

P359. Cariprazine, a Dopamine D3 Receptor-Preferring Partial Agonist, Reduces Footshock-Induced Ultrasonic Vocalizations in a Rat Model of Anxiety: Synergy With Serotonergic and GABAergic Mechanisms

Nasser Haddjeri, Bence Farkas, Béla Kiss, Viktor Román, Balázs Lendavi, Nika Adham*

Abbvie, Madison, New Jersey, United States

Background: Symptoms of anxiety are frequent in patients with schizophrenia, bipolar I disorder, and major depressive disorder, highlighting the need for treatments that can address the complex symptomatology characteristic of these disorders. Cariprazine is a dopamine D3 receptor-preferring D3/D2 receptor partial agonist approved by the US Food and Drug Administration to treat adults with schizophrenia as well as manic, mixed, and depressive episodes associated with bipolar I disorder. In addition to dopaminergic actions, cariprazine also acts as a partial agonist at serotonergic 5-HT1A receptors, which are implicated in the development of anxiety symptoms. The objective of this study was to assess the anxiolytic-like effects of cariprazine in rats, tested alone and in combination with two clinically used anxiolytics: the selective serotonin reuptake inhibitor (SSRI) escitalopram and the benzodiazepine diazepam.

Methods: Anxiety-like symptoms were tested using an electric footshock-induced ultrasonic vocalization (USV) model in male Wistar Unilever rats (150–200 g). The testing apparatus was a 20 cm×20 cm x 25 cm chamber equipped with a grid floor, through which footshocks (1 mA; 4 s) could be delivered. Day 1 consisted of two 10-min sessions, separated by 60 min, during which rats received 1–4 randomly distributed footshocks until USV emission was detected. The following day, rats were placed in the chamber and received a single shock 30 min after injection of vehicle (saline with 10% 2-hydroxypropopyl-β-cyclodextrine; i.p.). On day 3, the test was repeated 30 min after i.p. injection of cariprazine (1, 5, 50, 100, or 300 µg/kg), escitalopram (0.2, 1, or 3 mg/kg), or diazepam (1, 3, or 6 mg/kg). In a second experiment, sub-effective doses of the drugs (determined in the first experiment) were administered alone or in combination 30 min before testing on day 3. USVs were defined as vocalizations with frequencies higher than 20 kHz. For both experiments, duration of USVs on each testing day was determined using the 5-min period beginning immediately after the first footshock-induced USV. For analysis, USV durations were converted to a percentage of USV duration after vehicle treatment on day 2.

Results: Cariprazine treatment dose-dependently decreased the duration of footshock-induced USVs. Compared with vehicle, significantly lower USV durations were observed at doses ≥50 µg/kg ($P < 0.05$) and USVs were eliminated by the 300 µg/kg dose ($P < 0.001$). Dose-dependent decreases were also observed with escitalopram and diazepam and were statistically significant versus vehicle at doses ≥1 mg/kg ($P < 0.001$) and ≥3 mg/kg ($P < 0.05$), respectively. At the highest doses tested, maximum decreases in USV duration relative to vehicle were 89% with escitalopram and 55% with diazepam. Higher doses of diazepam produced confounding sedative effects, making it difficult to determine specific anxiolytic effects. In the second experiment, combination treatment using sub-effective doses of cariprazine (1 µg/kg) and escitalopram (0.2 mg/kg) reduced footshock-induced USV duration by 42% compared with vehicle ($P < 0.001$), whereas combination treatment using sub-effective doses of cariprazine (5 µg/kg) and diazepam (1 mg/kg) resulted in a 26% reduction ($P < 0.01$ versus vehicle).

Conclusions: In a rodent footshock-induced USV model, cariprazine demonstrated potent anxiolytic-like activity, similar to the SSRI escitalopram and benzodiazepine diazepam. Synergistic anxiolytic-like effects were observed for cariprazine applied in combination with escitalopram or diazepam at sub-effective doses of each compound. Although further research is needed to understand the mechanisms underlying these synergistic effects, therapies including cariprazine, either alone or in combination with lower-dose anxiolytic agents, may be effective in the treatment of recalcitrant anxiety disorders or anxiety associated with schizophrenia.

Keywords: Cariprazine, Dopamine, 5-HT, Anxiety, Ultrasonic Vocalization (USV) Model

Disclosure: AbbVie: Employee (Self)

P360. A Potent 5-HT2A Receptor Agonist TCB-2 Exerts Rapid Antidepressant-Like and Anxiolytic-Like Effects in Mice

Hiroyuki Koike*, Ichiro Horinokita, Mikio Suzuki, Takashi Futamura

Otsuka Pharmaceutical Co., Ltd., Tokushima, Japan

Background: Classic serotonergic psychedelics have gained renewed interest both in clinical and preclinical fields because of robust antidepressant and anxiolytic responses after a single dose. As a result, understanding the mechanisms of action has been an area of great interest in the basic science. Recent studies emphasize the importance of 5-HT2A receptor agonism in the therapeutic effects of psychedelics, yet the exact mechanism remains unclear. Relevant and useful animal models are necessary to obtain a better understanding of behavioral outcomes and biological correlates associated with the antidepressant and anxiolytic effects of psychedelics, especially through activation of 5-HT2A receptor. In this study, we investigated behavioral

effects of a potent 5-HT2A receptor agonist TCB-2 in corticosterone-induced anhedonia and novelty suppressed feeding test (NSFT) to evaluate the potential utility of these models for preclinical research on psychedelics.

Methods: Male C57BL/6 J mice were used for this study. We treated mice with corticosterone to induce anhedonia, observed as reduced sucrose preference. NSFT was carried out in naive mice to measure anxiety-induced hypophagia. To evaluate antidepressant-like and anxiolytic-like effects of TCB-2 in these assays, we injected mice intraperitoneally with it 24 hours before the test. To evaluate hallucinogenic potency, we measured head-twitch response immediately after administration. Experiments employed 6-8 mice per treatment. The data were analyzed by one-way ANOVA followed by Dunnett's post hoc test ($p < 0.05$ was considered statistically significant).

Results: TCB-2 improved the chronic corticosterone-induced reduction in sucrose preference; the response was maximal at an intermediate dose (0.1 mg/kg) but slightly declined at higher doses. TCB-2 also shortened the latency time until feed in the NSFT. The effective doses of TCB-2 in these behavioral assays elicited the head-twitch response.

Conclusions: We demonstrated that TCB-2 exerted the antidepressant-like and anxiolytic-like effects after a single injection in a behavioral model of anhedonia, sucrose preference test in corticosterone-treated mice, as well as a model of anxiety, NSFT. Given that traditional antidepressants are effective in these models after chronic treatment, our findings indicate that the effects of TCB-2 is rapid onset. Furthermore, because the effects of TCB-2 were observed 24 hours after treatment and psychedelics are reported to promote long-lasting neural plasticity through 5-HT2A receptor activation, it is likely that TCB-2 at the effective doses induce neural plasticity which potentially mediates it's antidepressant-like and anxiolytic-like effects via the same mechanism as psychedelics. Future work that leads to a precise delineation of the mechanisms underlying the observed effects of TCB-2 is needed to identify and develop effective treatment for alleviating symptoms of depression and anxiety, and the behavioral models we employed may be useful to detect the robust effects of psychedelics and elucidate the mechanisms of their action.

Keywords: Serotonin 5-HT2A Receptor, TCB-2, Psychedelics, Animal Models

Disclosure: Nothing to disclose.

P361. In Vitro and In Vivo Profile of CYB003: A Novel, Deuterated Psilocybin Analog for the Potential Treatment of Major Depressive Disorder

Michael Palfreyman*, Joan Krakowsky, Michael Morgan, Clinton Canal, Pradip Pathare, Ken Avery, Amy Reichelt, Joshua Hartsel, Alex Nivorozhkin, Amir Inamder, Geoffrey Varty

Palfreyman BioPharm Advisors, Inc., St. Petersburg, Florida, United States

Background: Phase 2 clinical studies have demonstrated that short-term administration of psilocybin has beneficial effects on Major Depressive Disorder (MDD) and Treatment Resistant Depression (TRD). While psilocybin is efficacious and inherently safe, there is significant variability between patients in their psychedelic and side-effect experiences. This variability is likely due to psilocybin acting as a pro-drug that requires de-phosphorylation to the psychoactive metabolite, psilocin. CYB003 is a novel, deuterated analog of psilocybin that may offer benefits over psilocybin. The aim of these studies was to compare the in vitro and in vivo activity of CYB003 to psilocin and conduct Investigational New Drug (IND)-enabling studies to allow the initiation of clinical studies.

Methods: Pharmacological profiles of CYB003 and psilocin were compared using serotonin (5-HT) receptor binding and functional assays to evaluate potency, efficacy, and selectivity at serotonin receptors; both compounds were also screened for activity at a panel of over 100 proteins. Additionally, both compounds were evaluated in the mouse head twitch response (HTR) assay (a behavioral model of psychedelic-like effects). ADME and pharmacokinetic (PK) studies were also completed followed by dose-range finding studies in rodents and non-rodents species. Finally, IND-enabling studies conducted to Good Laboratory Practice (GLP) standards were completed; specifically, repeat dose toxicological studies, and cardiovascular, respiratory, and CNS safety pharmacology assessments.

Results: The pharmacological and selectivity profile of CYB003 was similar to that seen with psilocin. CYB003 binds and activates the 5-HT2A receptor (5-HT2A Ki: 37 nm; psilocin 31 nM) and induces HTR in mice (No.of HTR (0-30 min): CYB003 8.9 _+ 1.3; psilocin 12.6 + _1). ADME studies indicate little metabolism in vitro and PK/PD studies showed a good correlation between plasma levels and in vivo activity. Safety pharmacology study findings were consistent with activation of 5-HT receptors, reflecting findings seen with other psychedelic compounds, including psilocybin. GLP-toxicological studies established safe, and potentially effective doses, and will support multiple-dose clinical studies. Collectively, these data permitted the initiation of Phase 1/2 A clinical studies www.clinicaltrials.gov ID: NCT05385783: A study of a Psilocybin Analog (CYB003) in Participants with Major Depressive Disorder. The Phase 1/2 A protocol in participants with MDD is now ongoing (Clinilabs, Eatontown, NJ).

Conclusions: Our findings confirm that CYB003 has the appropriate pharmacology and safety profile to proceed to clinical trials in patients with MDD. These data further support the potential of CYB003 to have superior properties to psilocybin and to offer considerable clinical benefits.

Keywords: Psychedelics, Psilocybin Analog, Pharmacology, IND-Enabling GLP Studies, Major Depressive Disorder

Disclosure: Cybin, Inc.: Employee (Self), Palfreyman BioPharm Advisors, LLC: Owner (Self)

P362. Pharmacokinetic Profile of CYB003: A Novel, Deuterated Psilocybin Analog for the Potential Treatment of Major Depressive Disorder

Amir Inamdar*, Michael Morgan, Joan Krakowsky, Amy Reichelt, Clinton Canal, Tina Mueller, Ken Avery, Pradip Pathare, Joshua Hartsel, Alex Nivorozhkin, Geoffrey Varty, Michael Palfreyman

Cybin, London, United Kingdom

Background: Phase 2 clinical studies have demonstrated that short-term administration of psilocybin has beneficial effects on a range of psychiatric conditions such as Major Depressive Disorder (MDD), anxiety, and substance use disorders. While psilocybin is efficacious and inherently safe, literature shows there is significant variability between patients in their psychedelic and side effect experiences. This variability is likely due to psilocybin acting as a pro-drug that requires de-phosphorylation to the psychoactive metabolite, psilocin. Once metabolized, psilocin is absorbed into the bloodstream and crosses the blood-brain barrier to interact with central serotonergic receptors, including 5-HT2A receptors. Improving the absorption, distribution, and elimination of psilocybin, while retaining its therapeutic effects, may offer significant benefits to the patient. CYB003 is a deuterated analog

of psilocybin that may offer such benefits. The aim of these studies was to compare the pharmacokinetic (PK) profile of CYB003, a novel psilocybin analog, to psilocybin and psilocin.

Methods: To compare the PK profile, CYB003, psilocin, and psilocybin were administered orally to male Sprague-Dawley rats. Plasma and brain samples were collected over a 4-hour post-dosing period and were analyzed by LC-MS/MS for levels of CYB003 and psilocin. The PK profiles of CYB003 following both oral and intravenous administration to the mouse, rat, and dog were also determined. PK parameters were determined using non-compartmental analysis.

Results: CYB003 demonstrates potential advantages over psilocybin in terms of its plasma PK profile. Specifically, compared to psilocybin, the rat plasma exposure profile of CYB003 has a shorter time to reach peak effect, has reduced half-life (T1/2), and importantly, results in reduced variability. The time to reach peak plasma exposure (Tmax) was 30 min for CYB003, compared to 60 min for plasma psilocin following psilocybin administration. The half-life (T1/2) for CYB003 was 45 mins, compared to greater than 240 mins for psilocin from psilocybin, and was corroborated by a reduction in CYB003 MRT of 36%. Furthermore, the inter-subject variability of CYB003 at the maximum achieved plasma concentration (Cmax) was reduced by 53% compared to psilocin from psilocybin. The rank order for oral bioavailability (%F) across species was mouse=dog»rat. The CYB003 brain to plasma ratio in the rat after IV administration was 11.5 compared to 8.1 for psilocin from psilocybin, suggesting improved penetration into the CNS, and therefore, the potential to reduce the systemic dose of CYB003 needed to achieve a target therapeutic level in the brain. This trend was also observed in mice.

Conclusions: These findings suggest that the unique PK properties of CYB003 provide a meaningful advantage over psilocybin for the treatment of an array of psychiatric disorders, including MDD.

Keywords: Psychedelics, Pharmacokinetics, Major Depressive Disorder, Psilocybin Analog, Psilocin

Disclosure: Cybin: Employee (Self), Astrazeneca: Employee (Self)

P363. CVL-354, a Novel, Brain Penetrant and Selective Kappa Opioid Receptor Antagonist

Georgette Suidan, Galen Missig*, Sokhom Pin, Srinivas Chakilam, Sridhar Duvvuri, Philip Iredale

Cerevel Therapeutics, Cambridge, Massachusetts, United States

Background: Kappa opioid receptors (KOR) have strong activity throughout many key regions of the brain whose activity defines psychological phenomena, including motivation incentivized by reward or addictive substances, as well as anxiety. As a regulator of both cellular excitability and synaptic transmission of key circuits involved in reward and mood, KORs present a unique therapeutic target for clinical investigation for the treatment of major depressive disorder and substance abuse disorder.

Methods: In vitro pharmacology studies were conducted to evaluate binding affinity and functional antagonism (cAMP) of CVL-354 at human KOR and human mu opioid receptor (MOR). Pharmacokinetic studies were run in both mouse and nonhuman primate to determine brain penetrance. In mouse, target engagement studies were conducted to investigate the relationship between drug exposure and occupancy of CVL-354 at both mKOR and mMOR to determine in vivo selectivity. Opioid-induced thermal sensitivity measured by the tail flick assay was utilized to determine in vivo pharmacological selectivity for mKOR over mMOR. Lastly, CVL-354 reversal of KOR-agonist induced deficits in motivation were measured in the progressive ratio responding task.

Results: In vitro binding data demonstrated that CVL-354 has 31-fold binding affinity for hKOR over hMOR indicating selectivity for KORs. Furthermore, CVL-354 was determined to be an antagonist at both KOR (IC50 = 0.042 nM) and MOR (IC50 = 9.1 nM) and, importantly, did not demonstrate agonist activity at either receptor up to 1uM (0.01 nM-1 μM tested). Pharmacokinetic studies in mouse, rat and nonhuman primate demonstrated that CVL-354 is brain penetrant and suggested greater brain penetration in higher species. In vivo target engagement studies in mouse revealed 27-fold selectivity for KOR (IC50 = 2.2 nM) over MOR (IC50 = 59.7 nM), similar to human in vitro binding studies. The thermal sensitivity (tail flick) assay was utilized to demonstrate functional antagonism at both KOR and MOR of analgesia ($p < 0.05$) and, although qualitative, suggested ~10- fold pharmacodynamic selectivity. Lastly, the effects of CVL-354 on progressive ratio responding were evaluated. Administration of the KOR agonist, spiradoline, in mice induces an anhedonic-like phenotype in which the animals are less motivated to seek rewards in the progressive ratio paradigm ($p < 0.05$). CVL-354 dose dependently reversed the effects of spiradoline on motivation (ED50 = 0.09 mg/kg, ~2 nM). One-way ANOVA (tail flick and progressive ratio responding) were used and Dunnett's posthoc tests comparing treatment to control groups were done when appropriate.

Conclusions: In vitro and in vivo pharmacology assays demonstrated that CVL-354 is a novel, brain-penetrant, potent and selective kappa opioid receptor (KOR) antagonist. Additionally, CVL-354 reverses KOR agonist induced deficits in motivation.

Keywords: Kappa Opioid Receptor, Kappa Opioid Receptor Antagonist, Motivation, Anhedonia

Disclosure: Cerevel Therapeutics: Employee, Stock / Equity (Self)

P364. Fibroblast Growth Factor 21 (FGF21) is Increased in Patients With Bipolar Depression

Krista Wartchow*, Giselli Scaini, Tina Li, Giovana Zunta-Soares, Rodrigo Machado-Vieira, Jair Soares, João Quevedo

UTHealth Science Center, Houston, Texas, United States

Background: One leading hypothesis is that Bipolar Disorder pathology is in part due to failure of mitochondrial function to support adequate neurotransmission and synaptic plasticity, thus affecting mood regulation, memory, and executive function. Several studies support this hypothesis, showing BD patients present atypical mitochondrial metabolism, oxidative stress, and mitochondrial DNA (mtDNA) damage. Moreover, studies in primary mitochondrial diseases have shown a high prevalence of self-reported affective syndromes. Plasma levels of Fibroblast-growth-factor 21 (FGF-21) is an established biomarker for mitochondrial disorders.

Methods: In this pilot study, we included 31 healthy controls and 34 patients with BD (11 BD euthymic and 13 BD depressed). All subjects underwent a comprehensive clinical interview and diagnosis of BD and TRD according to the DSM-IV-TR. Mood symptoms and functional status were assessed with the Montgomery Asberg Depression Scale (MADRS), Young Mania Rating Scale (YMRS), Global Assessment of Functioning (GAF) and Functioning Assessment Short Test (FAST). Quantitative analysis of FGF-21 plasma levels was performed using commercial kit.

Results: One-Way ANCOVA after controlling for age, gender, BMI, ethnicity, and smoking status showed that FGF-21 plasma yielded no statistically significant between BD patients and HCs. However, after stratifying patients between depressed and euthymic, we found that patients with depressed BD present higher levels of FGF-21 compared to euthymic BD patients.

Another notable finding was that FGF-21 plasma levels predicted changes in the depression-rating scale and functional status.

Conclusions: In summary, our results suggest that increased plasma FGF-21 may act as a mood state marker in BD. Also, our findings corroborate previous studies and support the notion that mitochondrial dysfunction plays a role in the pathophysiology of BD and may play a role in clinical and functional outcomes.

Keywords: Mitochondria, Bipolar Disorder, FGF-21, Bipolar Depression

Disclosure: Nothing to disclose.

P365. Epigenetic GrimAge Acceleration and History of Suicide Attempt in Bipolar Disorder

Camila De Carvalho-Lima*, Emese H. C. Kovács, Alexandra Del Favero-Campbell, Alexandre Paim Diaz, Salahudeen Mirza, Consuelo Walss-Bass, Benney M. R. Argue, Joao Quevedo, Jair C. Sores, Marie E. Gaine, Gabriel R. Fries

UTHealth Houston, Houston, Texas, United States

Background: Bipolar disorder (BD) is associated with reduced life expectancy, with excess mortality in nearly all categories of natural causes. Suicide is a major cause of mortality in BD, and a previous history of suicide attempt has been associated with decreased lifespan and worse clinical outcomes. Although the risk of suicide in BD is high, excess mortality is mainly explained by somatic comorbidities, with recent studies suggesting that premature cellular senescence contributes to the shortened life expectancy seen in psychiatric disorders. Therefore, this premature mortality may be explained by observations that BD is associated with changes in age-related biomarkers of inflammation, oxidative stress, epigenetics, and metabolism. In this study, we investigated the acceleration of GrimAge, a novel epigenetic clock trained on time-to-death data and uniquely associated with mortality and lifespan, as well as its subcomponents, in patients with BD with and without a lifetime history of suicide attempt.

Methods: Study participants from a discovery cohort (Houston) included BD patients with no history of suicide attempt (BD/non-SA, $n = 66$), BD patients with a lifetime history of suicide attempt (BD/SA, $n = 77$), and healthy controls (HC, $n = 51$) matched for age, sex, and race. An index of GrimAge acceleration (AgeAccelGrim) was computed based on peripheral blood genome-wide DNA methylation (DNAm) levels measured by the Infinium MethylationEPIC Beadchip (Illumina) and chronological age for all participants. ANCOVA models were used to compare groups for AgeAccelGrim as well as DNAm-based smoking pack-years and seven age-related plasma proteins (adrenomedullin, beta-2-microglobulin, cystatin C, growth differentiation factor 15 (GDF-15), leptin, plasminogen activation inhibitor 1 (PAI-1), and tissue inhibitor metalloproteinases 1). Results from the patient-specific comparisons were independently validated in a replication cohort (Iowa) including BD/non-SA ($n = 47$) and BD/SA ($n = 47$).

Results: In the discovery cohort, HC, BD/non-SA, and BD/SA significantly differed for AgeAccelGrim after controlling for age, sex, population genetic stratification, years of education, body mass index, smoking status, and blood cell counts ($F(2,175) = 7.864$, $p < 0.001$), with the highest AgeAccelGrim found in BD/SA, with an excess of mortality of 3 years compared to HC ($p = 0.001$). BD/SA also showed a significantly higher AgeAccelGrim than BD/non-SA in the unadjusted model ($P = 0.002$) and after adjusting for covariates in the discovery cohort ($p = 0.027$). Furthermore, this between-group difference was also replicated in an independent cohort in both unadjusted ($P = 0.02$) and adjusted models ($P < 0.001$). Finally, in the discovery cohort, unadjusted models showed that BD/SA had significantly higher DNAm-based PAI-1 levels ($P = 0.019$) and smocking pack-years ($P = 0.029$) compared to HC. After covariate adjustment, BD/SA showed significantly higher PAI-1 levels ($P = 0.016$), smocking pack-years ($P = 0.022$) and GDF-15 ($P = 0.035$) compared to HC.

Conclusions: Epigenetic GrimAge acceleration may contribute to premature morbidity and mortality in BD patients with a lifetime history of suicide attempt. These findings pair with existing evidence that not only BD, but also suicide attempt, may be associated with an acceleration of biological aging, and provide putative biological mechanisms for premature mortality in these conditions (for example, through the actions of PAI-1 and GDF-15). Future studies are warranted to explore the role of epigenetic aging in the pathophysiology of BD and suicidal behavior, as well as to dissect their shared and unique biological underpinnings.

Keywords: Bipolar Disorder, Suicide Attempt, GrimAge Acceleration

Disclosure: Nothing to disclose.

P366. Reduced Glutamatergic Cortical Facilitation Associated With Glutamate-Related Gene Expression Abnormalities in Treatment-Resistant Depression

Masataka Wada*, Shinichiro Nakajima, Shiori Honda, Mayuko Takano, Keita Taniguchi, Yu Mimura, Sakiko Tsugawa, Masaru Mimura, Yoshihiro Noda

Keio University School of Medicine, Sinuku-ku, Japan

Background: One third of patients with depression do not respond to optimal antidepressant treatment, defined as treatment-resistant depression (TRD). Recent studies noted the benefit of ketamine on TRD and abnormal glutamatergic neurometabolite levels in TRD suggest that abnormalities in glutamatergic neurotransmission are implicated in the pathogenesis of TRD. Furthermore, a recent lesion network mapping study reported that the symptom network of depression may involve the dorsolateral prefrontal cortex (DLPFC) on which transcranial magnetic stimulation (TMS) treatment is also effective on TRD. The intracortical facilitation (ICF) paradigm with a combined TMS and electroencephalography (TMS-EEG) method allows the evaluation of primarily N-methyl-D-aspartate receptor-mediate neural function in target brain regions. The objectives of this study were 1) to compare glutamatergic neural activity as indexed by the ICF paradigm at the DLPFC between patients with TRD and healthy controls (HCs) using TMS-EEG and 2) to explore the links between the cell-specific glutamatergic gene expression and abnormalities of glutamatergic neural activity in TRD using virtual histology approach.

Methods: This study was approved by the ethical committee at Keio University School of Medicine and the registration identification number of this study is UMIN000028863. Sixty patients with TRD and thirty HCs received 80 single-pulse TMS and paired-pulse TMS at an interstimulus interval of 10 ms at the left DLPFC to measure the evoked potential. The degree of ICF was assessed by the difference between paired-pulse and single-pulse evoked potentials. Differences in ICF between TRD and HCs groups were calculated by cluster-based permutation analysis. Additionally, we calculated the correlations of interregional profiles between altered glutamatergic neural activity and the glutamate-related gene expressions derived from the Allen Human Brain Atlas dataset.

Results: ICF level at the electrode sites above the left DLPFC was not significantly different between the two groups; however, ICF level at the left DLPFC just beneath the TMS coil was decreased compared with that of HCs ($p = 0.014$). Furthermore, the distribution on the brain surface of the decreased ICF level correlated with the distribution on the brain surface of the specific glutamate-related gene expression ($p = 0.016$).

Conclusions: A reduced level of ICF at the left DLPFC represents a pathophysiological endophenotype of TRD and may be associated with glutamate-related gene expression.

Keywords: TMS-EEG, Glutamate, Treatment-Resistant Depression, Imaging-Genetics

Disclosure: Nothing to disclose.

P367. Developing Neuroimaging Biomarkers for Depression Secondary to Neurological Illnesses

Mark Kvarta*, Yizhou Ma, Si Gao, Heather Bruce, Joshua Chiappelli, Peter Kochunov, Elliot Hong

University of Maryland School of Medicine, Maryland Psychiatric Research Center, Catonsville, Maryland, United States

Background: Depression is highly prevalent in individuals with neurological disease, in particular epilepsy, migraine, multiple sclerosis, and stroke, with lifetime depression rates after neurological illness onset as high as ~50%. Co-morbidities between depression and neurological insults are well described, although data on the neurobiological underpinnings are lacking. DSM-defined Major Depressive Disorder (MDD) is associated with well replicated structural brain changes, e.g., reduced volumes in frontal cortices and hippocampus, while the neurobiological origin of Depressive Disorder Due to Another Medical Condition (incl. neurological) is poorly understood and rarely rigorously studied.

If biophysical aspects of brain function contribute to MDD, then a neurological illness might change brain structures in a way that resembles patterns associated with MDD as a systems-level neurobiological explanation why some neurological patients develop depressive symptoms. Using neuroimaging and longitudinal behavioral data from the UK Biobank (UKBB) and applying a Regional Vulnerability Index (RVI) to quantify individual brain-wide phenotypic similarity to MDD (RVI-MDD), we tested the hypothesis that a neurological insult can cause brain changes to resemble the deficit patterns observed in MDD to increase risk for depressive symptoms.

Methods: UKBB is a large prospective study to identify determinants of health in middle to old age. We used data from two UKBB visits: the initial assessment visit (v0) and the second visit with brain imaging (v2). A continuous measurement of depression symptomatology obtained at v0 and v2 was obtained using the Recent Depressive Symptoms-4 (RDS-4) scale, a measure validated for neuroimaging study within UKBB with questions corresponding to several MDD criteria (Dutt et al., 2022). RDS-4 has high agreement with other scales like PHQ-9 when obtained concurrently, with an advantage that RDS-4 questions were collected at repeat UKBB visit dates including when imaging data was collected. Total depressive symptom scores at either visit were the sum of the answers given (including up to 1 imputed missing score). 46,910 participants (51.3% female) in UKBB completed at least 3 of the RSD-4 depressive symptom questions at both visits and were included. We identified individuals with new report of neurological illness between v0 and v2, a prospective approach in which v0 data can be considered baseline prior to onset of neurological illness (NI). The list of neurological diagnoses included infectious, multiple sclerosis, epilepsy, migraine, intracerebral hemorrhage, and stroke.

Imaging data was obtained from UKBB neuroimaging data release version 1.6. RVI calculations require large imaging samples aggregated from independent imaging studies of an illness to establish its 'gold standard', here using the ENIGMA consortium data for MDD. UKBB phenotypes included 24 regional white matter tract FA values, 33 regional estimates of cortical gray matter (GM) thickness, volumes of the lateral ventricles, and 7 subcortical gray matter volumes per hemisphere corresponding to these derived by ENIGMA workflows. RVI-MDD scores were calculated using 'RVIpkg' in [R]. After regressing out covariates, imaging phenotypes were transformed to z-scores, and the Pearson correlation coefficient calculated between a participant's z-scores and corresponding effect sizes for MDD patient-control group differences from the ENIGMA consortium for MDD (Schmaal et al., 2017).

Results: In first validating RVI-MDD in UKBB data outside of neurological illness, we found individuals with MDD had higher cortical RVI-MDD compared to those without MDD ($p = 4\times10\text{-}7$). Amongst the non-neurological control group (NC), those with an increase in depressive symptoms between visits had higher RVI-MDD than those with no worsening in mood ($p = 3\times10\text{-}6$). Thus, both cross-sectional and longitudinal data supported validity of applying the RVI-MDD derived cortical deficit pattern to UKBB depression symptom data.

There were no significant differences in age and sex ratio between NI ($n = 890$) and NC group ($n = 46{,}020$). NI had higher RDS-4 depression scores at v0 prior to neuro illness onset compared to NC ($p = 4\times10\text{-}7$), while both NI and NC showed reduced depression from v0 to v2 ($p = 3\times10\text{-}15$). A higher proportion of individuals in the NI group reported worsening depression scores (30.8%) than the NC group (24.9%) at v2 ($p = 6\times10\text{-}5$). Among NI, RVI was higher for those with worsened depressive score compared to those with no worsening ($p = 0.021$). In a full 2×2 ANCOVA model, there was a significant group*depression change interaction ($F(1,35550)=4.02$, $p = 0.045$). NI with worsened depression had the highest RVI-MDD, supporting that RVI can separate NI by depression score status. In comparison, polygenic risk score for MDD was not different between NI and NC groups ($p = 0.70$).

Conclusions: This study demonstrates longitudinal, quantitative changes in mood scores before and after serious neurological events, and that these changes drive cortical deficit changes towards a brain pattern similar to MDD. Those individuals with a neurological insult during the study period who reported worsening depression scores had a higher RVI than those who did not. These data add new neurobiological insight into the etiology of depression after neurological illness. RVI for MDD may provide a biomarker to index those who are vulnerable to develop depression associated with neurological illnesses.

Keywords: Human Neuroimaging, Brain Based Markers for Depression, Neurological Disorders, Depression, Big Data

Disclosure: Nothing to disclose.

P368. Persistent Brain Connectivity Changes in Healthy Volunteers Following Nitrous Oxide Inhalation

Charles Conway*, Peter Nagele, Thomas Zeffiro, Britt Gott, Thomas Nguyen, Charles Zorumski, Ben Julian Palanca

Washington University School of Medicine, Saint Louis, Missouri, United States

Background: Nitrous oxide (N2O), an N-methyl D-aspartate (NMDA) receptor antagonist with similarities to ketamine, has shown promise as a therapeutic in treatment-resistant major depressive disorder (TRMD) and PTSD. Similar to ketamine, the antidepressant effects persist well beyond a single one-hour inhalation. Previous studies have demonstrated that during N2O

inhalation changes in electroencephalographic signals and altered brain networks occur. Of note, electroencephalographic studies of ketamine in depressed patients demonstrate persistent plasticity changes in occipital cortex. This brain imaging trial examines subacute and persisting network brain connectivity changes 2 hours and 24 hours following exposure to one hour of inhaled N2O using resting state functional magnetic resonance imaging (rs-fMRI).

Methods: Sixteen healthy volunteers of both sexes were recruited in a double-blinded crossover study. Individuals underwent randomized inhalation sessions of 50% nitrous oxide/oxygen or oxygen/air mixture for one hour. Three 10-minute rs-fMRI scans were obtained prior to inhalation, and at 2-hours and 24-hours after inhalation sessions. Based on literature support of involvement in mood disorders, four a priori canonical network seeds were chosen: default mode, dorsal attention, affect, and reward networks. rs-fMRI inter-regional correlations were measured using echo-planar imaging. Inter-regional correlations were computed and converted to Fisher z scores from time series of preprocessed, motion artifact-scrubbed, and nuisance covariate-regressed time series data. To avoid bias introduced by serial testing, false discovery rate (FDR) corrections were employed. First-level whole brain modeling estimated change in connectivity strength (pre- vs post-N2O) for both a local connectivity strength using local correlation (LCOR) metric and global connectivity strength using a global correlation metric (GCOR). Seed-based functional connectivity was subsequently used to characterize the spatial pattern of connectivity changes associated with areas that showed changes in LCOR and GCOR. Second-level modeling (pre- vs post-N2O) built upon the first-level model findings used a repeated measure, mixed-effects model, with fixed effects of replication, session, condition and subject as a random effect, selecting the primary visual cortex as the seeded region.

Results: In comparisons of pre- to post-nitrous oxide inhalation, the a priori selected canonical networks did not reveal significant changes in resting-state functional connectivity. However, exploratory whole brain analyses demonstrated that, compared to placebo, N2O inhalation was associated with statistically significant changes in global brain connectivity that persisted in occipital cortex at 2- and 24-hours post inhalation (p-FDR = 0.019). Further, second-level modeling across 200 grey matter regions demonstrated stronger post-N2O correlations with the bilateral insula, lateral parietal, and middle cingulate regions, as well as bilateral cerebellar regions (all $p < 0.05$, FDR-corrected with region-based statistics). Further, analysis of resting-state networks demonstrated robust strengthening of connectivity between regions of the visual network and those of the dorsal attention network, both at 2- and 24-hours after inhalation (Cluster TFCE = 55.94 p = 0.041).

Conclusions: N2O inhalation in healthy volunteers revealed persistent increases in global connectivity between regions of primary visual cortex and dorsal attention network. These findings suggest that N2O inhalation induces neurophysiological cortical changes for at least 24 hours and are consistent with reports of synaptic plasticity in visual networks induced by ketamine. The sustained nature of these findings is compelling, as it parallels the observed N2O antidepressant clinical findings as well as the previous neuronal plasticity ketamine studies and suggests that these NMDA-antagonists may bring about their antidepressant effects, in part, by persistently altering network connections in the visual and attention realms. Further brain imaging studies of N2O in depression are warranted.

Keywords: Nitrous Oxide, Antidepressants, Functional Connectivity, Networks, NMDA

Disclosure: Nothing to disclose.

P369. Longitudinal Effects of Subcallosal Cingulate Deep Brain Stimulation on the Resting Electroencephalogram in Treatment-Resistant Depression

Tanya Nauvel*, Sankar Alagapan, Mosadoluwa Obatusin, Stephen Heisig, Jacob Dahill-Fuchel, Patricio Riva-Posse, Christopher Rozell, Allison Waters, Helen Mayberg

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

Background: Major depressive disorder affects approximately 14.8 million adults in the US (Kessler et al, 2005). While most cases of depression are treatable, up to 10% of the patients do not respond to conventional interventions using various combinations of medication, psychotherapy and somatic treatments including ECT. Deep brain stimulation (DBS) of the subcallosal cingulate (SCC) white matter is an emerging new treatment strategy for such treatment resistant depression (TRD) with published studies demonstrating sustained long-term antidepressant effects in 73% of implanted patients (Riva-Posse 2018).

While long-term effects are generally maintained once achieved, clinical observation and ratings as well as local LFP electrophysiological recordings in the SCC (Alagapan et al, 2021) show that recovery appears to have multiple stages: depressed (defined as <50% Hamilton change); first well (acute behavioral effects in the OR); rough (improved but unstable); and well (>50% Hamilton Depression Ratings Scale). These changes occur with a variable timecourse across individuals. Here we test if the resting EEG demonstrates comparable changes to the LFP and behavioral changes previously observed.

Methods: EEG was recorded in two cohorts of TRD patients receiving chronic SCC DBS using two DBS systems: Medtronic Activa PC + S r (n = 8) and Medtronic Summit RC + S (n = 5). All patients were tested at 8 time points: before DBS implantation surgery; 4 weeks post-surgery without ongoing DBS; once a month for 6 months during active stimulation. Recordings consist of 5-minute eyes open and eyes closed resting EEG data (EGI, 256 channels), collected with bilateral stimulation both ON and OFF.

Time frequency domain analyses of the resting EEG data over time were first calculated over 3 second windows using the Thomson multitaper method (5 tapers) and average power over 5 frequency bands (θ: 4-8 Hz, α: 8-12 Hz, low β: 12- 20 Hz, high β: 20-30 Hz and γ: 30-50 Hz) was calculated and compared across timepoints.

To track the time course of recovery, we clustered the spectral information from a subset of the 256 channels and tracked the percentage of the "well" cluster (extracted from the final 6 months visit) compared to all the other timepoints. We then compared this time course to the transitions defined by a neural network classifier used to distinguish SCC LFP dynamics that reflect differences in clinically defined 'depressed' and'well' states.

Results: Changes in EEG power over time show that there are differential early (1 month) and late (2-6 month) band-specific EEG changes in the first PC + S cohort of patients (Two Group Test, $P \leq 0.05$). These are similar to the timelines reported in PET and LFP studies within the same cohort of patients. The EEG spectral findings are replicated in the second cohort. Topoplots show different patterns of power changes across each frequency band: alpha and low beta frequency changes localize to parietal/occipital channels, whereas beta power increases are more centrally located around Cz. Resting EEG spectral clusters transitions from 'sick' to 'well' occur at similar timepoints as the LFP classifier transitions.

Conclusions: These findings provide evidence that the resting EEG can identify distinct early and late response change patterns

over the course of successful SCC DBS treatment for TRD. The response trajectory over 6 months involves 3 to 4 specific brain states transitions. The similarity of the identified sick-well transitions using EEG to those previously identified in the SCC LFP recordings suggest a nonlinear evolution of SCC-cortical network changes over time with ongoing DBS. These findings invite further consideration of the role of serial resting EEG to identify treatment milestones that might guide treatment optimization.

References:

1. Kessler et al. Arch Gen Psychiatry, 2005. 62:593–602.
2. Riva-Posse et al. Mol Psychiatry, 2018. 23:843-849
3. Alagapan et al. Brain Initiative Poster, 2021

Keywords: Brain Based Markers for Depression, Deep Brain Stimulation, Quantitative Electroencephalography (qEEG)

Disclosure: Nothing to disclose.

P370. Sex and Drinking: Sex Differences of the Effects of Chronic Stress on Drinking Behavior of C57Bl6 Mice in the Sucrose Preference Test Measured With Lickometry

Andreas Wulff, Scott Thompson*

University of Colorado School of Medicine, Aurora, Colorado, United States

Background: Depression is a devastating and prevalent mental illness. Rodent models are widely used to understand the neurobiology of depression and in the testing of potential novel antidepressants. As chronic stress is a major risk factor for the development of depression in humans, it is often used to precipitate depression-like behaviors in rodents. A wide range of behavioral outputs are used to measure cognitive, rewarding and other behavioral impairments following chronic stress. Among these is the sucrose preference test used to measure an anhedonic-like phenotype following chronic stress. However, sex as a biological variable has largely been overlooked in these rodent experiments and it is still not clear how the drinking behavior of male and female mice during the sucrose preference test relates to hedonic state.

Methods: We used a custom-built homecage lickometry setup to record the drinking behavior of male and female C57Bl6 mice undergoing a sucrose preference test (n = 8-17 per group and sex). We analyzed the changes in drinking as the night progressed and further identified 'bursts' of drinking, which has been used as a measure of palatability in prior studies of licking microstructural analysis. We then perturbed the sucrose preference test by either replacing 1% sucrose with 0.1% sucralose, or by subjecting the mice to acute stress or mild food restriction. Finally, we tested how drinking behavior was affected following chronic multimodal stress. When testing the effects of chronic stress, we further complemented the sucrose preference test with other behavioral tests of hedonic state, curiosity, and pain sensitivity.

Results: At baseline, male and female mice exhibited a phasic drinking behavior with increased drinking at the beginning and end of the dark phase of a 12:12 light:dark cycle. Both male and female mice exhibited a strong preference for the 1% sucrose solution as observed by a preference in volume consumed as well as by increased drinking at the sucrose bottle and increased number and length of drinking bouts. Mice also exhibited a strong preference for the non-caloric 0.1% sucralose which could also be observed in the number of licks, as well as number and length of lick bouts. Neither acute stress nor mild food restriction appeared to affect the drinking behavior significantly in either male or female mice but chronic stress precipitated a decrease in sucrose preference. When comparing the drinking behavior of male and female mice, we observed that female mice drank in more, but shorter, bouts compared to the males. The female mice were also less susceptible to the chronic stress with males exhibiting a strong decrease in drinking at the sucrose bottle. However, while male mice had a strong decrease in the number of drinking bouts at the sucrose bottle following chronic stress the length of the bout was increased at both the water and sucrose bottle and the relative length of the bout was unchanged between sucrose and water following chronic stress. This effect of stress was also seen in female mice. In this cohort however, we also did not see impairments of other tested hedonic behaviors or of curiosity, but we did observe a heightened pain sensitivity in both male and female mice following chronic stress. Chronic stress also affected the phasic drinking with a stronger peak in drinking at the beginning of the dark phase but no increase in drinking towards the end of the dark phase.

Because the length of drinking bouts is usually taken as a measure of acute rewarding response to a palatable solution, our data indicates that a loss in sucrose preference following chronic stress may not be due to a lack of reward responsivity. Instead we saw a decline in the number of lick bouts which could indicate impairment of metabolic or cognitive processes instead. What causes a loss to the sucrose preference may depend on species, strain and/or stressors.

Conclusions: Male and female C57Bl6 mice both exhibit a preference for sucrose, but the microstructural drinking behavior is slightly different. We further found that female mice were less susceptible to chronic stress than male mice. Finally, we found indications that under the conditions used in this experiment, the impaired sucrose preference following chronic stress may be due to impairments of cognitive or metabolic impairments rather than impairments in reward responsitivity.

Keywords: Reward Deficit, Anhedonia, Stress

Disclosure: Terran Biosciences: Patent (Self), Ambit: Patent (Self)

P371. Sex-Specific Anti-Angiogenic Mechanisms Unite Brain and Vascular Dysfunction in the RGS2 Knockout Model of Psycho-Obstetric Risk

Serena Gumusoglu*, Michaela Kiel, Brandon Schickling, Kaylee Weaver, Marisol Lauffer, Hannah Sullivan, Kaylie Coulter, Yuping Zhang, Eric Devor, Donna Santillan, Mark Santillan

University of Iowa Carver College of Medicine, Iowa City, Iowa, United States

Background: Gestational hypertensive disorders such as pre-eclampsia share high rates of comorbidity with mood disorders and anxiety, which in-turn appear to increase risk up to 3-fold for preeclampsia. This bi-directional risk suggests shared mechanisms, which may reveal insights into potential treatment targets for these often-dangerous disorders. Prior work by our group revealed that loss of RGS2, a risk gene for preeclampsia and psychiatric disorders including anxiety, depression, and suicide, results in preeclampsia-like obstetric phenotypes such as gestational hypertension and placental dysfunction in a murine model. Here we examine conserved, RGS2-mediated mechanisms that might disrupt both brain and vascular function to increase risk for preeclampsia and psychiatric disease in females in particular.

Methods: Adult RGS2 knock out (KO) and wildtype (WT) littermate controls (n = 6 per genotype per sex) were behaviorally tested using the Y-maze, open field, sucrose preference test, tail suspension, social approach, and elevated plus maze (EPM) assays. Brains from these animals were hemisected, with half reserved fresh for qPCR (dissected cortex, midbrain, and hypothalamus/paraventricular nucleus) via qPCR and half formalin-fixed and labeled with IB4 lectin (cerebrovascular marker), NeuN, and DAPI. ImageJ and stereology were used to calculate vascular and cellular

density. Platelet-free plasma serotonin (5-HT) was measured by ELISA. A separate cohort of animals (n = 3-7 per genotype per sex) were tested for peripheral vascular dysfunction. Blood pressure was assessed by tail cuff plethysmography (CODA) and aortas were dissected and studied for redox and angiogenic factors by qPCR. Males and females were analyzed separately. PRISM was used for data analysis and depiction.

Results: Male RGS2KO mice exhibited hyperlocomotion (P = 0.05), increased latency to explore (Y-maze) (P = 0.048), and impaired social memory (P = 0.02). Female RGS2KOs exhibited decreased center exploration (P = 0.05) and increased struggle on the tail suspension assay (P = 0.005). Both male and female RGS2KO mice exhibited anxiety-like (EPM) and hedonic (increased sucrose preference) behaviors (P = 0.04, 0.02 respectively). RGS2KO mice also exhibited significant reductions in cortical vascular density (males P = 0.05, females P = 0.144). Cortical thickness, white matter, commissural thickness, and cortical cell density were unchanged. Molecular profiling of the cortex, midbrain, and hypothalamus/paraventricular nucleus via qPCR revealed a significant increase in cortical HTR2A (P = 0.006) and MAOA (P = 0.03) in females, and in hypothalamic SLC6A4 in males (P = 0.02). Peripheral, platelet-free plasma 5-HT was unchanged by sex or genotype. In preliminary assessments, blood pressure was also unchanged by sex or genotype. Aorta assessments revealed significant upregulation of NOX4 (P = 0.002) and HIF-1A (P = 0.04) in females, and PDGFRb (males P = 0.07, females P = 0.05). MMP-9 was also significantly downregulated in RGS2KO males (P = 0.05).

Conclusions: Collectively, these results demonstrate that in the RGS2KO, animals exhibit sex-specific alterations in mood and other behaviors, serotonergic gene expression in the brain, in peripheral vascular redox and angiogenesis, and in cerebral vasculature density. Some of these results demonstrate female-specific vulnerabilities. The disruptions we report are not driven by hypertension, rather shared vascular redox and angiogenic dysregulation mechanisms may underlie both peripheral and cerebrovascular and brain function changes. These mechanisms may also link the shared pathogenesis of anxiety/depression to preeclampsia. This points to valuable future therapeutic targets and avenues for further mechanistic exploration.

Keywords: Pregnancy, Cardiovascular Function, Serotonin, Depression and Anxiety, Angiogenesis

Disclosure: Nothing to disclose.

P372. Inflammation Moderates Antidepressant Response to Ketamine in a Rodent Model of Treatment-Resistant Depression: Bioenergetic and Inflammatory Markers of Response

Susannah Tye*, J. Blair Price, Adam Walker, Jennifer Kruse, Kathryn Cullen, Mark Frye, Lilly Schwieler, Sophie Erhardt

The University of Queensland, St Lucia, Australia

Background: Low dose ketamine directly stimulates neurotrophic signaling to promote neuroplasticity and antidepressant response. Despite a clear biological mechanism for target engagement, moderating factors are not well understood. The present study examined the effects of inflammation on response to ketamine in a preclinical rodent model of treatment resistant depression.

Methods: Male Wistar rats (n = 86) received adrenocorticotropic hormone (ACTH; 100 µg/day, 22 days), lipopolysaccharide (LPS; 750-1250 µg/kg, days 17-22), and/or control vehicle saline (0.9% w/v, 22 days). Sucrose preference testing was conducted every second day across the course of this treatment. Animals were administered ketamine (10 mg/kg, final two days) before the open field and forced swim tests (FST) to screen for antidepressant effect. Prefrontal cortex (PFC) tissue was collected from animals' postmortem to assay kynurenine, kynurenic acid (KYNA), and 3-hydroxykynurenine (3HK) using high-performance liquid chromatography. Enzyme-linked immunosorbent assays (ELISA) were used to determine protein levels in the PFC (i.e., mammalian target of rapamycin (mTOR), 5'adenosine monophosphate-activated protein kinase (AMPK), and glycogen synthase kinase-3 α/β (GSK3α/β). Serum levels of C-reactive protein (CRP) and corticosterone were determined via ELISA.

Results: Significant interaction effects (p = 0.0493) and effects of LPS and ketamine treatment (p = 0.0017) on behaviors in the FST were observed for total immobility time. Notably, a robust antidepressant-like response was observed in animals receiving ACTH + LPS + ketamine, reducing immobility levels relative to ACTH-controls (p = 0.0017) and animals receiving ACTH + LPS (p = 0.0439). Similarly, immobility levels expressed by ACTH + ketamine treated animals were significantly lower than ACTH-controls (p = 0.0027), but not ACTH + LPS-treated animals. No significant effects of ketamine treatment on immobility time were observed among saline-treated animals, and no group differences were observed for other behavioral measures. ACTH + LPS treated animals had significantly higher levels of prefrontal KYNA than animals receiving ACTH + LPS + ketamine (p = 0.0037) or ACTH-controls (p < 0.0001). Conversely, ACTH + LPS + ketamine treated animals had significantly higher kynurenine/KYNA ratios compared to ACTH + ketamine treated animals (p = 0.0008) and ACTH + LPS-treated animals (p = 0.0001). ACTH-controls had higher kynurenine/KYNA compared to ACTH + ketamine treated animals (p = 0.0117) and ACTH + LPS-treated animals (p = 0.0014). ACTH + LPS-treated animals also had significantly greater KYNA/3-HK compared to ACTH-controls (p = 0.0393) and ACTH + ketamine treated animals (p = 0.0102). No significant differences were observed among saline-treated animals for tryptophan metabolites, however Saline+LPS + ketamine treated animals had significantly higher levels of prelimbic AMPK and GSK3α/GSK3β relative to saline-controls (p < 0.05). A significant positive correlation was observed between FST immobility time and serum CRP concentration across all treatment groups.

Conclusions: Ketamine directly attenuated LPS-induced elevations in levels of tryptophan metabolites in the PFC of animals treated with ACTH, potentially contributing to the antidepressant-like behavioral response within the context of this model. Neither LPS nor ketamine had any significant effect on prefrontal tryptophan metabolite levels in control saline-treated animals. CRP levels across all groups were associated with immobility time in the FST, further supporting the role of inflammation in moderating stress coping. Targeted research in clinical populations is needed to determine the utility of inflammatory markers in precision medicine treatment stratification. Ketamine's mechanism of action may optimally promote antidepressant response in individuals with elevated inflammatory markers within the context of TRD.

Keywords: Treatment Resistant Depression, Ketamine, Insulin, mTOR, Immune System

Disclosure: Nothing to disclose.

P373. Dissociable Gene and Circuit Networks in VTA Dopamine Subpopulations

Larry Zweifel*, Jordan Elum, Barbara Juarez, Grigory Loginov, Scott Ng-Evans, Sam Golden

University of Washington, Seattle, Washington, United States

Background: Dopamine neurons of the ventral tegmental area (VTA) are genetically and functionally distinct. How the circuit

connectivity and gene expression patterns within these populations endow their unique functionality is not resolved. We have endeavored to resolve how genes and circuits intersect at the level of the mesolimbic dopamine neuron subpopulations to establish a more granular understanding of the specialized roles of this system in motivated behavior, reinforcement learning, and decision making.

Methods: Fiber photometry was used to measure calcium dynamics in genetically distinct dopamine populations during cue-induced reinstatement, probabilistic reinforcement, and reward valuation. Sice electrophysiology was performed to measured synaptic connectivity and intrinsic properties of these cells. Analysis of single nuclear RNA sequencing was employed to establish gene network differences. Cell-specific rabies tracing, light sheet microscopy, and brain wide connectivity analysis was used for 3D reconstruction of circuit connections.

Results: We discovered that dopamine subpopulations that project to either the nucleus accumbens core or nucleus accumbens shell display differential calcium dynamics during cue-induced reinstatement, probabilistic reinforcement, and reward valuation. Whole-brain connectivity analysis revealed differential inputs to these cells that was confirmed by in vivo optical stimulation of these inputs and imaging of subpopulations. Electrophysiology revealed that these subpopulations have differential inhibitory connectivity and intrinsic excitability. Consistent with these findings disinhibitory networks evoke dissociable response profiles in these cells. Finally, our gene expression analysis reveals that differential ion channel expression within these cells contributes to their excitability and stimulus response profiles.

Conclusions: By combining circuit, gene, systems, and behavioral analyses we have established that both distinct and graded difference in circuit connectivity, gene expression, and intrinsic excitability of VTA dopamine subpopulations converge to impart the unique functionality of these cell types within the mesolimbic system of the brain.

Keywords: Dopamine, Behavior, Neural Circuits, Molecular Genetics, In Vivo Imaging

Disclosure: Nothing to disclose.

P374. The Bed Nucleus of the Stria Terminalis Mediates the Expression of Two-Way Active Avoidance

Justin Moscarello*, Diana Guerra, Wei Wang, Karienn Souza

Texas A and M University, College Station, Texas, United States

Background: Although avoidant behavior is a unifying symptom of many anxiety disorders, its underlying neural circuitry remains a mystery. Here, we report a series of experiments using a signaled active avoidance (SAA) procedure to model avoidant behavior in male rats. In SAA, the subject rapidly acquires an association between a conditioned stimulus (CS) and an aversive unconditioned stimulus (US). As training proceeds, the subject learns to perform a response (two-way shuttling) during the CS in order to prevent US delivery. Over the course of SAA training, the US presentation decreases drastically and thus transforms from a certain threat during initial acquisition into a distal threat as the avoidance response reaches asymptotic levels of expression. Prior research demonstrates that the bed nucleus of the stria terminalis (BNST) mediates defensive responses to possible/distal conditioned threats, suggesting a possible role for this region in the expression of active avoidance behavior. However, its role in SAA has yet to be examined.

Methods: We used a virally mediated DREADD (Designer Receptors Exclusively Activated by Designer Drugs) approach to explore the role of BNST in SAA. Rats received stereotactic infusions of a solution containing one of the following AAVs, depending on the experiment: AAV5-hSyn-hM4D(Gi)-mCherry, AAV5-hSyn-hM3Dq-mCherry, or AAV5-hSyn-EGFP. Following recovery, subjects received SAA training in which a tone CS (15 sec, 70 dB) preceded a footshock US (0.7 mA, 0.5 sec). Training occurred in a rectangular chamber separated into two compartments by a divider with an open aperture that allowed free movement between compartments. If, during the CS, the subject shuttled across the chamber to the opposite side, the CS terminated, and the US was omitted. Following the completion of training, all subjects received test sessions preceded by administration of 3 mg/kg of the DREADD agonist clozapine-N-oxide (CNO) or vehicle (10% DMSO).

Results: We first tested the hypothesis that BNST is necessary for the expression of the two-way active avoidance response. Male rats received intra-BNST infusions of AAV containing the gene construct for either the inhibitory hM4Di DREADD or GFP. After recovery, animals received four days of SAA training in order to reach stable levels of avoidance. This was followed by two additional days of SAA training preceded by counterbalanced IP injections of either the DREADD ligand CNO or vehicle. We found that CNO decreased avoidance responses and increased avoidance latencies in hM4Di subjects but not in GFP controls, demonstrating that the BNST is necessary for expression of the two-way avoidance response. Because some subjects in this experiment showed hM4Di expression in the medial septum (MS), we followed up with an anatomical control experiment specifically targeting this region. We found no evidence of a role for MS, indicating that the effects observed in our initial experiment were BNST-specific. We then compared DREADD activation (hM3Dq) and inhibition (hM4Di) of BNST in a test of avoidance under extinction conditions (10 CSs, no USs) in which all groups receive a common series of stimuli. After AAV infusions and recovery, subjects received six days of avoidance training. 24 hours later, all subjects were tested following the administration of either CNO or vehicle. This experiment confirmed that CNO decreased the expression of the avoidance response in hM4Di-expressing subjects. Indeed, CNO caused the hM4Di group to avoid at levels that were highly similar to the poor avoider group, which was comprised of subjects that never successfully expressed the response during training, illustrating the degree of the behavioral decrement induced by BNST inactivation. In contrast, administration of CNO to hM3Dq-expressing subjects caused a period of potentiated shuttling that extended beyond the presentation of the CS, whereas other groups quickly dropped back to baseline levels of shuttling following CS offset. Analyses of other ongoing behavior at test revealed that these effects on two-way shuttling in hM4Di and hM3Dq groups cannot be attributed to alterations in freezing or overall locomotor activity.

Conclusions: We conclude that BNST is not only necessary for normal levels of avoidance but is also sufficient to potentiate the output of the response, establishing a region identified as a crucial substrate for distal threat processing as a key mediator of avoidant behavior. This is in contrast with prominent conceptual models of the behavioral processes underlying SAA, such as two-factor theory, which identify putative fear states most related to imminent threat processing as the primary trigger of the avoidance response. Our results argue for an updated multi-factor model in which post-encounter responses to certain threat are supplanted by pre-encounter responses to possible threat over the course of the SAA learning curve.

Keywords: Fear, Anxiety, Avoidance, Bed Nucleus of the Stria Terminalis, Predatory Imminence

Disclosure: Nothing to disclose.

P375. (R,S)-Ketamine and (26,6 S)-HNK Attenuate Learned Fear by Differentially Modulating Brain-Wide Neural Activity

Alessia Mastrodonato*, Noelle Kee, Marcos Lanio, Michelle Jin, Andrea Muñoz Zamora, Christine Ann Denny

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

Background: Stress exposure is one of the greatest risk factors for fear and anxiety disorders, such as major depressive disorder and post-traumatic stress disorder (PTSD). We previously showed that a single injection of (R,S)-ketamine prior to stress reduces behavioral despair and attenuates learned fear by modulating hippocampus ventral CA3 activity in male mice. Recently, we found that a metabolite of (R,S)-ketamine ((2 S,6 S)-hydroxynorketamine (HNK)) also attenuates learned fear. However, the whole-brain regions mediating (R,S)-ketamine and (2 S,6 S)-HNK effects on fear behavior are still largely unknown.

Methods: Here, we used a 3-shock contextual fear conditioning (CFC) paradigm as a stressor 1 week following a single injection of saline or (R,S)-ketamine (30 mg/kg) or (2 S,6 S)-HNK (0.075 mg/kg) to adult (8-week-old, $n = 10$ per group) 129S6/SvEv male mice. Five days later, mice were re-exposed to the aversive context and sacrificed an hour later to quantify neural activity (i.e., c-fos expression) across the whole brain using an analysis pipeline developed in our laboratory.

We then transformed the c-fos cell counts separated by brain region into correlation matrices and created whole-brain networks to provide information on functional connectivity.

Results: (R,S)-ketamine and (2 S,6 S)-HNK administration attenuate learned fear compared to saline mice ($p < 0.01$ for both drugs). (R,S)-ketamine increases fear-related neural activity and connectivity in several brain regions (e.g., ventral CA3, restrosplenial cortex and temporal associative area ($p < 0.01$)), while (2 S,6 S)-HNK increases the connectivity specifically within and between the prefrontal cortex and amygdalar regions ($p < 0.05$).

Conclusions: Our results indicate that (R,S)-ketamine and (2 S,6 S)-HNK attenuate learned fear by differentially altering network correlated activity. We found new nodes in the network that are altered by fear behavior so that we can target with pharmacological manipulations to alleviate fear. This work contributes to the understanding of prophylactic drugs as therapeutic aids for fear-related disorders.

Unique Data: All data are new and unpublished

Keywords: (R,S)-ketamine, (2 S,6 S)-HNK, Fear Conditioning

Disclosure: Nothing to disclose.

P376. A Stress-Sensitive Frontostriatal Circuit Supporting Effortful Reward-Seeking Behavior

Robert Fetcho*, Puja Parekh, Margaux Kenwood, Laura Chalencon, David Estrin, Jolin Chou, Megan Johnson, Conor Liston

Weill Cornell Medical College, New York, New York, United States

Background: Effort valuation—a process for selecting actions based on the anticipated value of rewarding outcomes and expectations about the amount of work required to obtain them—plays a fundamental role in decision-making. Effort valuation is disrupted in chronic stress states and is supported in part by the anterior cingulate cortex (ACC), but the circuit-level mechanisms by which the ACC represents effort-related signals and regulates effort-based decision-making and reward-seeking are not well defined.

Methods: We developed an automated, physiology-compatible platform for high-throughput assessments of effort valuation in freely moving mice. In an elevated T-maze apparatus, animals (male C57Bl/6 mice, age 8-12 weeks) were presented with the choice between water rewards of varying magnitudes that are associated with varying effort expenditure requirements. We utilized fiber photometry and optogenetics to record from and manipulate nucleus accumbens projecting ACC neurons (ACC-NAc) as mice performed this effort-based decision-making task. Following training, a subset of mice underwent chronic corticosterone exposure - a model of the neuroendocrine response to stress - and following exposure were retested on the behavioral paradigm alongside ACC-NAc recordings.

Results: We found that ACC-NAc activity anticipates and responds to reward acquisition; however, the circuit does not appear to encode reward magnitude. Instead, the magnitude of ACC-NAc reward-related activity scaled with the level of effort expenditure required to obtain the reward ($N = 16$ mice, 4384 behavioral trials, 3 experiments; linear mixed effects model: significant interaction between reward type and effort level – $F(2,4378) = 135.38$ $p < 0.0001$). We next found that optogenetic silencing of this effort-sensitive reward signal led to significant reductions in future effortful decisions within a behavioral session (Two-way repeated measures ANOVA: $N = 18$ mice (11 control, 7 experimental), 2 experiments. Significant interaction $F(1,16) = 13.06$, $p = 0.0023$. Post-hoc bonferonni testing control: baseline vs. stim $p = 0.7$, experimental: baseline vs. stim $p = 0.0001$). Finally, chronic corticosterone treatment led to significant impairments in effortful reward-seeking behavior that strongly correlated with impaired ACC-NAc circuit function ($N = 5$ mice, 10 sessions from 2 experiments. Linear mixed effects model $T(10) = 4.56$, $p = 0.002$, $R2 = 0.72$).

Conclusions: Our results show that ACC neurons support effort valuation behavior through projections to the NAc. Specifically, ACC-NAc circuit activity integrates both reward- and effort-related information and this activity is critical for reinforcing future effortful decisions. Chronic corticosterone leads to disruptions in this effort-sensitive reinforcement signal that correlate with impaired effortful reward-seeking behavior, suggesting one potential mechanism underlying stress-induced motivational deficits.

Keywords: Anterior Cingulate Cortex (ACC), Effort, Reward, Stress

Disclosure: Nothing to disclose.

P377. Nucleus Accumbens Glutamatergic Afferents Integrate Outcomes in Reward-Learning

Eshaan Iyer*, Jessie Muir, Serena Wu, Karen Wassef, Rosemary Bagot

McGill University, Montreal, Canada

Background: Alterations in reward learning are associated with depression and other psychiatric disorders. The nucleus accumbens (NAc) is a key region implicated in motivation and reward and disrupted in depression. Susceptibility to depression-relevant behavior is mediated by afferent glutamatergic projections from the ventral hippocampus (vHip) and the medial prefrontal cortex (mPFC) to NAc. The role that these glutamatergic inputs to the NAc in supporting reward learning remains relatively unexplored.

Methods: Here, using in vivo fiber photometry, we simultaneously record the population-level activity of mPFC and the vHip projections to the NAc in adult male ($n = 12$) and female mice ($n = 12$) in a two-armed bandit task.

Results: Both neural projections dynamically encode information about the outcomes of a given trial. Rewarded vs non-rewarded outcomes are associated with greater suppression in the

mPFC-NAc both shortly following a choice ($p < 0.001$) and throughout the inter-trial-interval (ITI) ($p < 0.01$). In the vHip-NAc, reward-associated suppression emerged at a longer delay following a choice ($p < 0.01$) and also continued through the ITI ($p < 0.0001$). In both pathways, this reward-associated suppression continued into following trials (mPFC: $p < 0.0001$; vHip: $p < 0.001$) revealing neural representations of outcome modulated by reward history. We observed notable pathway-way specific differences with mPFC-NAc projections tracking outcome history, while vHIP-NAc projections preferentially encode unrewarded outcomes, tracking history of loss but not reward.

Conclusions: Together, these findings demonstrate that mPFC-NAc and vHip-NAc projections integrate outcomes over time in a task-dependent manner to support reward learning. In light of earlier evidence identifying altered neural activity in these pathways in stress-induced states, these neural circuits may be relevant to understanding alterations in reward learning in depression and other stress-related disorders.

Keywords: Probabilistic Reward Learning, Glutamatergic, Nucleus Accumbens Glutamatergic Afferents, mPFC, Ventral Hippocampus

Disclosure: Nothing to disclose.

P378. Identifying the Mechanisms of Noradrenergic Modulation of BLA-mediated Avoidance Behaviors

Sean Piantadosi*, Veronica Porubsky, Madison Martin, Avi Matarasso, Tammy Nguyen, Michael Bruchas

University of Washington, Seattle, Washington, United States

Background: Acute stress and threat produce physiological anxiety thought to facilitate planning and allow an organism to adapt its behavior for future exploration of the environment. This serves as an adaptive mechanism that allows anxiety-like behavior and avoidance to be tuned and selected. However, in many mental health disorders this homeostatic behavioral response becomes maladaptive and dysfunctional, leading to excessive anxiety in scenarios where it is unwarranted or undesirable. Anxiety disorders, which are typified by this maladaptive response, are the most common mental illness in the U.S., affecting roughly 20% of the adult population. Clinically, we know that neuromodulators such as norepinephrine (NE) play pivotal roles in long-term outcomes following stress exposure. Despite this, the mechanism by which these monoamine neuromodulatory signals regulate circuit activity associated with anxiety-like behavior remains poorly understood. One key modulatory system well positioned to mediate these behaviors is the locus coeruleus noradrenergic system (LC-NE), as it is one of the first engaged following stressful events and stimuli and activation of the LC and its projections to the basolateral amygdala (BLA) is anxiogenic. However, we know very little about how this critical neuromodulatory LC-BLA circuit generates anxiety-like behavior at the network, circuit, cell type, transmitter, and receptor level.

Methods: We conducted two-photon calcium imaging of individual LC-NE neurons using an endoscopic prism lens while mice ($n = 5$, 3 male, 2 female) were exposed to the potent predator stressor 2MT. Next, we modeled the effect of stress on LC activity and measured NE release in the BLA using a NE sensor (GRABNE2m) while activating LC terminals in the BLA at a tonic (5hz) frequency using combined optogenetics and fiber photometry ($n = 6$, 6 male). We then performed freely moving microendoscopic calcium imaging of BLA neurons in classic anxiety-like behavior assays while manipulating LC terminals optogenetically at the stress-induced tonic frequency (Dbh-cre mice; $n = 8$, 5 male, 3 female). Pharmacological manipulations (using selective α1 and β2 antagonists) were used to discern which adrenergic receptors were mediating the anxiogenic effect of LC-BLA stimulation. Support vector classifiers were trained to predict whether mice were in an anxiogenic context based solely on BLA population activity. Graph theory analyses were developed to test whether tonic LC terminal activation would cause correlated increases in activation of BLA neurons as might be expected of a gain control signal. Finally, CRISPR-SaCas9 knockdown of gene which encode adrenergic receptor (AR) subtypes (α1- and β2-ARs) within the BLA was combined with microendoscopic calcium imaging to evaluate the necessity of these receptors for the expression of anxiety-like behavior following stress and how each of these receptor subtypes on specific cell types contributes to BLA population encoding of anxiety-state.

Results: LC-NE neuron activity was found to be highly synchronous when mice consumed an appetitive stimulus. When mice were exposed to the predator odor stressor 2MT, exploratory licking behavior was dramatically reduced ($t(3)=2.82, p < 0.05$). This reduction in exploratory licking coincided with an increase in tonic firing rate in a large subset of LC-NE neurons ($t(37)=18.1, p < 0.0001$), suggesting that in many LC-NE neurons stress induced tonic activation, though responses became less synchronous as a population. We next modeled the effect of this increased tonic activation on NE release within the BLA, finding that 5hz activation of LC-BLA terminal activation produced sustained increases in NE release as measured by GRABNE2m signal ($p < 0.001$). This effect was blocked by the α2-AR antagonist yohimbine. Using combined optogenetics and freely moving microendoscopy, we recorded the activity of individual BLA neurons while manipulating LC-BLA terminals at a tonic frequency and assessed anxiety-like behaviors. We found that tonic LC-BLA activation was anxiogenic (RM-ANOVA, $p < 0.05$; post-hoc $t(7)=3.41, p < 0.05$), promoted a shift in activity of BLA neurons that were activated when mice entered the anxiogenic context (center of open field, and open arms of elevated zero maze), and improved the ability of BLA population activity to classify mouse location within the arenas ($t(6)=2.95, p < 0.05$). These effects were blocked by the β2-AR antagonist propranolol ($p > 0.05$). Tonic LC-BLA activation produced sustained changes in the correlation structure of BLA neurons that lasted after stimulation ended, suggesting that mimicking stress by specific activation of LC inputs to BLA produces sustained changes in BLA network activity. CRISPR-SaCas9 knockdown of Adrb2 (gene encoding β2-AR) in Vglut1 neurons in the BLA was effective and resulted in a roughly 40% knockdown.

Conclusions: Neuromodulation, and specifically the neuromodulator norepinephrine, has long been suggested to serve as a gain control signal capable of shifting the activity of large populations of downstream neurons. Here using a combination of behavioral, optical, and molecular approaches we show that sustained release of norepinephrine following stress or optogenetic activation produces lasting alterations in BLA network activity, consistent with this theory, and identify the potential receptor (β2-AR) that might be necessary for this type of network shift.

Keywords: Locus Coeruleus (LC), Norepinephrine, In Vivo Calcium Imaging, Basolateral Amygdala, Stress and Anxiety Behavior

Disclosure: Nothing to disclose.

P379. Early Life Adversity Causes Fear Generalization by Impairing Serotonergic Modulation of the Ventral Dentate Gyrus

Lauren Malave*, Rushell Dixon, John Bickle, Christoph Anacker

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

Background: Having a history of early life adversity (ELA), such as physical or emotional trauma experienced during sensitive

periods in early development, increases risk for psychiatric disorders later in life and reduces responsiveness to antidepressant treatment. ELA exerts long-lasting changes on the developmental trajectory of neural circuits and neurotransmitter systems, such as the serotonergic system, in a way that may be different from the effects of adult stress or genetic predisposition. Previously, we found that hyperactivity of the ventral dentate gyrus (vDG) region of the hippocampus increases stress responses. Here, we investigate the interaction between ELA, serotonin (5-HT), and DG function on fear overgeneralization, which is a form of cognitive distortion that is characteristic for variety of mental illnesses. Understanding the neurobiological mechanisms underlying the specific contributions of ELA to fear overgeneralization may thus help us develop better treatments or preventions for psychiatric disorders.

Methods: To induce ELA in mice, we used the limited bedding and nesting (LBN) model from postnatal day (P) 3-10. LBN causes fragmented and unpredictable maternal care, as well as rough handling and stepping on pups. To examine if the effects of ELA can be rescued by increasing 5-HT signaling, we used a transgenic mouse model to conditionally knockdown 5-HT1A auto receptors on raphe 5-HT neurons (Pet1-tTS; Htr1atetO/tetO mice), resulting in increased 5-HT signaling starting at birth. Control and LBN-exposed offspring with and without 5-HT1A knockdown were tested during adolescence (P35) and adulthood (P56) in a fear discrimination task. Mice received a foot shock in context A on day 1 and were then re-exposed to context A on day 2. Two hours after re-exposure to the shock-associated context A, mice were placed in a new, safe context B. Freezing behavior was recorded using FreezeFrame software (Actimetrics) to compare fear expression between context A and context B on day 2. We also used immunohistochemistry for the immediate early gene, Fos, to measure the activity of raphe 5-HT neurons and of vDG granule neurons after exposure to context B. Tissue levels of 5-HT in the vDG of control and ELA-exposed mice were measured using HPLC.

Results: Female, but not male, mice exposed to LBN overgeneralize between a foot-shock associated context A and a safe context B in adulthood at P56, as indicated by a lower discrimination ratio between the two contexts (Wild-Type (WT) control: 0.63 ± 0.04 $n = 26$; WT ELA: 0.34 ± 0.1 $n = 14$; 2way ANOVA ELA vs. genotype interaction $F(1,55)=7.7$, $p = 0.01$; Tukey's post hoc, WT control vs. ELA $p = 0.01$). Increasing 5-HT levels rescued the behavioral deficits observed in adulthood (WT ELA: 0.34 ± 0.1 $n = 14$; 5-HT1A knockdown ELA: 0.66 ± 0.06 $n = 11$; 2way ANOVA ELA vs. genotype interaction $F(1,55)=7.7$ $p = 0.01$; Tukey's post hoc, $p = 0.02$). These differences were only observed at P56 and not at P35 for all groups, indicating that LBN leads to cognitive deficits in adulthood. At P56 in females, we found increased Fos expression in the vDG of ELA-exposed mice following exposure to the safe context B compared to controls (WT control: 67.4 ± 4.1 $n = 3$; WT ELA: 42.5 ± 5.6 $n = 6$; 2way ANOVA ELA effect $F(1,11) = 8.5$, $p = 0.01$; Tukey's post hoc, control vs. ELA $p = 0.02$). This increase in Fos expression was only found in the vDG, an area involved in anxiety-related behaviors, and not in the dorsal DG, an area involved in spatial navigation. This increase in vDG Fos expression was rescued by 5-HT1A knockdown, similar to the effect of 5-HT1A on rescued fear overgeneralization. Percentage of Fos+ 5-HT neurons in the median raphe nucleus (MnR), the nucleus projecting to the hippocampus, was reduced in mice that experienced ELA (Control: 16.4 ± 1.02 $n = 5$, ELA: 8.9 ± 1.3; $n = 5$; 2way RM ANOVA ELA vs. region interaction $F(1,8)=28.2$, $p = 0.001$; Sidak's post hoc, control vs. ELA, $p = 0.0004$). No effects of ELA were observed in the dorsal raphe nucleus, or on the total number of 5-HT neurons, suggesting that ELA causes dysfunction in MnR 5-HT neurons independent of total neuron number. Additionally, we found that 5-HT levels in the vDG were decreased in ELA-exposed animals at P56 (Control: 232.1 ± 35.6 $n = 6$; ELA: 112.2 ± 13.9 $n = 6$; unpaired t test, $p = 0.01$) but not at P35, suggesting an impairment of 5-HT modulation of the vDG that manifests in adulthood.

Conclusions: Our results show that ELA causes vDG hyperactivity and fear overgeneralization in adult female mice, and that increasing 5-HT levels from birth can rescue these neurobiological and behavioral deficits. Understanding how 5-HT regulation of the vDG mediates ELA-induced behavioral impairments may provide new potential treatment options for psychiatric disorders that have their origin early in life.

Keywords: Early Life Adversity, Serotonin, Dentate Gyrus, Early Life Stress, Neural Circuits

Disclosure: Nothing to disclose.

P380. Frequency-Dependent Entrainment of Brain Networks Using Transcranial Magnetic Stimulation

Juliana Corlier*, Andrew Wilson, Cole Citrenbaum, Andrew Leuchter

University of California - Los Angeles, Los Angeles, California, United States

Background: Transcranial Magnetic Stimulation (TMS) is an effective treatment for several psychiatric and neurological conditions such as depression, chronic pain, obsessive compulsive disorder, smoking cessation and others. While the therapeutic efficacy is hypothesized to arise from the effects on widespread networks beyond the stimulation site, the role of the stimulation frequency is not clear. In the past, various frequencies have been successfully used for therapeutic purposes, including 1, 5, 10, 18, 20 Hz as well as theta-burst stimulation. It remains to be elucidated how the elicited patterns of activation vary by frequency. Additionally, it is unclear if brain circuits have resonant frequencies (RF) to which the networks respond in a preferential manner and if these frequencies differ between individuals.

We hypothesized that different frequencies would elicit spatially distinct patterns of electroencephalographic (EEG) activity that would reflect how different brain circuits respond to stimulation. We have also hypothesized that preferred RFs vary across individuals. We are using a newly developed TMS-EEG interrogation paradigm to evaluate TMS-evoked spectral changes and the entrainment of oscillations in the source-localized EEG signal and the identification of RF across individuals.

Methods: 80 subjects with Major Depressive Disorder (MDD) underwent a TMS interrogation at 71 frequencies ranging from 3-17 Hz (in steps of 0.2 Hz) administered to left dorsolateral prefrontal cortex (DLPFC). Each stimulation frequency consisted of 40 pulses with 26 s off period in between frequencies. Frequency order was randomized for all subjects.

64-channel EEG was recorded at baseline, during the entire interrogation procedure and after the session. We used a custom developed artifact removal pipeline to remove TMS-pulse artifact, decay artifacts and conventional EEG artifacts (electrical noise, eye, muscle, cardiac artifacts) following each TMS train. The period [-1000 0 ms] pre stimulation and [0-1000 ms] post every frequency was extracted and used for source localization using Brainstorm. We created a head model with the Boundary Element Method (BEM) using digitized electrode locations and the OpenMEEG toolbox. The source time series were reconstructed using the Minimum Norm Estimation (MNE) method to obtain the inverse solution, which addresses the ill-posed nature of EEG source estimation through regularization. For each frequency, we obtained an average cortical source activation map for each frequency band. Only circuit activation greater than z-scores $> =$ 2.5 compared to baseline activity are displayed.

Results: We present average source-localized activation maps (Figure 1) as well as individual subject examples (Figure 2) for all

71 stimulation frequencies in the broadband and the bandpass filtered frequencies of delta [1-4 Hz], theta [4-8 Hz], alpha [8-12 Hz] beta [12-20 Hz] and low gamma [20-30 Hz]. Different TMS stimulation frequencies elicit widespread cortical activations well beyond the stimulation site. The spatial distribution of the elicited patterns varies substantially between individuals based on the stimulation frequency.

Conclusions: We demonstrate that different TMS stimulation frequencies elicit largely different spatial activation patterns in the source localized EEG signal and that these activation maps vary as a function of frequency and across individuals. These findings have clinical implications suggesting that a personalized treatment frequency should be used to enhance the entrainment of target brain circuits.

Keywords: Brain Stimulation, TMS-EEG, Frequency Optimization

Disclosure: Nothing to disclose.

P381. Distinct Populations of Ventral Pallidal Cholinergic Neurons Encode Valence of Olfactory Stimuli

Ronald Kim*, Mala Ananth, Niraj Desai, Lorna Role, David Talmage

National Institutes of Health, Bethesda, Maryland, United States

Background: Basal forebrain cholinergic neurons (BFCNs) play a critical role in a wide array of behaviors. In addition to an established role of BFCNs in learning and memory, recent evidence demonstrates BFCNs encode innate behaviors. For example, we have shown that mice display approach to an appetitive odor (2-phenylethanol) and avoidance to an aversive odor (predator urine). These innate behaviors are associated with an increase in the number of activated cholinergic neurons in the ventral pallidum (VP). The goal of the present studies is to expand on these findings, and further examine the functional significance of VP cholinergic neurons.

Methods: To better understand how VP cholinergic neurons respond to an appetitive vs. aversive odor, fiber photometry was used to characterize in-vivo calcium activity. Chat-cre mice were injected with AAV-syn-Flex-GCaMP6F and received a fiber optic implant targeting the VP. Following habituation, calcium activity of VP cholinergic neurons was assessed in response to either the appetitive or aversive odor one day, and the opposite odor the following day. To test whether same or distinct subsets of VP cholinergic neurons are activated in response to each odor, Chat-cre x cFos-tTA/GFP mice were injected with an activity- and cre-dependent viral vector in the VP (ADCD-hM4Di). This strategy utilizes a Tet-Off system, where in the absence of a doxycycline (DOX) diet, activated cholinergic neurons are permanently labeled with mCherry. cFos-GFP can be used to label activated neurons in a distinct context. Mice underwent behavioral testing across 3 days, Day 0 = habituation (DOX-on), Day 1 = odor exposure 1 or saline (DOX-off, activated VP cholinergic neurons labeled with mCherry), Day 2 = odor exposure 2 or saline (DOX-on, immunohistochemistry for GFP and Chat). To examine the role of VP cholinergic neurons in innate behavioral responses to odor exposure, Chat-cre mice were injected with syn-DIO-hM4Di. Fifteen minutes following an IP injection of a subthreshold dose of clozapine, preference to either the appetitive or aversive odor was assessed in a Y-maze. For selective chemogenetic inhibition of previously activated VP cholinergic neurons, Chat-cre x cFos-tTa/GFP mice were injected with ADCD-hM4di. Following habituation, mice were taken off a DOX diet and exposed to either the appetitive or aversive odor (activated VP cholinergic neurons labeled with mCherry). Twenty-four hours later, mice were injected with clozapine to inhibit the previously activated VP cholinergic neurons, and preference to the same odor was assessed in a Y-maze.

Results: Preliminary fiber photometry results show that VP cholinergic neurons demonstrate reliable, time-locked increases in calcium activity in response to both the appetitive and aversive odor. We also examined if activated cholinergic neurons in the VP were the same or distinct neurons following exposure to appetitive vs. aversive odor. When exposed to the same odor on days 1 and 2, mice exhibited a significant increase in colocalization of mCherry (activated VP cholinergic neurons on day 1) and GFP (activated VP cholinergic neurons on day 2). This indicates re-activation of previously activated VP cholinergic neurons when exposed to the same odor. However, when mice are exposed to a distinct odor on day 2, no colocalization of mCherry and GFP were observed. These results are consistent with there being two distinct subpopulations of VP cholinergic neurons: one population activated in response to an appetitive odor and a second, distinct population activated upon exposure to an aversive odor. Our chemogenetic inhibition experiments with syn-DIO-hM4Di revealed that general inhibition of all subpopulations of VP cholinergic neurons abolished normal approach to the appetitive odor. Mice spent more time in the saline paired arm, signifying the appetitive odor was now aversive. In contrast to the VP cholinergic block of approach behavior, avoidance to the aversive odor was still observed. Similar results were observed when specifically targeting VP cholinergic neurons that were previously activated. Selective inhibition of VP cholinergic neurons that were previously activated in response to the appetitive odor abolished approach behavior and led to avoidance of the appetitive odor during the preference test. Targeted inhibition of VP cholinergic neurons that were previously activated after exposure to the aversive odor resulted in normal avoidance to the aversive odor.

Conclusions: The results from the present studies reveal (1) increases in in-vivo calcium activity of VP cholinergic neurons in response to both an appetitive and aversive odor; (2) the VP contains two distinct and non-overlapping subpopulations of cholinergic neurons which are uniquely engaged by either appetitive or aversive stimuli; and (3) chemogenetic inhibition (either general inhibition or selective inhibition of previously activated neurons) of VP cholinergic neurons alters innate behavioral responses to appetitive but not aversive odor. In ongoing studies, we are exploring how these subpopulations of VP cholinergic neurons differ, with a focus on mapping their projections and measuring their baseline electrical properties.

Keywords: Acetylcholine, Ventral Pallidum, Valence

Disclosure: Nothing to disclose.

P382. Differential Vulnerabilities to Acute and Chronic Variable Stress in Mice Carrying a Parkinson's Disease-Linked LRRK2-G2019S Mutation

Christopher Guevara*, Kumayl Alloo, Romario Thomas, Alexander Tielemans, Kyomi Blake, Jamal Magoti, Swati Gupta, Alexandra Magee, Deanna Benson, George Huntley

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

Background: Parkinson's disease (PD) is associated with psychiatric non-motor symptoms such as depression and anxiety that emerge early, appear to be independent of dopamine neuron loss, and are poorly understood. The G2019S mutation in the Lrrk2 gene is one of the most commonly associated PD risk gene mutations found in both sporadic and familiar late-onset PD. Risk for both PD and depression is increased by stress, and previous work in the lab has shown that young adult Lrrk2-G2019S knockin mice display behavioral, synaptic and non-synaptic plasticity

adaptations to social stress that differ significantly from those of wildtype mice.

Methods: In order to determine whether social stress-effects are generalized to other forms and magnitudes of behavioral stress, we subjected young adult wildtype (WT) and G2019S knock-in (GS) mice to a standard, daily variable stress (VS) paradigm consisting of one, 1-hr stressor per day over three days in the following order: 100 mild foot shocks, tail suspension and restraint. This stress paradigm was applied for 6 days (6d-VS) or 28 days (28d-VS). Following the last stressor, home-cage unstressed controls and stressed mice underwent a battery of assays (open field test, social interaction test, and novelty suppressed feeding) to probe for stress-induced behavioral changes.

Results: In WT mice, we found no major changes in behavioral assays after 6d-VS, but significant stress-effects in post-stress behavioral assays emerged at 28d-VS, as expected from prior work. In contrast, GS mice already displayed significant stress susceptibility after 6d-VS which persisted after 28d-VS. These behavioral adaptations are driven specifically by stressful experiences, as no differences were observed between genotypes in unstressed control conditions.

Conclusions: These data show that the G2019S mutation lowers the threshold for stress susceptibility broadly across stress paradigms, mounting a temporally evolving set of neural and behavioral adaptations that differ from WT. Such differential vulnerabilities may impact the onset of psychiatric symptoms in human PD patients. Future studies are probing how different brain regions are impacted at a cellular and synaptic level by the differing durations of stress. Understanding these interactions will provide insight into the neural adaptations of individuals harboring the G2019S mutation, revealing novel targets for ameliorating mood-related symptoms associated with PD.

Keywords: Acute and Chronic Stress, Synapses, Glutamate, Parkinson's Disease, Depression

Disclosure: Nothing to disclose.

P383. Contribution of Adult Born Granule Cells to the Mechanism of Action of Electroconvulsive Therapy

Julia Castello Saval*, Victor Luna, Hannah Chung, Rene Hen

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

Background: Major depressive disorder is the leading cause of disability worldwide and about one third of patients do not respond to pharmacotherapy. In cases of treatment-resistant depression, electroconvulsive therapy (ECT) is an effective alternative with a fast onset of action. It is known that ECT increases neurogenesis in the dentate gyrus (DG) of the hippocampus, a brain region highly vulnerable to stress and thought to play an important role in the development of depression. In this study, we investigate which is the contribution of adult born granule cells (abGCs) to the mechanism of action of ECT.

Methods: Electroconvulsive stimulation was delivered on alternate days for 10 sessions. Mice were anesthetized with isoflurane and electrical stimulus were delivered through ear clipped electrodes (120 Hz, 0.5 mA, 1 sec). Behavior was assessed one week after the last session in the forced swim test and the novelty suppressed feeding test (males, $n = 9$-10 per group). In the forced swim test, mice were placed in a container with water (25°C) and allowed to freely swim. Immobility time was quantified using Videotrack. In the Novelty suppressed feeding test mice were starved for 19 hours and placed in a novel, bright arena with a food pellet in the center, the latency to eat the food pellet was measured. For hippocampal irradiation after anesthesia, mice were placed in a stereotaxic frame, and cranial irradiation applied using a lead shield for focal Xray. A cumulative dose of 5 Gy was given over the course of 3 sessions. Results between groups were compared using one-way ANOVA for statistical group comparisons or multiple t-tests when appropriate.

For fluorescence images, animals (males, $n = 5$ per group) were perfused and brains were sectioned. Sections underwent washing steps, antigen retrieval with sodium citrate buffer and blocking step with 10% normal donkey serum. Slices were incubated overnight at 4°C in primary antibody (rabbit anti-doublecortin, 1:400, rabbit cFos, 1:400, Synaptic Systems). The next day, sections were washed and incubated in secondary antibody (anti-rabbit Alexa Fluor 488). Sections were prepared for confocal imaging.

Slice electrophysiology was performed with a Nestin-CreERT2 line crossed with a Channel Rhodopsin line to drive expression of Channel rhodopsin in adult born neurons. Tamoxifen was injected 6 weeks before whole cell current clamp recordings to induce Cre recombinase. Whole-cell recordings (-70 mV) were obtained using a patch pipette (4.5-6.5 M). For optogenetic stimulation of abGCs, light pulses were delivered through a 40x objective directly into brain slices.

Results: Electroconvulsive stimulation, the mouse model of ECT, rescues the depressive-like phenotype of mice administered chronically with corticosterone (CORT) in the novelty suppressed feeding test an anxiety related test. Vehicle/Sham 100.4 ± 48.61; Vehicle/ECS 113.5 ± 65.8 unpaired student's t-test: $p = 0.6325$. CORT/Sham 272.3 ± 33.76; CORT/ECS 175.7 ± 41.58 unpaired student's t-test: $p = 0.0458$). In the forced swim test, a stress coping test, ECS rescued the depressive-like phenotype in the Vehicle group and the CORT group. Vehicle/Sham 124.0 ± 11.02; Vehicle/ECS 80.86 ± 27.92 one-way ANOVA multiple comparisons test: $p = 0.006683$. CORT/Sham 126 ± 17.41; CORT/ECS 87.46 ± 11.04 $p = 0.005860$.

X-Ray ablation of abGCs renders mice unresponsive to ECS novelty suppressed feeding and forced swim test. Sham/Sham 272.3 ± 33.76; Sham/ECS 164.1 ± 38.95 unpaired t-test: $p = 0.0501$; XRay/Sham 309.1 ± 29.15; XRay/ECS 318.2 ± 28.24 one-way ANOVA multiple comparisons test: $p = 0.8283$.

ECS stimulates the production of abGCs as shown by increased levels of doublecortin (DCX), a marker for young neurons. In addition, using transgenic mice expressing YFP-Synaptophysin in the DG, we visualized presynaptic boutons. After ECS, there is an increase in the number of puncta in the granule cell layer, suggesting increased presynaptic terminal density In contrast, when abGCs are ablated, the ECS-induced increase in synaptic boutons is comparable to Sham treated animals Sham 0.922 ± 0.03984; ECS 1.240 ± 0.03745 XRay 0.3468 ± 0.03327 One way ANOVA multiple comparisons test $p < 0.001$.

Intrigued by this finding, we used a Nestin-CreERT2 mouse line crossed with a Channelrhodopsin-2(ChR2)-EYFP floxed mouse line to express ChR2 in abGCs. This allowed whole cell clamp recordings in mGCs after optogenetic stimulation of abGCs. When comparing mice that received 10 sessions of ECS with Sham animals, we found that stimulation of abGCs induced an inhibitory current in mGCs that is revealed after pharmacological suppression of the excitatory current component using the AMPA antagonist, NQBX and the NMDA antagonist, APV. Furthermore, addition of the mGluR2/3 antagonist APICA suppressed the inhibitory current in mGCs

All results shown as Mean±S.E.M

Conclusions: To integrate these results, we propose that ECS induces neurogenesis and axonal sprouting from abGCs which results in an increase in the number of presynaptic boutons from abGCs in the granule cell layer which in turn results in increased activation of mGluR2 receptors on mGCs. The resulting decrease in DG activity may be responsible for the antidepressant-like effects of ECS.

Keywords: Electroconvulsive Therapy, Adult Hippocampal Neurogenesis, Major Depressive Disorder, Dentate Gyrus

Disclosure: Nothing to disclose.

P384. VTA-NAc Neural Activity Underlying Chronic Stress-Induced Reward-Seeking Deficits

Lucille Johnston, Amanda Amilcar, Rachel Ding, Alexander Harris*

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

Background: Anhedonia, defined as reduction in the pursuit of pleasure, is one of the core symptoms of depression. Stress, a risk factor for depression, decreases reward seeking in both humans and mice. We recently demonstrated that ventral tegmental area (VTA)-nucleus accumbens (NAc) activity underlies the reduction in anticipation seen after a single episode of restraint stress (Lowes et al., Nature Communications 2021). However, the precise nature of the behavioral changes (e.g., 'wanting' vs 'liking') that underlie deficits in reward seeking after chronic stress remain unknown. Moreover, the impact of chronic stress on reward circuit activity remains controversial, with studies reporting both increases and decreases in VTA dopamine firing rates (Tye et al. Nature 2013; Chaudhury et al. Nature 2013). In this study, we investigate VTA and NAc firing rates during reward seeking after mice have undergone chronic stress.

Methods: We implanted chronic electrodes simultaneously in VTA and NAc of male and female mice to record both local field potential (LFP) and single unit activity (C57BL/6 J 3 males, 4 females). We trained the mice to associate a tone with reward availability (CS +) and another tone with no condensed milk reward (CS-) to a criterion of 70 percent correct responses to the CS + for two consecutive days. We then recorded neural activity as mice performed the task. The mice then underwent 10 days of chronic social defeat stress (CSDS) and 24 hours after the final day of defeat, performed a social interaction task and the reward task. We analyzed the data using custom MATLAB script and used Pearson's correlation to correlate the impact of stress on social interaction and reward seeking.

Results: We found that chronic stress produced reductions in anticipatory and post-consumption lick rates that were sustained for over 24 hours and correlated with SI ratio ($N = 7$, $r = 0.79$ for anticipation, $r = 0.87$ for consumption, $p < 0.05$). We also are sorting neurons into their putative identities (e.g., VTA dopaminergic, VTA GABAergic, NAc MSN, etc) and analyzing the lick- and cue-evoked neural activity and synchrony between regions to determine the neural activity associated with these reductions in reward seeking.

Conclusions: Together, these data suggest that chronic stress induces longer-lasting reward processing deficits and allows for the dissection of the contribution of individual cell populations to anhedonia.

Keywords: Reward, Neural Circuits, Chronic Stress

Disclosure: Genetika: Advisory Board (Self)

P385. Somatostatin Peptide Signaling Dampens Cortical Circuits and Promotes Exploratory Behavior

Nicole Crowley*

Penn State University, University Park, Pennsylvania, United States

Background: Somatostatin (SST) neurons in the prelimbic (PL) cortex mediate a variety of behavioral states, ranging from alcohol consumption to fear learning and avoidance-related behaviors. However, little is known about the role of somatostatin peptide signaling itself to cortical functioning and behavior. Here, we sought to characterize the unique physiological and behavioral roles of the SST peptide in the PL cortex.

Methods: We employed a combination of ex vivo electrophysiology, in vivo calcium monitoring, and in vivo peptide pharmacology to explore the role of SST neuron and peptide signaling in the mouse PL cortex. Whole-cell slice electrophysiology was conducted in C57BL/6 J male and female mice in pyramidal and GABAergic neurons of the PL cortex to characterize the pharmacological mechanism of SST signaling. Fiber photometry recordings of GCaMP6f fluorescent calcium signals from SST neurons were conducted to characterize the activity profile of SST neurons during exploration of an elevated plus maze (EPM) and open field (OF). We further used local delivery of a broad SST receptor (SSTR) agonist into bilateral PL cortex to test causal effects of SST signaling on these same exploratory behaviors.

Results: SSTR activation broadly hyperpolarized layer 2/3 pyramidal neurons in the PL cortex in both male and female mice ex vivo, through both monosynaptic and polysynaptic GABA neuron-mediated mechanisms of action. This included reductions in the resting membrane potential in females ($t13 = 2.205$, $p = 0.0460$) and males ($t16 = 2.889$, $p = 0.0107$) of pyramidal neurons – an effect that was greater in the presence of TTX in both females ($t6 = 5.095$, $p = 0.0022$) and males ($t4 = 3.448$, $p = 0.0261$). Multiple other measurements of excitability were also significantly altered in both cases. Hyperpolarization was blocked by pre-application of the SSTR antagonist cyclo-somatostatin (cyclo-SST) and was non-reversible. SST neurons in PL were activated during EPM and OF exploration, indicating task-related recruitment of these neurons. Specifically, SST neurons were more active while mice were in the open arms as compared to the closed arms of the elevated plus maze ($t10 = 6.101$ $p < 0.001$), and were more active while mice were in the center as compared to the edges of the open field ($t10 = 2.797$, $p = 0.0189$). No sex differences were seen in the fiber photometry experiments. Lastly, in line with this exploration-related activity profile, SSTR agonist administration directly into the PL enhanced open arm exploration in the elevated plus maze of male mice, with no effect in female mice (2-way ANOVA; $Fsex(1,26) = 0.6452$, $p = 0.4291$; $Fdrug(1,26) = 3.462$, $p = 0.0741$, Fsex x drug $(1,26) = 7.868$, $p = 0.0094$). We also saw a significant increase in the number of dead dips over the open arms of the elevated plus maze in male mice (2-way ANOVA; $Fsex(1,26) = 5.917$, $p = 0.0222$; $Fdrug(1,26) = 3.264$, $p = 0.0824$, Fsex x drug $(1,26) = 5.614$, $p = 0.0255$).

Conclusions: Here we reveal a novel role for the SST peptide system within the PL cortex, by demonstrating a peptide-induced hypoexcitability of PL circuits and modulation of PL-dependent exploratory behaviors.

Keywords: Somatostatin, Prelimbic Cortex, Slice Electrophysiology, Fiber Photometry

Disclosure: Nothing to disclose.

P386. Changes in Emotion Regulation-Relevant Neurocircuitry Following Standard RTMS for Treatment Resistant Depression

Kristen Ellard*, Walker Pederson, Samadrita R Chowdhury, Tracy Barbour, Joan A Camprodon

Harvard Medical School, Boston, Massachusetts, United States

Background: Transcranial magnetic stimulation (TMS) of the dorsolateral prefrontal cortex (DLPFC) is an FDA-approved standard intervention for treatment resistant depression. However, the mechanisms by which TMS of the DLPFC relates to treatment response remains poorly understood. Examining the

mechanisms of TMS is crucial towards improving treatment outcomes, which currently show high response but low remission rates (~30%). Deficits in emotion regulation (ER) have been repeatedly demonstrated in depression, linked to symptom severity and treatment response. Healthy ER includes detection of salience, the initiation of regulation, and cognitive control in accordance with internal and external contextual demands, processes that require adaptive coordination between neurocircuits comprising the salience, frontoparietal control, and default mode functional networks. Here, we examined the effects of a course of repetitive (r) TMS to the left DLPFC on resting-state functional connectivity across the broader neurocircuitry supporting ER, spanning canonical functional networks.

Methods: Based upon existing literature (e.g. Kohn et al., 2014; McTeague et al, 2020), an analysis of functional connectivity between individually-derived DLPFC target ROIs and a priori selected ER-related regions of interest (ROIs) was conducted on data from 24 depressed patients (50% female, mean age 39.91 ± 3.04, 92% Caucasian) receiving an interventional course (36 sessions, 120% MT, 10hz rTMS, 3,000 pulses) of left DLPFC rTMS. Selected ROIs comprised regions within the limbic (LIMB) salience/ventral attention (SalVA), dorsal attention (DA), frontoparietal control (FPC) and default mode networks (DMN). Significant pre-post TMS changes in DLPFC target - ER ROI functional connectivity were followed up with linear regressions to elucidate the relationship between ER functional neurocircuit changes and changes in ER-relevant clinical measures in responders (Resp; $n = 10$) versus non-responders (Non-Resp; $n = 14$). To understand TMS-related changes within the broader ER-related neurocircuitry, ROIs showing significant pre-post TMS functional connectivity with the DLPFC target and demonstrating a significant relationship to changes in ER-related clinical response were further characterized using graph metrics of centrality, based upon whole-brain functional connectivity.

Results: Significant pre-post TMS changes in functional connectivity were found between the DLPFC target and regions in the LIMB (l. amygdala, Resp: $t = -1.82$, $p = .05$, Non-Resp ns; r. hippocampus, Resp: -2.07, $p = .03$, Non-Reps ns), SalVA (r. anterior insula, Resp: $t = -2.11$, $p = .03$, Non-Resp ns; r. VLPFC, Resp: $t = -1.12$, $p = .10$, Non-Resp: $t = 2.08$, $p = .02$), and DMN networks precuneus/posterior cingulate, Resp: $t = 2.12$, $p = .03$, Non-Resp ns). Changes in target-ER ROI functional connectivity were significantly associated with improvements in ER measures (delta DLPFC-amygdala/delta negative affect, $z = -1.96$, $p = .05$; delta DLPFC-hippocampus/delta affective control, $z = -2.50$, $p = .02$; delta DLPFC-anterior insula/delta negative affect, $z = -2.45$, $p = .01$; delta DLPFC-VLPFC/delta affective control, $z = 2.89$, $p = .004$; delta DLPFC-precuneus, PCC/delta affective control, $z = 2.15$, $p = .03$). Follow-up graph analysis identified significant pre-post TMS increases in betweenness-centrality (a measure of the amount of influence a region has on the flow of information between regions and networks), in the right VLPFC/anterior insula ($p = .03$). Significant changes in functional connectivity between this node and the broader ER neurocircuitry were found following TMS (all p's < .05), with the strongest effect between VLPFC/anterior insula and DMN functional connectivity (precuneus/PCC, $t = -2.63$, $p = .007$). A significant relationship was found between these functional connectivity changes and measures of negative affect ($t = 2.00$, $p = .05$) and rumination ($t = 2.01$, $p = .05$).

Conclusions: Treatment with TMS applied to individualized DLPFC targets resulted in broad changes in functional connectivity between the DLPFC target site and regions of the limbic, salience and default mode networks in treatment responders. These changes were significantly related to changes in ER relevant clinical measures. The right VLPFC/anterior insula showed an increased influence on the broader emotion regulation neurocircuitry as measured by betweenness centrality, and TMS-related changes in functional connectivity between this region and regions of the DMN were significantly related to changes in negative affect and rumination, two core features of emotion dysregulation. These results suggest more precise targeting of emotion regulation related neurocircuitry with TMS may be one avenue towards increasing overall response rates in treatment resistant depression.

Keywords: Repetitive Transcranial Magnetic Stimulation (rTMS), Emotional Regulation, Depression

Disclosure: Nothing to disclose.

P387. Sex Moderates the Relationship Between Functional Connectivity and Remission in Late-Life Depression

Andrew Gerlach*, James Wilson, Helmet Karim, Howard Aizenstein, Carmen Andreescu

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: Major depressive disorder accounts for more disability-adjusted life years than any other mental illness, yet the predominant treatment approach is essentially trial-and-error. This resulting delays in recovery exacerbate the personal, social, and even economic costs. This problem is amplified in late-life depression (LLD), where the disorder presents with greater heterogeneity, longer time to response, lower remission rates, and higher risk of relapse. Thus, biomarkers of early treatment response in LLD to improve response time and outcome are a major strategic goal of neuroimaging research. However, the role of biological sex has not been adequately studied in assessment of MRI-based biomarkers.

Methods: Demographic information, baseline depression severity, and resting state fMRI scans at baseline and after commencing antidepressant treatment (day 1) were collected in two LLD studies (NEMO [R01 MH076079-15]: $n = 28$, Circuits2 [R01 MH076079-09]: $n = 51$). Remission was defined as a final MADRS score of 10 or less for at least 2 weeks and subject to blinded clinician assessment. The Shen50 atlas was used to calculate region-to-region functional connectivity (FC) for 82 non-cerebellar regions (excluded due to poor coverage). Differential connectivity (DC) was calculated by subtracting the baseline FC from the day 1 FC. The role of biological sex was assessed in both explanatory and predictive frameworks. For the explanatory framework, sex-based differences in remitters and non-remitters were assessed using two-sample t-tests on the DC for each pair of regions. For the predictive framework, a random forest classifier was used to predict the remission status of each participant. Models were fit and tested separately using all participants, only females, and only males. Monte Carlo cross-validation was used to evaluate the predictive performance of clinical and DC covariates to predict remission status in seven different model specifications for each study. Variable importance was evaluated using the Gini importance measure assessed through the trained random forest models. The average accuracy and area under the receiver operating characteristic curve metrics were used to assess the predictive performance of each model.

Results: The location and direction of differences in DC among remitters and non-remitters, as well as the brain regions important to the prediction of remission substantially differed by sex. Prediction of remission was significantly improved by fitting separate models for males and females. Further, males showed stronger group DC differences between remitters and non-remitters than females, with large differences in the bilateral caudate, bilateral temporal pole, and left postcentral gyrus. These regions were also important for prediction in male-only models. Females, on the other hand, showed weaker group differences in DC between remitters and non-remitters, but a much more diverse set of regions important for prediction. Connectivity of the

left caudate nucleus was the leading predictor of remission for both males and females.

Conclusions: Early FC indicators of antidepressant treatment response in LLD may differ fundamentally between males and females. Simply including sex as a covariate in the model is insufficient to capture these difference, as there is a clear moderating effect of sex. Separate models for males and females or sex by region interaction terms may be necessary to provide sufficient sensitivity and specificity.

Keywords: Late Life Depression, Resting State Functional Connectivity, Antidepressant Response, Predictive Models, Sex Differences

Disclosure: Nothing to disclose.

P388. Utilizing the Systemic Immune-Inflammation Index and Inflammatory Cytokines in Predicting Treatment Responsiveness Amongst Patients With Treatment Resistant Bipolar Depression

Kyle Decker*, Nausheen Baig, Stephen Murata, Angelos Halaris, Jawed Fareed, Debra Hoppenstaed

Loyola University School of Medicine, Saint Charles, Illinois, United States

Background: Inflammation is associated with depressive illness and treatment resistance. These advances have prompted a search for predictive biomarkers of treatment response. The Systemic Immune-Inflammation Index (SII) has shown promise as a prognostic indicator for solid malignancies and cardiovascular disease while its use in psychiatric conditions remains underexplored. In a recent randomized, double-blind, placebo-controlled trial we found that combined treatment with celecoxib (CXB) and escitalopram (ESC) was more effective in reversing treatment resistance and augmenting antidepressant response in treatment-resistant bipolar depression (TRBDD) compared to ESC plus placebo. Our follow-up analysis characterizes treatment response in relation to SII, as well as inflammatory and kynurenine pathway (KP) biomarkers.

Methods: The sample (N = 69) included 65.2% female, 65.2% white, mean age 42 years (SD = 12.7). The study included healthy controls (n = 32) and TRBDD subjects (n = 47). The TRBDD group consisted of an ESC + CBX arm (n = 26) and ESC + PBO arm (n = 21). SII was calculated from the complete blood count with differential (SII = platelets x neutrophils/lymphocytes) at baseline and completion (8 weeks). Plasma inflammatory and KP markers levels were also obtained at baseline and week 8. Depressive symptom severity (main outcome) was measured both continuously (HAMD17 total score) and dichotomously (treatment remission, defined as HAMD17 total score < 7 by week 8). Statistical analysis was conducted using R-3.6.3.

Results: Group comparison revealed no significant differences in SII by treatment arm at baseline or remission status by week 8. SII at baseline was trended with an elevated inflammatory cytokine profile including higher IL-2 at baseline ($p = 0.073$), IL-1B at baseline ($p = 0.051$), hsCRP at week 8 ($p = 0.08$), and lower anti-inflammatory IL-4 ($p = 0.086$). Baseline SII significantly correlated with lower baseline vascular endothelial growth factor (VEGF) ($p = 0.029$). On multivariate linear modeling (R2/ R2 adjusted = 0.49/0.41) HAMD17 at week 8 (outcome) was significantly associated with ESC + CBX treatment ($p < 0.008$), an interaction with age and SII-baseline (Beta-estimate = 0.001, (95% CI [< 0.001, 0.001]), $p < 0.001$, Cohen's D = 0.5). In a separate model using individual cell counts (R2 /R2 adjusted = 0.413/0.334), HAMD17 by week 8 was associated with baseline neutrophil count amongst older patients (b-estimate = 0.17 (95% CI [0.06, 0.27]), $p < 0.003$, Cohen's D = 0.48), but not baseline platelet or lymphocyte counts ($p = 0.312$, $p = 0.201$, respectively)

Conclusions: On univariate analysis, baseline SII trended with elevated baseline proinflammatory markers (IL-2, IL-1B, hsCRP) and lower baseline anti-inflammatory markers (IL4). SII significantly correlated with lower vascular endothelial growth factor (VEGF) which is associated with neuroprotection and was previously shown to be elevated in our TRBDD cohort compared to healthy controls. In older patients, lower pretreatment SII predicted lower depressive severity by week 8 irrespective of treatment arm, and this association appeared driven largely by the neutrophil component of SII. Taken together, SII appears to be a candidate poor prognostic indicator in TRBDD. Future studies with a larger sample sizes should further investigate the potential clinical utility of SII as it is readily available and accessible through the routinely drawn blood counts.

Keywords: Bipolar Depression, Treatment Resistance, Inflammation

Disclosure: Nothing to disclose.

P389. Assessing the Generalizability and Stability of Biologically-Based Subtypes of Depression

Katharine Dunlop*, Logan Grosenick, Jonathan Downar, Fidel Vila-Rodriguez, Faith Gunning, Zafiris Daskalakis, Daniel M. Blumberger, Conor Liston

University of Toronto, Toronto, Canada

Background: Major depressive disorder (MDD) is associated with considerable symptom variability; a comprehensive understanding of this variability may lead to individualized intervention approaches and therefore improved treatment response rates. Recent efforts by our group and others have sought to understand symptom heterogeneity in MDD using functional neuroimaging. Although these recent studies mark significant progress in understanding heterogeneity in MDD, replication and validation of these results is critical given the limitations imposed by sample size and depth of clinical characterization. To address these limitations, this study has three aims. First, we sought to understand the neurobiological basis of MDD symptom heterogeneity by extending our earlier work defining robust and reproducible brain-behavior dimensions to new data. An L2-norm regularized multivariate model was generated using a large MDD dataset recruited from a single site and incorporated additional items assessing anhedonia and anxiety symptoms. Second, we tested for the existence of MDD subtypes and evaluated their stability and reproducibility. Third, we characterized these MDD subtypes regarding atypical resting-state functional connectivity (RSFC), clinical symptoms, and antidepressant response to non-invasive brain stimulation.

Methods: L2-regularized canonical correlation analysis (RCCA) was evaluated in a large, single-site MDD dataset (n = 328, 215 female (65.6%); mean age=40.35 ± 12.05 SD) using RSFC and clinical symptomatology. First, to optimize three RCCA hyperparameters, we performed a nested grid search (with training, validation, and test splits); the optimal hyperparameter combination was defined as the highest median canonical correlation in held-out validation data for the first dimension. Next, we examined the stability and hold-out performance of dimensions (on held-out test data not used for training or validation) and tested for significant dimensions using random permutation testing. Afterwards, we generated a final optimized RCCA model and evaluated the performance and stability of hierarchical clustering. Upon identifying the optimal clustering solution, we characterized latent variables representing co-occurring RSFC and symptomatology, and symptom/RSFC differences by subtype.

Lastly, we identified subtype differences in repetitive transcranial magnetic stimulation response and remission rates.

Results: The performance and stability of the first three RCCA dimensions were significant ($p < 0.05$, random permutation test). These three dimensions represented: depressed mood, and thalamic and default mode RSFC; anhedonia, and cingulo-opercular and higher-order visual and network RSFC; and insomnia, and sensorimotor and posterior insula RSFC, among other connectivity features. Hierarchical clustering identified four significant depression subtypes ($p < 0.05$, random permutation test), each with distinct clinical symptom profiles, abnormal RSFC patterns, and responsivity to repetitive transcranial magnetic stimulation (rTMS) over the dorsomedial or dorsolateral prefrontal cortex. Subtypes with lower anhedonic symptoms were most responsive to rTMS. Subtypes did not differ by age or sex.

Conclusions: In an extension of our previous work, we sought to characterize regularized CCA and clustering performance in a large, single-site MDD dataset. RCCA yielded three significant, stable and generalizable brain-behavior dimensions that resembled well-documented MDD symptom-brain associations, and four categorical subtypes. Both categorical and dimensional approaches to parsing heterogeneity may be beneficial in different contexts. We note several study design choices that may affect RCCA models, including participant inclusion/exclusion criteria, medication use, and choice of symptom severity measures. Taken together, these results represent an important step forward in assessing data-driven subtyping methods and provide evidence that RCCA is an effective tool to identify stable and generalizable associations between RSFC and behavior.

Keywords: Resting State Functional Connectivity, Depression Subtypes, Transcranial Magnetic Stimulation, Canonical Correlation Analysis (CCA)

Disclosure: Nothing to disclose.

P390. Probing Dopaminergic Deficits in Adolescent Depression

David Pagliaccio*, Emily Zhang, Alma Bitran, Kenneth Wengler, Guillermo Horga, Randy Auerbach

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

Background: Depression is a chronic and impairing mental health condition that often peaks during adolescence. Prior work implicates alterations within the RDoC Positive Valence Systems in adolescent depression, including anhedonia and blunted striatal reward response. These alterations generally rely on dopaminergic projections from the substantia nigra and ventral tegmental area in the midbrain, to striatal and prefrontal circuits, respectively. Yet, the hypothesized role of dopamine is largely based on animal, pharmacological, post-mortem human, and adult studies utilizing methods too invasive for pediatric research (e.g., lumbar puncture, positron emission tomography). Recently, a safe and non-invasive alternative means of characterizing midbrain dopamine has been developed using magnetic resonance imaging (MRI) to assess neuromelanin, a key byproduct of dopamine metabolism.

Methods: As part of ongoing data collection, we are collecting high-resolution neuromelanin-MRI data from adolescents (13-18 years old; Target $N = 60$; no exclusions based on sex or gender), primarily with a history of major depressive disorder. Adolescent complete clinical assessment, self-reports, and smartphone-based data collection over the subsequent months. Preliminary analyses entail linear regression models among $N = 29$ adolescents. All analyses controlled for age, sex, and head motion.

Results: Adolescents with current depression exhibit reduced neuromelanin signal in the substantia nigra pars compacta ($b = -1.88$, $t = -2.51$, $p = .02$). Lower SNpc neuromelanin-MRI signal associates with more severe depression (CDRS; $B = -0.45$, $t = -2.67$, $p = .01$), social anhedonia (ACIPS; $B = -0.46$, $t = -2.89$, $p = .009$), and suicide ideation (SSI; $B = -0.59$, $t = -4.23$, $p < .001$.

Conclusions: These novel data support the role of midbrain dopaminergic deficits in adolescent depression. Ongoing analyses will examine associations with real-world affect and anhedonia via smartphone ecological momentary assessment as well as associations with other neuroimaging modalities, i.e., linking midbrain neuromelanin with reward response during functional MRI. We aim to probe the utility of neuromelanin-MRI, in combination with other measures, as predictors of course and continuity of depression symptoms in this time of rapid adolescent development. Future studies will build on this to probe potential dopaminergic risk markers in earlier child development to predict depression onset before this adolescent peak.

Keywords: Adolescent Depression, Anhedonia, Neuromelanin-Sensitive MRI, MRI

Disclosure: Nothing to disclose.

P391. Development and Characterization of a Novel Fluorinated Etonitazene Analog as a Potential Radiotracer for Mu Opioid Receptors

Juan Gomez*, Agnieszka Sulima, Arianna Rizzo, Zelai Garcon-Poca, Emilya Ventriglia, Jordi Bonaventura, Kenner Rice, Mike Michaelides

National Institute on Drug Abuse, Baltimore, Maryland, United States

Background: Opiates have long been used and studied for their analgesic properties and synthetic mu opioid receptor (MOR) agonist medications are the most effective analgesics available. However, such medications have known adverse effects including constipation, respiratory depression, and abuse liability. As such, there is a desperate need to develop novel MOR agonists with lower adverse effect profiles. A critical tool for such an endeavor involves the development of a MOR-selective radiotracer for in vivo target engagement studies using positron emission tomography (PET). The only MOR selective PET radiotracer developed to date is [11 C]carfentanil. [11 C]carfentanil has been used in many studies to measure MOR binding in humans and laboratory animals. Despite its use, [11 C]carfentanil has two key limitations. It has very high potency, which necessitates achieving very high specific activity in its radiosynthesis, and its use is restricted to PET studies in centers with on-site cyclotrons. Accordingly, an 18F-labeled MOR selective agonist PET radiotracer which has longer half-life and lower potency than [11 C]carfentanil would be desirable, but such a radiotracer has not been previously developed. Etonitazene is a selective and potent MOR agonist that has not been studied extensively. The aim of these studies was to characterize a novel fluorinated etonitazene analog (aka fluornitrazene, FNZ) and assess its potential to be radiolabeled and used as a MOR selective in vitro 3H-labeled radioligand and 18F-labeled PET radiotracer.

Methods: To assess it selectivity, FNZ was screened against a panel of >100 receptors and enzymes at 100 nM and 10 μM concentrations. To assess its propensity for brain entry, FNZ was also screened for its drug transporter inhibition activity against a panel of several drug transporters using these same concentrations. FNZ and [3H]FNZ were studied via competitive binding assays using rat brain membrane suspensions (minus cerebellum) (40 μg of protein/ml) incubated in 50 mM Tris-HCl (pH 7.4) containing 10 mM MgCl2, 10 nM of [3H]DAMGO (46 Ci/mmol) or 1 nM [3H]FNZ (43 Ci/mmol) and increasing concentrations of the

tested compounds (DAMGO, FNZ) incubated of 2 hours at room temperature. Non-specific binding was determined in the presence of 100 μM naloxone. In all cases, free and membrane-bound radioligand were separated by rapid filtration in a 96-well plate harvester and washed with 2 mL of ice-cold Tris-HCl buffer. Microscint-20 scintillation liquid (65 μL/well) was added to the filter plates, incubated overnight, and radioactivity counts were determined in a MicroBeta2 plate counter. One-site competition curves were fitted, and Ki values were calculated using the Cheng-Prusoff equation. FNZ was also evaluated in its propensity to stimulate cAMP and β arrestin-signaling using HEK293 cells transfected with hMOR cDNA and the respective genetically encoded sensors.

Results: At 10 μM, FNZ inhibited binding to serotonin (5-HT1A, 5-HT2A, 5-HT2B), adrenergic (α1A, α1D), cannabinoid (CB2), opioid (delta, kappa, MOR) receptors, as well as to calcium (L-type, diltiazem site), and potassium (hERG) channels. At 100 nM, FNZ inhibited binding only to MOR. In the efflux transporter panel, at 10 μM, FNZ inhibited transporter activity of OCT2, BSEP, MATE1, MATE2-K, OAT3, OATP1B1, OATP1B3, and p-glycoprotein (P-gp). At 100 nM, FNZ inhibited the activity of only OCT2 and MATE2-K, which are restricted to the periphery. Competitive binding assays against [3H]DAMGO showed that FNZ had a Ki = ~1.0 nM. Similarly, [3H]FNZ showed a Kd = ~1.3 nM. FNZ showed an EC50 of ~0.1 nM and Emax ~100% for cAMP and EC50 of ~10 nM and Emax ~100% for β arrestin.

Conclusions: FNZ is a selective agonist for MORs and [3H]FNZ exhibits favorable properties as an in vitro radioligand. FNZ exhibits minimal interaction with efflux transporters expressed in the periphery and therefore may have potential as a PET radiotracer.

Keywords: Mu-Opioid Receptors, Functional Characterization, Fluornitrazene

Disclosure: Nothing to disclose.

P393. GPR6 as a Modulator of Striatal-Based Motor Plasticity Under Conditions of Dopamine Inhibition

Brenna Williams*, Anuj Patel, Zoe Frank, Maellie Midroit, Hailey Huddleston, Patrick LaChance, Michael J. Frank, Kevin G. Bath

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

Background: Parkinson's disease stems from depleted striatal dopamine (DA), leading to motor impairments such as bradykinesia, akinesia, and tremor. Theoretical models have suggested that the source of these impairments is not in motor execution per se, but is rather motivational, whereby DA depletion enhances the brain's representation of the cost of action. Moreover, DA depletion not only induces a performance effect (amplifying the motivational cost), but is associated with aberrant learning, such that motor performance degrades with experience as the cost is learned in a particular context. Experimental studies manipulating DA blockade or depletion separately during learning and performance phases have provided strong evidence for the aberrant learning hypothesis, resulting from a lack of D2 receptor binding in the indirect corticostriatal pathway. GPR6, a G- protein coupled receptor, is expressed highly specifically to D2-containing striatopallidal neurons of both mice and humans; hence manipulation of GPR6 function provides a novel opportunity to intervene with the aberrant learning process. However, the mechanisms by which GPR6 affects performance or learning remain unknown. Given the putative role of GPR6 in plasticity-dependent processes, together with its selective expression on D2 cells, we predicted that loss of GPR6 would diminish aberrant learning under pharmacological DA receptor blockade and would result in paradoxically more rapid recovery following drug removal than is observed in wildtype mice.

Methods: Male and female wildtype C57Bl/6 N and GPR6 KO mice (n = 8-10/group/sex) were assessed for motor learning and performance degradation on an accelerating rotarod to assess the effects of D1 and D2 receptor antagonists (including a D1/D2 cocktail) on the acquisition of a novel motor learning task and subsequent performance when drug was removed. Ongoing experiments are underway to assess animals for biochemical markers of plasticity. Based on previous studies investigating reinforcement learning, and our knowledge of signaling pathways affected by GPR6, we will specifically test for the activation of plasticity markers DARPP-32, CREB, and MAPK.

Results: We found that DA receptor blockade impaired motor performance and that D2 and D1/D2 cocktail, but not D1 blockade alone, significantly impaired recovery in motor performance following drug removal. All mice showed impaired performance under dopamine antagonists (Days 1-5, "direct performance effects"). Following drug removal (D2 or D1/D2 cocktail), wildtype mice showed slowed recovery (WT + Drug vs WT/KO + Sal, one-way repeated measures ANOVA, post-hoc Tukey: Days 8-12, $p < 0.05$). Consistent with our hypotheses, GPR6 KO mice demonstrated enhanced performance after drug removal and showed a more rapid and complete progression to "normal" performance when compared with wildtype mice (GPR6 KO + Drug vs WT/KO + Sal, one-way repeated measures ANOVA: Days 8-9, $p < 0.05$, D10-12, ns). This effect was most dramatic in conditions in which both D1/D2 receptors were blocked. Notably, GPR6 did not impact the direct performance effects of DA receptor blockade, and thus appears to selectively impact the aberrant learning component (GPR6 KO + Drug vs WT + Drug, one way repeated measures ANOVA: Days 1-5, ns).

Conclusions: Together, these data demonstrate that loss of GPR6 receptors diminishes the behavioral learning effects that occur under D2 blockade, suggesting that variation in GPR6 function modulates the extent to which aberrant learning occurs via plasticity of D2 striatopallidal neurons. Further experiments will elucidate whether, in wildtype mice, motor learning under D2 blockade will lead to enhanced activation of proteins involved in plasticity in D2-positive cells. We anticipate that loss of GPR6 will attenuate motor learning induced expression of phosphorylated DARPP-32, CREB, and MAPK, indicating decreased plasticity of D2 positive cells. These findings provide evidence that GPR6 represents an under-studied but promising target and pathway for intervention in various frontostriatal disorders that affect motor, motivational, and cognitive function, including but not limited to Parkinson's disease.

Keywords: Neuropsychiatric Disorders [Schizophrenia, Parkinson's Disease, Major Depressive Disorder], Dopamine, GPCRs, Motor Learning

Disclosure: Nothing to disclose.

P394. Nanoscale Imaging of pT217-Tau in Aged Rhesus Macaque: Trans-Synaptic Propagation and Seeding of Tau Pathology in Entorhinal Cortex

Dibyadeep Datta*, SueAnn Mentone, Yury Morozov, Rosalinda Roberts, Christopher van Dyck, Amy Arnsten

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

Background: Tau pathology in Alzheimer's Disease (AD) targets higher cortical circuits, with evidence that phosphorylated tau propagates from the entorhinal cortex (ERC) to "seed" pathology throughout the neuronal network. Recent discoveries indicate that tau phosphorylated at threonine 217 (pT217-tau) can be captured

in cerebrospinal fluid (CSF) and plasma as an early biomarker of ensuing disease. pT217-tau levels correlate with disease stage, progression and longitudinal rates of change. Importantly, CSF pT217-tau permits early identification of at-risk presymptomatic individuals and shows high specificity for conversion to mild cognitive impairment and dementia. More recently, pT217-tau in blood plasma has been shown to distinguish AD from other neurodegenerative disorders with high diagnostic accuracy (>98%) and was more accurate than blood-based tests for pT181-tau, neurofilament light or Aβ-42/40 ratio. Plasma pT217-tau collected during life correlated with neuropathological neurofibrillary tangle (NFT) density measured postmortem. CSF and plasma pT217-tau is increased ~7-fold in AD, and the levels increase 20 years before onset of cognitive impairment. Recent high-resolution quantitative proteomics map of post-translational modifications (PTMs) on multiple isoforms of tau revealed pT217-tau as a crucial epitope distinguishing AD from other neurodegenerative disorders and indicative of disease progression. However, the role of pT217-tau in brain tau pathology is unknown, especially as soluble tau species are dephosphorylated postmortem in humans. Rhesus macaques naturally develop the same qualitative pattern and sequence of tau and amyloid pathology, with NFT's comprised of paired helical filaments, identical to human AD. Perfusion fixation of monkey tissue preserves phosphorylation state and allows imaging of molecular location and interactions with nanometer resolution not possible in humans due to postmortem degradation. The current study examined the ultrastructural localization of pT217-tau in layer II ERC of the aged rhesus macaques, focusing on potential evidence of propagation between neurons, and exposure to the extracellular space.

Methods: We used immunohistochemistry to examine the anatomical localization pattern of pT217-tau in aged rhesus macaques and compared to human postmortem AD subjects. We performed high spatial-resolution immunoelectron microscopy (immunoEM) in aged rhesus macaques (18-31 years) to localize pT217-tau in the stellate cell islands in ERC layer II, which show the earliest signatures of tau pathology in AD.

Results: Our data at the light-level reveals dense pT217-tau immunolabeling in stellate cells in ERC layer II, pyramidal cells in hippocampus CA3, CA1, and pyramidal cells in dlPFC layer III, all of which show tau pathology in human AD. The labeling shows aggregated, filamentous fibrillated structures within apical dendrites and basilar dendrites, often with a twisted morphology common in NFTs. pT217-tau immunolabeling was predominantly observed in postsynaptic compartments in macaque ERC layer II. pT217-tau accumulated on the calcium-storing smooth endoplasmic reticulum spine apparatus near axospinous asymmetric glutamatergic synapses in dendritic spines. We observed extensive, trans-synaptic pT217-tau trafficking between interconnected neurons within omega-shaped bodies and endosomes in ERC layer II, specifically near excitatory, but not inhibitory synapses. Within dendritic shafts, pT217-tau aggregated on microtubules often in concordance with autophagic vacuoles indicative of neurite dystrophy.

Conclusions: pT217-tau accumulates in ERC layer II subcompartments known to be the earliest to show pathology in humans. The data provide the first evidence of pT217-tau trafficking between neurons to "seed" tau pathology in higher brain circuits, potentially interfacing with the extracellular space to become readily accessible and captured in CSF and blood as a robust AD biomarker. Illuminating patterns of neurodegeneration with pT217-tau could potentially guide earlier intervention of therapeutics that might mitigate tau hyperphosphorylation in AD.

Keywords: Alzheimer's Disease, Tau, Entorhinal Cortex, Protein Trafficking, Neurofibrillary Tangle

Disclosure: Nothing to disclose.

P395. Replication of the N170 Response to Faces for Use as a Potential Stratification Biomarker in Clinical Trials for Autism Spectrum Disorder

Matthew Klein*, Wen-Hsuan Chan, Molly Lucas, Srinivasan Vairavan, Abigail Bangerter, Nikolay V. Manyakov, Gayle Wittenberg, Gahan Pandina

Janssen Research and Development, San Diego, California, United States

Background: Autism spectrum disorder (ASD) is a complex neurodevelopmental condition characterized by impairments in social communication, and restricted and repetitive behavior. It often occurs in combination with other psychiatric conditions. There are no approved medications for the core symptoms of ASD and, partly, this is due to the highly variable symptom presentation. Stratification of the heterogeneity could allow for more targeted interventions. Currently, there are no validated biomarkers for ASD. The N170, an early-stage neural response to faces, has been proposed as a potential stratification biomarker. Longer latency, or slower response, to faces has been found to be related to social communication skills, making it a relevant candidate. For utility in clinical trials, biomarkers must also be reliable and robust. In order to move towards the validation of the N170 as a biomarker, results must be replicable within the context of use. In this abstract, we present an event-related potential (ERP) study for N170 obtained in a large sample of autistic and typically developing (TD) children and adults within a clinical trial context. In addition, we use age-adjusted modeling to create subgroups based on N170 response and investigate whether there are phenotypic differences between groups.

Methods: Biosensor data, including electroencephalogram (EEG), was obtained from ASD participants ($n = 144$) as part of a battery of passive viewing tasks at 3 time points (0, 4, 8 weeks) during an observational study (NCT02668991). Data from a group of TD participants ($n = 41$) was obtained at a single time point. For the face task design, static upright faces stimuli with either direct or averted gaze were presented. EEG recording was conducted using ActiCHamp 32 with 19 electrodes placed in accordance with the standard 10-20 system. ERPs were computed for epochs extending from 200 ms pre-stimulus to 1000 ms post-stimulus onset. ERPs were averaged by stimuli type after subtraction of the 200 ms pre-stimulus baseline.

We focused on N170 at the parieto-occipital sites (P7, P8, O1, O2). We calculated group differences (all ages, 6-12, $12+$) in averages for N170 peak latency and amplitude including sex and age as covariates. We assessed stability of these measures for the ASD group across 3 timepoints using intraclass correlation coefficient (ICC) from a linear mixed model. Correlations between ASD N170 features, and phenotypic data (IQ, ADOS, Autism Behavior Inventory) were calculated. Finally, we used a linear mixed effect model to estimate age-adjusted N170 latency for both TD and ASD groups, and calculated residual differences between expected and measured N170 latency (Webb, 2022). Using a 90th percentile cut off based on TD residuals, we categorized ASD participants into either "slowed" or "standard" groups. This approach allowed us to stratify ASD subgroups and to evaluate these N170 latency defined subgroups in relation to the phenotypic behaviors.

Results: The final number of participants with data for analysis was TD 30/ASD (97, 95, 91) for each respective timepoint, TD 15/ASD (43, 47, 38) and TD 15/ASD (54, 48, 53) respective to the age groups under and over 12 years old. There were no between group differences in response to condition (averted or direct gaze). For the 'all ages' group there was a group difference in N170 to all faces, where the ASD group had longer latency and

reduced amplitude. The latency group difference remained when age and sex were included in the analysis, and when the groups were analyzed by age. The ICC value was 0.81 (0.61 and 0.85 for age under and over 12 years old), suggesting moderate stability over 4 and 8 weeks for the measured N170 latency in the ASD. There were no correlations between N170 latency and phenotypic data, other than a small negative correlation with restricted interests, which was observed at all 3 timepoints. We observed significant differences in latency defined ASD (slow/standard) subgroups analyses. For the under 12 s, increased severity of ASD core behaviors was seen in the slowed N170 group and for over 12 s, increased irritability was associated with the standard N170 group. There was a difference in IQ between the subgroups, with a slowed response related to a lower IQ.

Conclusions: These findings are an important step in demonstrating the reliability and robustness of the N170 latency as a potential stratification biomarker for ASD. The relationships observed between subgroups and ASD core symptom severity in the younger age group confirm the relevance of this biomarker to the ASD phenotype. The relationship of this biomarker to age and IQ needs continued exploration.

Keywords: Autism, EEG, N170, Biomarker

Disclosure: Janssen Research and Development, LLC: Employee (Self), Johnson and Johnson: Stock / Equity (Self)

P396. Identification of Stable Clinical Subtypes in Autism Spectrum Disorder Using the Autism Behavior Inventory

Srinivasan Vairavan, Savannah Gosnell, Abigail Bangerter, Matthew Klein, Molly Lucas, Nikolay V. Manyakov, Gayle Wittenberg, Gahan Pandina*

Johnson and Johnson, Titusville, New Jersey, United States

Background: Autism spectrum disorder (ASD) is a complex neurodevelopmental condition characterized by impairments in social communication, and restricted and repetitive behavior/interests. Other psychiatric comorbidities are common in ASD. Variability in etiology and phenotypic outcomes of ASD contribute to clinical heterogeneity. Identification of subtypes that maximize homogeneity using clinical characteristics could potentially improve detection of changes in response to treatment. In this abstract, we identified clinical subtypes in ASD using 5-core domains (i.e., social communication, restrictive behaviors, mood and anxiety, self-regulation, and challenging behavior) as part of the Autism Behavior Inventory (ABI) and replicated findings across two different cohorts.

Methods: Baseline ABI data were obtained from three large studies, including an online survey, an observational study (NCT02668991), and an efficacy study (NCT03664232). Age, gender, and symptom severity (5-core domains in ABI) of the ASD subjects across three studies were subjected to propensity score matching with the efficacy study as reference, leading to a sample size of 180, 45, 45, in the online survey, observational study, and efficacy study, respectively. The distribution of intelligence quotient (IQ) across studies was comparable with mean IQ ≥ 90. The 5-core domains of the ABI were subjected to Uniform Manifold Approximation and Projection (UMAP) based dimensionality reduction. The number of clusters was chosen in a data-driven way using eigen gap metric. The UMAP parameters, namely, number of neighbors and minimum distance, were optimized based on distance between training and validation cluster, computed as mean distance of each point in training cluster (online survey) to its centroid minus the mean distance of each point in validation cluster (observational study) to the training cluster centroid. The UMAP parameters corresponding to the minimum number of clusters corresponding to the top three positive distances between training and validation cluster were chosen as the optimal parameters.

Results: Three clusters were identified corresponding to high, medium, and low symptom report across different domains in ABI and were stable across validation (observational study) and testing datasets (efficacy study).

Conclusions: These findings highlight the inherent clinical subtypes in ASD using ABI. The stable three-cluster profile across ASD will be further evaluated in the efficacy study to understand the enrichment in treatment response. In addition, the three-cluster profile will also be evaluated for enrichment in digital signatures observed via heart rate variability, evoked response (N170 corresponding to neural processing of faces), eye tracking and facial expressions.

Keywords: Autism, ASD, ABI

Disclosure: Johnson and Johnson: Stock / Equity (Self), Janssen Research and Development, LLC: Employee (Self)

P397. Dysregulation of the Kynurenine Pathway is Related to Persistent Cognitive Impairments After Tick Borne Encephalitis (TBE)

Lilly Schwieler*, Jacob Ahlberg Weidenfors, Vytautas Griška, Xueqi Li, Fredrik Piehl, Aukse Mickiene, Sophie Erhardt

Karolinska Institutet, Stockholm, Sweden

Background: Tick-borne encephalitis (TBE) is caused by neurotropic flavivirus infection and is one of the most serious neurological tick-transmitted diseases. TBE patients present long-term post-encephalitic neurologic and neuropsychiatric symptoms including cognitive decline. Importantly, clinical biomarkers and targeted treatments for post-encephalitic symptoms are currently unavailable. It is well known that virus infections induce tryptophan degradation via a cascade of enzymatic steps known as the "kynurenine pathway" (KP). Modulation of kynurenine metabolites during infection represents a finetuned system for regulating immune responses. This KP is also responsible for the biosynthesis of neuroactive compounds such as quinolinic acid (QUIN) and kynurenic acid (KYNA), both capable of impacting cognition. Indeed, altered KP activity has repeatedly been demonstrated in several diseases from which patients suffer cognitive decline. CSF KYNA associates with both poor cognition and psychosis in psychiatric disorders, while concentrations of CSF KYNA and QUIN are found elevated in HIV-1 infected patients and in patients with COVID-19, disorders with a high incidence of long-term cognitive dysfunctions. The present study aims to measure KP metabolites in cerebrospinal fluid (CSF) and serum of TBE patients and to investigate their relation to long-term neurocognitive performance.

Methods: TBE patients were recruited upon admission to Lithuania University of Health Science hospital, Kaunas during which serum and CSF samples were obtained, and the severity of encephalitic illness was classified according to clinical criteria as mild, moderate, or severe. During two follow-up occasions, 6 and 18 months after hospital discharge, patients underwent neurocognitive performance testing (MATRICS Consensus Cognitive Battery, MCCB) and additional serum samples were obtained. Serum and CSF concentrations of tryptophan, kynurenine, KYNA, QUIN, 3-hydroxykynurenine, picolinic acid, and nicotinamide were measured by ultra-performance liquid chromatography-tandem mass spectrometry (UPLC-MS/MS) in TBE patients and a non-inflammatory neurological disease (NINDC) control group. Following data collection, statistical analysis was carried out to investigate relationships between KP metabolites and patient neurocognitive performance and symptomatology. Linear regression models using log-transformed metabolite concentration data

were employed to assess differences in KP metabolite levels between the control group and TBE patients of different clinical severity. Interactions between measured KP metabolite levels and neurocognitive performance were assessed via Spearman rank-correlation tests.

Results: TBE patients (n = 87, median age 53, median BMI 25.9, 39% females) did not significantly differ from NINDC controls (n = 12, median age 52.5, median BMI 25.3, 58% females) with regard to age, gender, and BMI.

Statistical analysis revealed extensive induction of the KP in TBE patients compared to the control group. In CSF, concentrations of all kynurenine metabolites were significantly increased with notable drastic elevations of the neurotoxic metabolite QUIN (TBE: median 544.24 nM (222.85, 1199.63), NINDC: median 10.31 nM (7.82, 11.29), $p < 0.001$). The kynurenine/tryptophan ratio, (rKT) a proxy for kynurenine pathway activity, was greatly elevated in the CSF of TBE patients. Similar alterations in KP activity were observed in the serum of TBE patients, with elevations of all measured kynurenine metabolites and notable exceptions of KYNA and QUIN. Interestingly, serum rKT remained significantly elevated among TBE patients up to 18 months, indicating persistent, long-term activation of the KP in TBE patients. In addition, increasing disease severities corresponded with significant elevation of CSF kynurenine and rKT.

Conclusions: Our results demonstrate inductions of the KP in both serum and CSF of TBE patients, with persisting serum rKT that has not normalized 18 months after infection. Statistical analyses showed the importance of the magnitude of KP induction for the performance within several neurocognitive domains among TBE patients measured at 18-month follow-up. Confirming that kynurenine metabolites are elevated during TBE infection as well as understanding their role in symptomatology and cognitive decline may open up for new treatment interventions with the aim of dampening the activity in the kynurenine pathway.

Keywords: Cognitive Decline, Virus Infection, Kynurenine Pathway

Disclosure: Nothing to disclose.

P398. A Reliable Measure of Excitatory / Inhibitory (E/I) Balance in Alzheimer's Disease

Juan Molina, Joyce Sprock, Dylan Iwanaga, Jo Talledo, Yash Joshi, Bradley Voytek, Lisa Delano-Wood, Christopher Gonzalez, Steven Huege, Gabriel Leger, Greg Light, Neal Swerdlow*

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

Background: The balance of cortical excitatory and inhibitory synaptic activity ("E/I balance") has been proposed to be a critical mechanism for fine-tuning neural network activity within a narrow temporal window associated with the processing of sensory stimuli. Several studies have proposed that aberrant E/I balance in Alzheimer's Disease (AD) may: 1) reflect pathology in cortical glutamatergic, cholinergic and GABAergic transmission; 2) contribute to cognitive decline in this disorder; and 3) be a target for AD therapeutics. The study of E/I balance typically requires invasive methods, such as single-unit or voltage-clamp recordings in rodents and/or non-human primates, which have precluded their application to human studies. Recent findings suggest that the aperiodic, 1/f-like component of the neural power spectra may index tonic E/I balance and can be studied non-invasively using electroencephalographic (EEG) recordings. Using this approach, we reported that the uncompetitive NMDA receptor antagonist, memantine (MEM), normalizes E/I balance in patients with schizophrenia. MEM is used clinically to treat moderate-to-severe Alzheimer's Disease (AD); as part of a longitudinal study of biomarker predictors of MEM sensitivity in AD, we assessed the effects of acute MEM (0 vs. 20 mg, po) on E/I balance in 18 patients with AD.

Methods: Subjects to date are 18 carefully screened individuals with AD (mean (range): age = 72.8 (61-82 y); MoCA = 16.4 (6-23); education = 16.6 (12-20 y); M:F = 9:9). Baseline neurocognitive measures included the MoCA and ADAS-cog; GDS and NPI-Q scales assessed depression and general psychiatric symptoms. Subjects completed a double-blind order-balanced study of MEM (placebo (PBO) vs. 20 mg; 2 test days separated by 1 week) on subjective, autonomic, cognitive and electroencephalographic (EEG) measures. At 275 min post-pill, neurocognition was assessed via the RBANS. At 345 min post-pill, EEG measures were used to assess mismatch negativity (MMN), P3a amplitude and the Auditory Steady State Response (40 Hz coherence and power; ASSR). To analyze the aperiodic and periodic spectral EEG features, EEG signals were decomposed into their frequency-domain components via power spectral density (PSD) estimation using Welch's method. PSDs from the 4–50 Hz range were used to characterize the aperiodic "background" or 1/f-like signal and oscillatory components using a robust linear regression algorithm per our published methods.

Results: Aperiodic slope (E/I) measurements were reliable, i.e., significantly correlated across weeks ($R = 0.58$, $p < 0.015$), and were not significantly impacted by acute MEM ingestion ($F < 1$) nor by ongoing AChE-I use ($F = 1.72$, df 1,16, ns). PBO-week E/I values were not significantly associated with cognition, whether assessed by baseline MoCA ($R = 0.27$, ns) or ADAS-cog total scores (Core: $R = -0.34$, ns; Optional: $R = -0.41$, ns), or PBO-week RBANS score ($R = 0.12$, ns). PBO-week E/I values were also not significantly associated with baseline GDS ($R = -0.375$, ns) or NPI-Q scores ($R = 0.05$, ns). Acute MEM effects on E/I values did not correlate significantly with those on cognition (RBANS Index Total Score; $R = 0.24$, ns) or on either MMN, ASSR or P3a amplitude (all ns). After completing testing, all subjects began a 24-week open-label trial of MEM (10 mg BID), with neurocognitive assessment at 8, 16 and 24 weeks; any predictive effects of E/I measures on sensitivity to therapeutic response to MEM will be reported.

Conclusions: An EEG-based measure of "excitatory/inhibitory balance" can be acquired in patients with AD and shows significant test-retest reliability in these patients. In this modest sample, E/I balance is not associated with baseline neurocognitive deficits, is not sensitive to acute challenge with MEM (20 mg) and appears to index processes that are distinct from other evoked EEG measures (MMN, ASSR and P3a). Ongoing studies will address whether baseline E/I values, or their sensitivity to acute MEM challenge, predict therapeutic response to a 24-week trial of MEM.

Keywords: Alzheimer's Disease, Memantine, Excitatory / Inhibitory Balance

Disclosure: Nothing to disclose.

P399. Dysregulation of the Chromatin Environment Leads to Differential Alternative Splicing as a Mechanism of Disease in a Human Model of Autism Spectrum Disorders

Calvin Leung, Shoshana Rosenzweig, Brian Yoon, Nicholas Marinelli, Ethan Hollingsworth, Abbie Maguire, Mara Cowen, Michael Schmidt, Jaime Imitola, Ece Gamsiz Uzun, Sofia Lizarraga*

University of South Carolina, Columbia, South Carolina, United States

Background: Autism spectrum disorder (ASD) affects 1 in 44 children. Chromatin regulatory proteins are overrepresented among genes that contain high risk variants in ASD. Disruption of the chromatin environment leads to widespread dysregulation of gene expression, which is traditionally thought of as a

mechanism of disease pathogenesis associated with ASD. Alternatively, alterations in chromatin dynamics could also lead to dysregulation of alternative splicing, which is understudied as a mechanism of ASD pathogenesis. The anticonvulsant valproic acid (VPA) is a well-known environmental risk factor for ASD that acts as a class I histone deacetylase (HDAC) inhibitor. However, the precise molecular mechanisms underlying defects in human neuronal development associated with exposure to VPA are understudied.

Methods: To dissect how VPA exposure and subsequent chromatin hyper-acetylation influence molecular signatures involved in ASD pathogenesis, we conducted RNA sequencing (RNA-seq) in human cortical neurons that were treated with VPA. Neurons of male origin from three independent neuronal inductions were used at day 65. Differentially expressed genes (DEGs) were detected using DESeq2 software package (version 1.32.0) (adjusted $P < 0.05$ and a fold change $\geq |1.5|$). Differential transcript usage (DTU) events were identified using DRIMseq software package (version 1.20.0). Statistically significant DTU events were identified following stageR (version 1.14.0) post-processing analysis as events that has an adjusted $P < 0.05$. Differential alternative splicing events were identified using rMATS (version 4.1.1). Statistically significant differential alternative splicing events were identified as events with $FDR < 0.05$ and $IncLevelDifference \geq |0.1|$. ClusterProfiler (version 4.0.5) was used for functional enrichment analysis (GSEA, GO, and DisGeNET) of DEGs and DTU events. Significant enrichment results were obtained using a q-value cutoff of < 0.05.

Results: We observed that differentially expressed genes (DEGs) were enriched for mRNA splicing, mRNA processing, histone modification, and metabolism related gene sets. We observed widespread and distinct changes in gene expression in the VPA-treated samples compared to control samples with 3545 upregulated and 2663 downregulated DEGs. Furthermore, we observed widespread increase in the number and the type of alternative splicing events. Skipped exon (SE) and retained intron (RI) events were the most frequent splicing events detected in VPA-treated neurons at 40% and 22% of total events, respectively. Analysis of differential transcript usage (DTU) showed that exposure to VPA induces extensive alterations in transcript isoform usage across neurodevelopmentally important genes. Finally, we find that DEGs and genes that display DTU overlap with known ASD-risk genes.

Conclusions: In summary, our work highlights the importance of the chromatin environment in the regulation of alternative splicing in the pathogenesis of ASD associated with environmental risk factors and potentially genetic risk factors (chromatin regulators).

Keywords: Autism, Epigenetics, Alternative Splicing

Disclosure: Nothing to disclose.

P400. Inconsistent Patterns of Alzheimer's Disease- and Anxiety-Targeting Medication Use are Associated With Faster Cognitive Decline and Higher Tau Pathology Burden in Cognitively Impaired Elderly

Daniel Felsky*, Hyun-Sik Yang

Centre for Addiction and Mental Health, Toronto, Canada

Background: Effective and tolerable pharmaceutical management of illness in elderly individuals with cognitive impairment is a major clinical challenge. Over time, psychological and somatic changes manifest as a broad set of symptoms requiring intervention, sometimes with agents that may impact mental or general wellbeing. Therefore, decisions to start or stop treatment are complex, and little is known about the consequences of these decisions on longer term outcomes for patients. We sought to understand how different patterns of medication prescription were related to cognitive changes in elderly who developed mild cognitive impairment (MCI) or dementia due to Alzheimer's disease (AD).

Methods: 1,035 elderly individuals (age 80.7, sd=7; 71% female) from the Religious Orders Study and Memory and Aging Project (ROS/MAP) were assessed annually (range 3-28 yrs follow-up, mean=9 yrs) with 19 cognitive tests. Analyses only included participants who received a clinical diagnosis of MCI ($n = 416$) or dementia ($n = 619$) at their last study visit. Prescription status for 47 drug types among 20 broad classes (Medi-Span database coding; e.g., anti-AD medications, such as cholinesterase inhibitors, and anti-anxiety medications, such as SSRIs, benzodiazepines, etc.) were ascertained at each time point by direct inspection of containers. Individuals were classified into five discrete trajectory categories for each drug type: 1) never prescribed during study ("never"); 2) prescribed at all study visits ("always"); 3) not prescribed at study entry, transitioning to prescribed during study ("transition on"); 4) prescribed at study entry, transitioning to not prescribed ("transition off"); 5) prescription status fluctuated at some point during study between [on-off-on], or [off-on-off] ("unstable"). Trajectories of diagnostic stability were calculated in a similar fashion, identifying individuals who: 1) transitioned only from CN to MCI or MCI to AD; 2) transitioned from a more impaired diagnosis to a lesser impaired diagnosis at any point during study. Medication trajectories were associated with longitudinal rates of cognitive decline - per-individual random slopes of a linear mixed model for composite scores of global cognition - using ANCOVA, adjusting for sex, education, APOE ε4 status, age at baseline, smoking and alcohol intake at baseline, number of self-reported medical conditions at baseline, cognitive performance at baseline, depressive symptoms at baseline, clinical diagnosis at last visit, and number of follow-up visits. For medication trajectories with effects on cognitive decline, associations were tested for 11 neuropathologies in a subset of 757 individuals with autopsy data, including postmortem interval and age at death as additional co-variates.

Results: After Bonferroni correction for multiple testing, significant associations with cognitive decline were found for AD ($F4 = 12$, $p = 1.5\times10\text{-}9$) and antianxiety medication ($F4 = 11$, $p = 7.6\times10\text{-}9$) trajectories. For AD medications, the always prescribed group ($n = 30$) had the slowest cognitive decline and those who transitioned off medication ($n = 19$) experienced the most rapid decline (post-hoc comparison corrected $p < 0.001$). Significant differences were also found between both the always and transition on ($n = 177$) groups vs. the unstable ($n = 132$) group; the unstable group had the second most rapid cognitive decline following the transition-off group. For anti-anxiety medications, those never prescribed antianxiety medication ($n = 847$) showed the slowest cognitive decline, a significant difference from the unstable ($n = 132$) group (corrected $p < 0.001$), which had the most rapid decline. In both analyses, the always-off and transitioned-off groups were small – removing these groups from analysis finds consensus that unstable trajectories of medication are linked to faster rates of cognitive decline than stable. Neuropathologically, associations were observed for AD medication trajectories and both neuritic plaques and neurofibrillary tangles, whereby the transitioned-off group had the highest pathological burden – possibly reflecting the common discontinuation of these medications at more severe stages of impairment. The unstable trajectory group showed no difference in pathology compared to those who transitioned onto medication. For antianxiety medication trajectories, the unstable group was the only one significantly different from the never medicated group, showing elevated counts of neurofibrillary tangles (corrected $p = 0.013$). Finally, the AD medication trajectory was weakly associated with stability of AD diagnosis over time (Fisher's

$p = 0.054$), whereas antianxiety trajectories were not (Fisher's $p = 0.89$).

Conclusions: Unstable longitudinal patterns of AD and antianxiety prescription are associated with faster rates of cognitive decline, independent of a multitude of clinical and biological risk factors. Antianxiety medication trajectory specifically was not related to the stability of an individual's clinical diagnosis but may be linked to increased neurofibrillary tangle accumulation. Our results suggest that fluctuations in AD medication or antianxiety medication use might have a detrimental effect on cognitive prognosis, although reverse causation cannot be fully ruled out. Further study is needed to fully understand potentially causal relationships between these fluctuations and the acceleration of decline in elderly suffering from cognitive impairment.

Keywords: Cognitive Decline, Prescriptions, Trajectories, Alzheimer's Dementia, Neuropathology

Disclosure: Nothing to disclose.

P401. Opioid Use Disorder is Associated With Alterations in Circadian Pathways: Proteomics Analysis of Human Postmortem Brains

Stephanie Puig*, Ryan Salisbury, Anastasia Yocum, Jill Glausier, David Lewis, Zachary Freyberg, Marianne Seney, Matthew MacDonald, Ryan Logan

Boston University School of Medicine, Boston, Massachusetts, United States

Background: Prevalence rates of opioid use disorder (OUD) have increased dramatically, and many patients being treated for OUD relapse within first year. Vulnerability to opioid relapse is associated with severe disruption to sleep and circadian rhythms. Thus, improving circadian rhythms may be an effective intervention for reducing relapse. While relationships between OUD and circadian rhythm alterations have been studied in rodents, an understanding of the molecular alterations that occur in human brains of people diagnosed with OUD remains limited.

Methods: We used mass-spectrometry based proteomics to investigate protein alterations in human postmortem brains of OUD and unaffected subjects. Postmortem brains, provided by the Department of Psychiatry at the University of Pittsburgh School of Medicine, were collected from people diagnosed with OUD and unaffected control subjects. Subjects were matched on age, sex, postmortem interval (PMI), brain pH, and RNA integrity (RIN). To enable analysis of rhythms of protein expression, we used time of death information to build cohorts distributed across 24-hour timescale. We collected the nucleus accumbens (NAc), and on the dorsolateral prefrontal cortex (dlPFC), two structures strongly interconnected and heavily implicated in OUD. Quantitative proteomics with TMT was used to measure protein in NAc and dlPFC tissue homogenates and synaptosomes fractions. Limma was used to analyze differentially expressed proteins with adjusted and unadjusted p-values and log-fold-changes (logFC).

Results: Identification of rhythmic proteins showed that, in homogenates, the total number of rhythmic proteins was reduced in OUD subjects in both the NAc and the dlPFC (NAc: 115 rhythmic proteins in unaffected controls vs. 56 OUD subjects; dlPFC: 88 rhythmic proteins in unaffected controls vs. 53 in OUD subjects). Conversely, in synaptosomes of OUD subjects, total rhythmic proteins in NAc and dlPFC were decreased or increased, respectively. Interestingly, rhythmic proteins were largely different between groups in both, homogenates and synaptosomes. Notably, in synaptosomes, pathways enriched in OUD rhythmic proteins included membrane potential, vesicle-mediated transport, and GPCR signaling, primarily involved synaptic function. Circadian pathways were also identified as top enriched pathways in synaptosomes.

Analysis of differential rhythmicity that compares periods phases, amplitude and variance (R2), highlighted proteins that strictly change rhythmicity between unaffected and OUD subjects. In the NAc 25 and 23 proteins changed rhythmicity in homogenates and synaptosomes respectively and the majority lost rhythmicity. In the dlPFC, only 10 and 18 proteins changed rhythmicity in homogenates and synaptosomes, respectively. Interestingly, changes were equally distributed towards gain or loss of rhythm. In NAc synaptosomes, top pathways enriched in OUD proteins that changed rhythm included membrane trafficking, response to light stimulus and signaling by Rho GTPases. In dlPFC synaptosomes, they included platelet-derived growth factor beta signaling, tyrosine phosphorylation and GTPase signal transduction

Conclusions: Our study is the first to analyze changes in protein rhythms caused by OUD in human postmortem brains at a large scale. Our findings demonstrate significantly altered circadian rhythms in synaptic function and signaling in the NAc and dlPFC associated with OUD. These results provide insight on processes mediating alterations in circadian rhythms and sleep/wake cycles caused by OUD. Ongoing studies are investigating the possible functional links between these pathways in mouse models of OUD.

Keywords: Postmortem Human Brain Study, Proteomics, Circadian Rhythms, Opioid Use Disorder, Synaptosomes

Disclosure: Nothing to disclose.

P402. Multimodal Investigation of Structural and Functional Connectivity Changes Associated With 7q11.23 Copy Number Variations

Michael Gregory*, J. Shane Kippenhan, Tiffany Nash, Daniel Eisenberg, Philip Kohn, Zachary Trevorrow, Madeline Hamborg, Oliva Kline, Danya Adams, Carolyn Mervis, Karen Berman

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

Background: Williams syndrome (WS) and 7q11.23 duplication syndrome (Dup7) are caused by hemideletion (leaving one gene copy) or duplication (yielding three gene copies), respectively, of ~25 genes on chromosomal locus 7q11.23. Previous research has demonstrated alterations in both functional connectivity and white-matter microstructure in WS (i.e., Gregory 2019, Marenco 2007). However, despite the opportunity to test for gene-dosage effects in research by comparing individuals who have hemideletions to individuals who have duplications of the same set of genes, little investigation of this nature has been carried out. Neuroimaging of myelination is a particularly good candidate for such investigation because rodent studies have demonstrated that knockout of the GTF2I gene, which is affected by these 7q11.23 copy number variations (CNVs), is associated with decreased brain myelination (Barak 2019). Here, we took a data-driven approach to identifying both structural and functional connectivity alterations associated with 7q11.23 CNVs by combining data from three neuroimaging modalities: After searching across the brain for regions with altered resting-state functional connectivity that was related to 7q11.23 CNV dosage, we identified white-matter tracts connecting these regions in each participant, and then tested for changes in myelination within these tracts, again, as a function of gene dosage.

Methods: Resting-state fMRI, diffusion tensor imaging (DTI), and mcDespot sequences for quantitative myelin imaging were

collected for 70 participants (WS: N = 20, age=13.8 ± 4.4, 15 females; typically developing: N = 34, age=14.3 ± 3.8, 21 females; Dup7: N = 16, age=14.8 ± 2.7, 8 females). Quality control measures identified 11 participants whose mcDespot data were not suitable for processing (four individuals with WS, five typically developing individuals, and two individuals with Dup7). First, for resting-state fMRI data, multivariate distance-based matrix regression (MDMR) controlling for age, sex, and motion was used to identify brain regions where whole-brain functional connectivity patterns significantly related to CNV dosage ($p < 1\times10\text{-}9$). Second, after preprocessing of DTI data to calculate voxel-wise tensors, significant regions emerging from the first step, the MDMR resting fMRI analysis, were inflated such that they extended to include underlying white matter; these inflated regions were then used as seed regions in probabilistic tractography analyses using AFNI's FATCAT software to identify white matter tracts connecting each pair of regions in each participant. Only white matter connections that could be identified in 90% of participants were carried forward in the analysis. Finally, voxelwise myelin water fraction (MWF) maps were computed for each participant from the mcDespot data using the publicly available QUIT pipeline; MWF was averaged across each tract identified from probabilistic tractography in each participant, and compared across CNV groups while controlling for age and sex using linear regression in R.

Results: Resting-state functional connectivity patterns that significantly related to CNV dosage were found in 22 gray matter regions. These regions were predominantly in areas known to subserve visuospatial and social functions, consistent with the neurobehavioral phenotypes observed in individuals affected by these CNVs. Probabilistic tractography identified 50 white matter tracts connecting these regions that were common across >90% of the participants. MWF in three of these tracts significantly related to CNV status (gene-dosage). These included tracts connecting posterior cingulate cortex with anterior cingulate cortex ($p = 0.0018$), right angular gyrus with right fusiform gyrus ($p = 0.0028$), and right parietal lobe with right prefrontal cortex ($p = 0.0038$). Myelination of all three tracts showed a pattern of increasing myelination with increasing copy number (Dup7 > typically developing > WS). No tracts showed the opposite pattern of significantly decreasing myelination with increasing copy number.

Conclusions: These data take a multimodal neuroimaging approach to identifying structural and functional connectivity changes associated with 7q11.23 CNVs. First, the 22 regions found to have altered resting-state functional connectivity largely subserve visuospatial and social functions, which are key components of the neurobehavioral phenotypes of WS and Dup7. Second, consistent with previous reports of decreased myelin basic protein in postmortem brain samples from individuals with WS (Barak 2019), we found that myelination of three tracts connecting these regions increased with increasing 7q11.23 copy number. Among the white matter tracts investigated, these three are particularly notable for their role in connecting regions known to be major hubs of structural and functional brain networks, including posterior cingulate cortex, anterior cingulate cortex, angular gyrus, parietal lobule and prefrontal cortex (Oldham 2019), supporting the notion that the genetic mechanisms underlying 7q11.23 CNVs lead to altered myelination and that these neural features may contribute to the behavioral phenotypes in WS and Dup7. Further work may better characterize these structural and functional connectivity changes and may relate the findings to neuropsychological or behavioral measures.

Keywords: Williams Syndrome, 7q11.23 Duplication, Multimodal Neuroimaging, Myelin Imaging, Resting State Functional Connectivity

Disclosure: Nothing to disclose.

P403. Genetic Influences on Psychotic and/or Affective Neuropsychiatric Symptom Phenotypes of Alzheimer's Disease

Inga Antonsdottir*, Mary Ann DeMichele-Sweet, Lambertus Klei, Byron Creese, Clive Ballard, Bernie Devlin, Constantine Lyketsos, Robert Sweet

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

Background: Neuropsychiatric symptoms (NPS) including delusions or hallucinations (psychosis) and depression, anxiety, and/or irritability (affective disturbance) are common among people living with Alzheimer's disease (AD). Symptoms such as these contribute to more rapid and severe cognitive decline, while causing significant disability, morbidity, and mortality.

Methods: This genome-wide association meta-analysis examined data from persons with AD assessed on the Neuropsychiatric Inventory or Questionnaire (NPI or NPIQ) who participated in one of the following six source studies: the Fundació ACE Barcelona Alzheimer Treatment and Research Center (ACE/GR@ACE), a Consortium of National Institute on Aging Alzheimer Disease Centers (ADC), Eli Lilly and Company (LILLY), the Norwegian, Exeter and King's College Consortium for Genetics of Neuropsychiatric Symptoms in Dementia (NEXGENS), the National Institute on Aging's Late Onset Alzheimer's Disease Family Study (NIA-LOAD), and the University of Pittsburgh Alzheimer Disease Research Center (PITT ADRC). The presence of psychotic symptoms (AD + P; hallucinations or delusions) or affective symptoms (AD + A; depression, anxiety, and/or irritability) was indicated if participants had at least one of these symptoms at any visit as assessed by the NPI or NPIQ. A GWAS analysis was performed separately on each of the two phenotypes. Results from the AD + P and AD + A phenotypes were combined, and genetic correlation and heritability analyses were conducted using GenomicSEM.

Results: Data from 8,714 individuals with AD (60.2% female) were analyzed. Of these individuals, 32% had neither psychotic (AD-P) nor affective symptoms (AD-A), 28% had affective but not psychotic symptoms (AD + A; AD-P), 28% had psychotic and affective symptoms (AD + P; AD + A), and 12% had psychotic but not affective symptoms (AD + P; AD-A). Significant association occurred between affective and psychotic symptoms, with affective symptoms present in 70.1% of participants with psychotic symptoms versus 46.9% in participants without psychotic symptoms ($\chi2 = df=1$, $p < 0.001$).

There was significant heritability to both the AD + P and the AD + A phenotypes as well as to the joint phenotype. Based on a prevalence of AD + P and AD + A in AD subjects of 0.50 and 0.40, respectively, the estimated h2 on the liability scale was $0.23 + 0.06$ and $0.06 + 0.07$ for AD + P and AD + A, respectively. The estimated correlation between the two phenotypes on the liability scale was $0.55 + 0.44$. The estimated h2 on the liability scale for the joint phenotype was $0.155 + 0.065$. Genetic correlation of the joint phenotype with AD + P was $0.95 + 0.31$ and with AD + A was $0.78 + 0.64$.

When assessing specific genetic associations, we first contrasted AD-P to AD + P where two loci achieved genome-wide significance. One was in ENPP6 (rs9994623, O.R. (95%CI) 1.16 (1.10, 1.22), $p = 1.26 \times 10 - 8$) and the other at 19q13 spanning three genes, APOE, TOMM40, and NECTIN2 (of note, rs429358 is one of two SNPs defining the APOEε4 genotype [O.R. 0.84 (0.79-0.90), $p = 2.34 = 10\text{-}8$]). These findings are similar to those previously reported in a GWAS analysis of AD + P, with changes in p-value significance likely attributed to the change in sample size.

When contrasting AD-A to AD + A, no SNP reached genome-wide significance; however, a locus at 9q31 spanning RAD23B

approached significance (rs1805331, O.R. (95% CI) 0.80 (0.74, 0.87), $p = 1.331\times10\text{-}7$) and may become significant if the sample size were larger, as assessed by probability-probability plot.

When bivariate association tests were performed, no SNP reached genome-wide significance, however three approached significance. The first was at 9q31 spanning RAD23B (rs1805331, O.R. 0.80 (0.74, 0.87), $p = 1.33\times10\text{-}7$), a single SNP, rs112368830, at 1q42 (O.R. 1.39 (1.22, 1.57), $p = 2.53\times10\text{-}7$), and a third locus at 15q22 (best SNP rs35669194, O.R. 0.78 (0.70, 0.86), $p = 6.11\times10\text{-}7$), spanning GTF2A2 and the 3' portion of BNIP2.

Conclusions: These results show common genetic variation accounts for a significant portion of heritability in both phenotypes. These findings confirm prior work in associating AD + P with common genetic variation and extend the association to AD + A. Of note is the novel finding that the joint AD + P/AD + A phenotype is associated with common genetic variation. AD + P samples had a larger maximum absolute Z, and smaller minimum p-value, than observed values for AD + A. This suggests affective symptoms in AD are less distinctly driven by genetics as opposed to psychotic symptoms and could instead be more environmentally driven.

We note that the bivariate analysis was slightly better powered to detect genetic associations of NPS in AD than in either phenotype alone, conditional to the sample size being equivalent. This illustrates the importance of utilizing a joint affective-psychotic phenotype when conducting research on genetic associations of NPS in AD.

Given the near universal prevalence and adverse impact of NPS for patients with AD and considering the relative paucity of effective treatment options further study of emerging genetic influences on distinct NPS phenotypes has the potential of contributing to better treatments.

Keywords: Alzheimer's Disease, GWAS, Neuropsychiatric Symptoms (NPS)

Disclosure: Nothing to disclose.

P404. Development of a CSF1R Pet Ligand [18 F]JNJ-4249

Hartmuth Kolb*, A. Katrin Szardenings, Wei Zhang, Chunfang Xia, Tamara Berdyyeva, Su-Tang Lo, Mani Salarian, Christian Constantinescu

Janssen Pharmaceutical, San Diego, California, United States

Background: There is increasing evidence that neuroinflammation promotes and modulates neuropsychiatric disorders. Microglia play a critical role as key cellular components of the inflammatory process. Being able to image activated microglia in-vivo would provide a valuable tool to study neuroinflammation and responses to new targeted treatments in the brain.

Colony-Stimulating Factor 1 Receptor (CSF1R) is a receptor tyrosine kinase, predominantly expressed on macrophages and microglia, that regulates microglia activation and density. It therefore provides a target that could be used not only to measure the extent of inflammation in the brain, but also to develop new effective therapies.

Here, we describe preclinical studies of the PET ligand [18 F]JNJ-4249, a selective and potent CSF1R inhibitor.

Methods: The novel CSF1R PET ligand, [18 F]JNJ-4249 was evaluated in a series of standard in vitro assays and in-vivo imaging experiments in rodents and non-human primates. To induce neuroinflammation, two preclinical rodent models were introduced. A local neuroinflammatory model was induced by intracranial injection of Lipopolysaccharide (LPS) into the striatum of Sprague Dawley rats. A systemic neuroinflammatory model was induced by intraperitoneal injection of LPS into C57BL6 mice. Both models were investigated by imaging with [18 F]JNJ-4249, followed by post-mortem analysis of the brains by IHC (Iba-1) and Western Blotting (CSF1R). The tracer distribution and radiometabolite analysis were further investigated in non-human primates.

Results: JNJ-4249 is a highly potent (IC50 1.2 nM) and selective (400 kinase panel) PET ligand. In-vivo imaging of healthy rats showed good initial brain uptake and fast washout. In a locally injected LPS rat model, an increased (30%) uptake on day 2 and 4 post-LPS injection (20 µg, right striatum) is reproducible and consistent. On the other hand, 48 h post-IP injection of LPS in mice showed 50% increase in whole brain compared to naïve mice. Western blotting of brain tissue shows low levels of CSF1R expression in normal tissue, but increased expression in LPS treated animals (47 kDa, cytoplasmic domain). A baseline scan in healthy non-human primates shows a similar brain uptake and washout pattern as in rodents. The distribution was moderately homogeneous and demonstrated reversible kinetics. The VT (240 min) at baseline in whole brain was 12.4 mL/cm3, with a range of 11.1 mL/cm3 (cerebellum) to 15.2 mL/cm3 (frontal). Metabolism of the tracer was fast, forming more polar metabolites (65% and 4% parent remaining at 6 and 60 minutes, respectively).

Conclusions: The preclinical evaluation of [18 F]JNJ-4249 demonstrates that this tracer is a suitable candidate for imaging CSF1R in the brain and has potential to be used as a biomarker of microglia activation and neuroinflammation. Further experiments in neuroinflammation models are ongoing in preparation for first-in human studies.

Keywords: Molecular Imaging, CSF1R, Neuroinflammation, Microglia

Disclosure: Janssen: Employee (Self)

P405. Calbindin- And Parvalbumin-Positive Neurons and Microglia Display Sexual Dimorphism in Long-Term Responses to Methamphetamine and HIV-Associated Brain Injury

Marcus Kaul*, Rohan Shah, Ricky Maung, Daniel Ojeda-Juárez, Ana Sanchez, TMARC Group

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

Background: Methamphetamine (METH) use is a frequent comorbidity of infection with human immunodeficiency virus type-1 (HIV-1) and exacerbates HIV-associated neurocognitive disorders (HAND). However, the combined pathological mechanisms of METH and HIV-1 are incompletely understood. Transgenic mice expressing the HIV-1 envelope protein gp120 in the brain (gp120tg) share characteristic neuropathological features and gene expression patterns with neurocognitively impaired HIV/AIDS patients.

Methods: We previously exposed age-matched female and male gp120tg mice and non-tg controls to an escalating METH binge regimen for 25 days (HIV-1 gp120tg SAL n = 16, HIV-1 gp120tg METH n = 14, WT SAL n = 15, WT METH n = 18) and analyzed the animals 7 months later, at about 12 months of age using behavioral assessments, neurohistopathology and gene expression (Hoefer et al., Exp. Neurol. 2015). Both, HIVgp120 and METH compromised neurites, synapses and behavioral performance. In this follow-up study, we analyzed calbindin (Calb)- and parvalbumin (PV)-positive neurons and glial cells in a subset of the animals (n = 3 - 4 each of males and females per experimental group) using quantitative immunofluorescence and quantitative reverse transcription polymerase chain reaction (qRT-PCR). Statistical analysis employed ANOVA and Fisher's PLSD post hoc tests for comparison of the experimental groups.

Results: Calb and PV revealed sex-dependent differences in cerebral cortex and hippocampus with regard to the number of neurons, the quantity of the cell marker's expression, and the response to gp120 and/or METH exposure (RNA and protein). Similarly, pronounced sexual dimorphism was observed in microglia, including in the response to METH. In contrast, astrocytosis indicated only in cerebral cortex a sex-dependent difference associated with gp120 expression but not in response to METH.

Conclusions: In summary, METH exposure and viral gp120 compromise learning and memory function and induce neuropathology in both sexes. While injury to MAP-2 and synaptophysin-positive neurites and presynaptic terminals, respectively, does not reveal sexual dimorphism, Calb- and PV-positive interneurons and microglia display significant sex- and brain region-specific differences in the long-term response to METH exposure and HIVgp120 expression.

Supported by NIH, MH087332, MH104131, MH105330, DA052209 and DA026306 (P5) to M.K.

Keywords: HIV-Associated Neurocognitive Disorder, Methamphetamine, Drug Use Disorders, Microglial Activation, Astrogliosis

Disclosure: Nothing to disclose.

P406. Exploring Codon-Edited tRNA Approaches to Correct Premature Stop Codons in Mouse and Human Models of SCN2A-Related Autism and Intellectual Disability

Olivia Klein, Jonathan Sikora, Marisol Lauffer, Karina Kruth, Christopher Ahern, Aislinn Williams*

University of Iowa, Iowa City, Iowa, United States

Background: The SCN2A gene encodes the alpha subunit of the neuronal voltage gated sodium channel NaV1.2, which is necessary for action potential generation and propagation. Allelic variants of SCN2A are associated with autism spectrum disorder (ASD), intellectual disability, and seizure disorders. Loss of function mutations, such as nonsense mutations, can cause ASD and intellectual disability. Nonsense mutations create a premature termination codon (PTC) in the reading frame of NaV1.2 and result in functional haploinsufficiency. Therapeutic repair of these nonsense mutations could restore NaV1.2 sodium channel expression.

Currently, treatment of PTCs is limited to using small molecules with to promote ribosomal readthrough of the nonsense codon, which often results in generation of a missense mutation. These therapies would likely offer minimal clinical improvement for SCN2A and other ion channelopathies since many ion channels are intolerant of mutations, and missense mutations in NaV1.2 are strongly associated with epileptic encephalopathy.

We have generated a novel mouse model of an SCN2A PTC, R1626X, as well as a human patient-derived neuronal model of a different PTC, C959X, to test therapeutic approaches. We have also developed a novel codon edited transfer RNAs (tRNA) approach to correct PTCs. Codon edited tRNAs bind to the target PTC in mRNA but are charged with the correct amino acid, thereby generating full-length wild-type protein. Codon edited tRNAs have limited interaction with native stop codons. Here, we show that heterozygous SCN2A R1626X mouse display SCN2A autism-associated behaviors, and that our codon edited tRNA system can rescue PTCs in mouse and human neurons.

Methods: Mice heterozygous for the SCN2A R1626X mutation were generated through the University of Iowa Genome Editing Facility. To record ultrasonic vocalizations (USVs), pups were separated from dams at postnatal day 6 (P6) and recorded for 5 minutes ($n = 10$-15 per genotype). For behavioral work in adult mice, mice were tested between 10-20 weeks of age ($n = 9$-15 per genotype) on open field, novel object exploration, Erasmus Ladder, three-chamber social interaction test, and free social interaction using standard protocols. Both male and female mice were used for all experiments and results were analyzed by two-way ANOVA or linear mixed models, where appropriate.

For mouse neuronal cultures, cerebella were dissected from P6 pups to isolate cerebellar granule neurons (CGNs). Plasmids were transfected into neurons using PolyJet or Mirus reagents or were nucleofected (Lonza). Cells were analyzed by immunofluorescence 96 hours after transfection.

For human neuronal cultures, induced pluripotent stem cells (iPSCs) were generated from a patient with an SCN2A C959X mutation and a neurotypical control. iPSCs were differentiated into neural progenitors using dual-SMAD inhibition, and progenitors were differentiated into neurons in BrainPhys medium with supplements.

Plasmids in this study contain fluorescent reporters (mOrange2 or eGFP) engineered to encode a PTC and codon-edited tRNA to correct the PTC. Plasmids were transfected into human neurons using Lipofectamine STEM transfection reagent. For quantitative PCR, SCN2A expression was quantified using PrimeTime primers (Integrated DNA Technologies), normalized to GAPDH. For Western blot, cerebella were dissected from adult SCN2A R1626X mice and WT littermates, then lysed and run on a gradient gel under standard conditions. NaV1.2 protein was detected using a custom antibody (gift of Dr. Geoff Pitt, Cornell University).

Results: Heterozygous SCN2A R1626X pups had lower mean frequency and longer USVs compared to WT littermates ($p < 0.01$), with no differences in total USV number. Adult heterozygous SCN2A R1626X mice were more likely to explore the center of the open field than WT littermates ($p < 0.05$), with no differences in total distance traveled. Erasmus Ladder testing showed that SCN2A R1626X mice display improved gait adaptation ($p < 0.05$), a form of motor learning, with no evidence of ataxia. We observed no differences in novel object exploration or social behaviors using the three-chamber social interaction test. SCN2A R1626X mice have decreased NaV1.2 protein in the cerebellum, consistent with the PTC causing functional haploinsufficiency.

Although human SCN2A C959X neurons contain a PTC, SCN2A mRNA levels were not lower in C959X neurons compared to control neurons throughout their maturation in vitro, suggesting that human SCN2A transcripts containing PTCs do not undergo substantial nonsense mediated decay. Codon-edited tRNA were able to rescue PTCs in fluorescent reporters in both mouse and human iPSC-derived neurons, showing that codon-edited tRNA can be expressed in neurons and facilitate PTC rescue.

Conclusions: Heterozygous SCN2A R1626X mice display behaviors consistent with aspects of autism and intellectual disability, some of which can be detected at very young ages, along with reduced NaV1.2 protein. Human SCN2A C959X neurons express SCN2A and, to our knowledge, this is the first study to show that codon edited tRNA can rescue PTCs in human neurons. These models will be useful for testing codon-edited tRNA therapeutic approaches. Future directions include transitioning from plasmids to viral vectors, which have greater translational potential, and attempting to correct endogenous PTCs in these human and mouse models.

Keywords: Ion Channels, Autism Spectrum Disorder and Related Syndromes, Intellectual Disability, Behavioral Tasks, Induced Pluripotent Stem Cells (iPSCs)

Disclosure: Nothing to disclose.

P407. Optimization and Validation of Profiling Efforts Within the NINDS Preclinical Screening Platform for Pain (PSPP) to Accelerate the Development of Novel Non-Opioid, Non-Addictive Pain Therapeutics

Smriti Iyengar*, Mark Varney, Mark Urban, Elizabeth Dugan, David Budac, Qing Chang, Taleen Hanania, Sarah Woller

NINDS, NIH, Rockville, Maryland, United States

Background: The NINDS Preclinical Screening Platform for Pain (PSPP) was developed under the NIH HEAL Initiative to facilitate the identification and development of new non-opioid, non-addictive pain therapeutics. PSPP provides researchers from academic, industry, and government institutions worldwide, an efficient, rigorous, resource to accelerate development of novel small molecule, biologic, natural product, and device treatments for pain. Such novel assets accepted into the program are profiled to evaluate their effects in vivo in rats in extensively validated pain related models and endpoints, paying close attention to pharmacokinetic parameters, understanding potential for neurological deficits in vivo and drug abuse liability in vitro as well as in vivo.

Methods: Male and female SD rats (Envigo, Indianapolis, IN) 180-250 g, were acclimated for at least a week prior to testing and appropriately housed. During the study, 12/12 light/dark cycles were maintained with lights on at 6 am. The room temperature was maintained between 20 and 23°C with a relative humidity maintained around 50%. Chow (Lab diet 5001, LabDiet, St. Louis, MO, USA) and water were provided ad libitum for the duration of the study. The tests were performed during the animal's light phase between 8 am – 4 pm.

Pharmacokinetic studies were conducted to guide dosing, select the route of administration, and to determine the time course, supporting subsequent behavioral studies. The modified Irwin ($n = 4$) and rotarod tests ($n = 10$) were conducted to evaluate potential neurologic, physiologic, and fine motor effects that may impact outcome measures in the pain models. Following side effect profile assessment, efficacy was evaluated in the plantar incisional pain ($n = 10$) and L5/L6 spinal nerve ligation (SNL; $n = 10$) models. The plantar incision model is an established model of acute post-operative pain induced by incision of the skin and the plantaris muscle (Brennan et al. 1996). The model is characterized by transient hind paw tactile allodynia and spontaneous guarding behaviors. SNL is a model of peripheral neuropathic pain resulting from chronic nerve compression in which tactile and cold allodynia are produced (Kim and Chung, 1992). All experiments were conducted in a blinded manner with both sexes included.

Statistics: Data were analyzed by either one-way ANOVA or Repeated Measures ANOVA using GraphPad Prism (Version 9.2.0), followed by Dunnett's or Bonferroni's post-hoc comparisons when appropriate. An effect was considered significant at $p < 0.05$ level. Data are presented as the mean and standard error of the mean (s.e.m). Power analysis was used to determine the group sizes for the various assays.

Results: Results from the evaluation of duloxetine, celecoxib and diazepam suggest that all three drugs had differing profiles of efficacy in pain related models.

Administration of duloxetine (10, 30, 60, 100 mg/kg PO) did not affect performance on the rotarod in male or female rats at the doses or times assessed. In the modified Irwin test evaluating similar doses, the 10 mg/kg dose was well tolerated, but the 100 mg/kg treated animals consistently displayed decreased body position, decreased locomotor activity, and sedation. In both the plantar incision model and in the L5/L6 SNL model, duloxetine was evaluated at 3, 10, 30, 60 mg/kg PO. In the plantar incision model, the 60 mg/kg dose of duloxetine robustly reduced mechanical allodynia and guarding behaviors in male and female rats with the maximal effect occurring at 1 hour while in the SNL model duloxetine (60 mg/kg) robustly reduced mechanical allodynia and acetone cold sensitivity in both male and female animals compared to vehicle treated animals with maximal effect occurring between 1 and 2 hours post treatment.

Ongoing studies with celecoxib (3, 10, 30, 100 mg/kg PO) show that the drug did not affect rotarod performance in male or female rats at the doses or times assessed. In the plantar incision model (10, 30, 60 mg/kg PO), celecoxib produced a significant attenuation of guarding behaviors in male and female animals at 1, 2, 4 and 6 hours but did not affect mechanical allodynia behaviors.

At doses of 1, 3, 10 and 30 mg/kg PO, diazepam treatment produced sedative effects, including decreased body position, decreased locomotor activity, ataxia, visual placement after nose contact, and decreased grip strength in a dose dependent manner in both male and female animals in the modified Irwin test and significantly impaired rotarod performance in males at 1 hour and in females at 1 and 2 hours, post-treatment. In the plantar incision model, while diazepam (1, 3, 10 mg/kg PO) showed a statistically significant effect on PWT in male and female rats and on guarding behavior in female rats, it did not robustly alter these behaviors. Administration of diazepam showed no effect on either PWT or acetone cold sensitivity in male and female animals in the L5/L6 SNL model at similar doses across a 6-hour period post administration.

The results will be presented in the context of pharmacokinetic parameters.

Conclusions: In summary, the evaluation of clinically used drugs duloxetine, celecoxib and diazepam showed differing profiles in the PSPP paradigm, that were consistent with their utility clinically. The NINDS PSPP program strives to accelerate the development of novel non-opioid, non-addictive therapeutics for pain.

Keywords: Pain Therapeutics Profiling, Clinical Pain Standards, Pain Models

Disclosure: Retiree, Eli Lilly and Company: Other Financial or Material Support (Self)

P408. Oliceridine Demonstrates a Reduced Effect on Neurocognitive Function in Humans, Compared to Morphine: A Phase 1 Randomized, Placebo-Controlled, Dose-Ranging Partial Block Cross-Over Study

Mark Demitrack*, Michael Kramer, Cathy Michalsky, Hemme Hijma, Laurence Moss, Geert Jan Groeneveld

Trevena, Inc., West Chester, Pennsylvania, United States

Background: Oliceridine is an agonist at the mu-opioid receptor, with preferential post-receptor engagement of G-protein signaling, while showing reduced beta-arrestin recruitment and receptor internalization (DeWire, 2013). Opioid medications are an essential component of pain management after surgery, though a variety of opioid-induced adverse events (AEs) complicate their use. Notable among these is the development or exacerbation of cognitive dysfunction, which may range from sedation to confusion or progress to delirium. Cognitive dysfunction can therefore have potential implications for post-operative recovery and health outcomes, and in some instances may result in deficits that persist beyond the immediate post-operative period. The mechanism of these cognitive complications is unclear, though it has been hypothesized that opioids, such as morphine, can bind to the toll-like receptor 4 (TLR4), and the subsequent neuroinflammatory response may contribute to these postoperative cognitive sequelae (Muscat, 2021). Rats treated with oliceridine demonstrate reduced levels of spinal cord TLR4 after experimental fracture compared to morphine-treated animals (Liang, 2018). The present study was designed to characterize the neurocognitive impact of IV oliceridine versus IV morphine using a validated cognitive test battery. We hypothesized that IV oliceridine would demonstrate a reduced effect on cognitive measures compared to IV morphine.

Methods: Twenty-three healthy subjects (13 males, 10 females; median age 26 years), provided their informed consent and were randomized to receive 3 of the 5 possible treatments as single IV doses in a partial block cross-over design: placebo, oliceridine 1 mg or 3 mg, or morphine 5 mg or 10 mg. The dose range for each drug was selected based on prior data confirming a relative potency of oliceridine to morphine of approximately 1 to 5, and the maximum approved single dose of oliceridine (3 mg; Olinvyk® product label). Neurocognitive function using the NeuroCart® test battery was assessed at: baseline, 30 min, 1 h, 2 h, 3 h, 4 h, 5 h and 6 h. The primary outcome of the study was saccadic eye movement peak velocity, a sensitive measure of sedation. Secondary outcome measures included saccadic eye movement reaction time and % inaccuracy, smooth pursuit eye movement, an adaptive tracking test of eye-hand coordination, postural stability measured by body sway, and the symbol-digit substitution test. Right and left pupillometry, and analgesia measured with the cold pain test assessed adequacy of target engagement. Outcomes were examined with a mixed-model ANOVA, with treatment, period, time, and treatment by time as fixed factors, subject, subject by treatment and subject by time as random factors and average baseline value as a covariate. For a significant main effect of treatment, between-group pairwise comparisons of the treatment conditions were performed.

Results: Consistent with the known relative potency of oliceridine and IV morphine, across the dose ranges studied, both drugs demonstrated expected effects on opioid-induced pupillary constriction, and analgesia in response to cold pain testing.

There was a statistically significant effect of treatment on the primary outcome measure of saccadic eye movement peak velocity (main effect of treatment, $P < 0.0001$), driven by a favorable, reduced impact of oliceridine versus IV morphine (LS mean treatment difference [95% CI]: -11.40 degrees/s [-21.19, -1.61], $P = 0.0236$). Similar outcomes favoring oliceridine were observed on the secondary outcome measures of saccadic eye movement reaction time (main effect of treatment, $P = 0.0201$; LS mean treatment difference [95% CI]: 0.0088 sec [0.0010, 0.0166], $P = 0.0273$), reduced body sway (main effect of treatment, $P = 0.0314$; LS mean treatment difference [95% CI]: 16.1% (mm) [-2.7%, 38.5%], $P = 0.0951$), and improved performance accuracy on the adaptive tracking test, a measure of eye-hand coordination (main effect of treatment, $P = 0.0011$; LS mean treatment difference [95% CI]: -1.519% [-3.505, 0.467], $P = 0.1303$).

Additional neurocognitive outcome measures, visual tracking and the symbol-digit substitution test, did not show statistical differences between oliceridine and IV morphine. No serious adverse events were observed in the study. Common AEs included expected opioid-related adverse events of nausea, vomiting and somnolence. Events were assessed as mild in all cases with the exception of one AE of moderate nausea in a morphine-treated subject.

Conclusions: IV oliceridine has a reduced impact on several clinically relevant measures of cognitive performance, compared to IV morphine, including certain measures of sedation, motor performance, and eye-hand coordination.

Keywords: Cognition, Opioids, Clinical Psychopharmacology, Postoperative Cognitive Dysfunction

Disclosure: Trevena, Inc.: Employee (Self)

P409. Kappa Opioid Receptor Antagonism Suppresses Biochemical and Behavioral Phenotypes in Mice Expressing an ADHD and ASD-Associated Variant of the Dopamine Transporter

Felix Mayer*, Adele Stewart, Michelle Velez, Maximilian Rabil, James Foster, Roxanne Vaughan, Lynette Daws, Sammanda Ramamoorthy, Randy Blakely

Florida Atlantic University, Jupiter, Florida, United States

Background: The availability of extracellular dopamine (DA) is tightly regulated by the dopamine (DA) transporter (DAT). The rare DAT Val559 variant has been identified in individuals with attention-deficit hyperactivity disorder (ADHD), bipolar disorder and autism spectrum disorder (ASD). DAT Val559 exhibits anomalous DA efflux (ADE), a property that is shared by multiple disease-associated DAT variants. Mice expressing DAT Val559 display elevated extracellular DA in the striatum and present with hyperreactivity to imminent handling, waiting impulsivity, working memory deficits, and enhanced motivation for reward. ADE supports continuous activation of D2-type DA autoreceptors (D2AR), leading to elevated surface expression and hyperphosphorylation of DAT Val559 in a sex and circuit specific manner. The synthesis of dynorphin, the endogenous ligand of kappa opioid receptors (KORs) can be elevated postsynaptically by increased DA signaling at D1 receptors. In turn, activation of KORs on DA terminals leads to elevated DAT surface trafficking. Consequently, we hypothesized that antagonism of KORs could, by reducing dynorphin signaling, diminish surface levels of the efflux prone DAT Val559, and thereby suppress neurochemical and behavioral phenotypes in DAT Val559 KI mice.

Methods: We performed surface-biotinylation and immunopurification assays using acute coronal slices containing the dorsal and/or ventral striatum of DAT Val559 and wild-type (WT) control mice to assess the effect of the KOR agonist U69,593 and the KOR antagonist nor-binaltorphimine (norBNI) on surface expression and phosphorylation of WT DAT and DAT Val559 ex vivo. To measure the effect of systemic norBNI (10 mg/kg, i.p.) on DA-release in vivo, we performed microdialysis and fiber photometry, the latter effort performed using virally-expressed, genetically-encoded sensors for DA. Following vehicle or norBNI (10 mg/kg, i.p.) administration, WT and DAT Val559 expressing mice were assessed in the Y-maze and open field test. All experiments utilizing mice were performed under a protocol approved by the Institutional Animal Care and Use Committee (IACUC) at Florida Atlantic University.

Results: Using acute slices from DAT Val559 and WT mice, we found that DAT Val559 remains amenable to regulation via KOR. Regardless of genotype, exposure to U69,593 enhanced DAT surface expression that was accompanied by phosphorylation at DAT threonine 53 (Thr53). In contrast, exposure to norBNI normalized the basal elevated DAT Val559 surface expression and hyperphosphorylation at Thr53 but was without effect on WT DAT. Assessment of DA dynamics in DAT Val559 in vivo compared to WT revealed that systemic norBNI normalized vesicular DA release in the dorsal striatum of male mice. Finally, norBNI was found to restore the alternation-deficit (Y-maze) and anxiety-related behaviors (reduced time in center of the open field) observed in DAT Val559 mice.

Conclusions: DAT Val559 mediated ADE occurs independently from neuronal activity and results in the tonic activation of D2ARs, supporting a feedback loop that recruits additional efflux-prone transporters to the plasma membrane. By targeting KOR, a well-established regulator of DA neuronal activity and DAT surface expression, we identified antagonism of KOR as a promising strategy to ameliorate neurochemical and behavioral phenotypes that arise from the expression of a disease-associated DAT variant. More generally, our observation that antagonism of KOR was without gross effects on WT animals for the measures obtained supports KOR antagonism as a potentially useful strategy to offset biochemical and behavioral deficits that are linked to hyperdopaminergic states.

Keywords: Dopamine Transporter, ADHD, Kappa Opioid Receptor, ASD, Dopamine

Disclosure: Nothing to disclose.

P410. TRV045, a Novel, Selective S1P Receptor subtype-1 Modulator that Does Not Cause lymphopenia, Is Efficacious in Acute and Chronic Rodent Epilepsy Models

Michael Kramer*, Ruihua Chen, Mark Demitrack

Trevena, Inc., Chesterbrook, Pennsylvania, United States

Background: Epilepsy is caused by the aberrant synchronized firing of neurons resulting from the imbalance in excitatory and inhibitory neurotransmission. The mainstream antiepileptic drugs (AEDs) are direct modulators of ion channels, but they do not control seizures in 30% of patients. There is a need to discover and develop AEDs with novel mechanisms to address refractory epilepsy.

S1P and its receptors, especially subtype 1 (S1PR1), play important roles in neuroinflammation, a process underlying seizures and epileptogenesis. S1P receptor expression is increased in experimental post status epilepticus (SE) TLE mouse models. Fingolimod, a nonselective S1PR modulator but with high potency for S1PR1, has shown anti-epileptic effects in a diverse range of preclinical epileptic models, possibly through anti-inflammatory mechanisms as well as preservation of neuronal and blood brain barrier integrity. However, nonselective S1PR modulators such as fingolimod cause lymphopenia.

TRV045 is a highly selective S1PR1 modulator that does not cause lymphopenia in animal models. Through collaboration with the NINDS Epilepsy Therapy Screening Program (ETSP) we previously showed that subcutaneous dosing of TRV045 reduced seizures in the corneal-kindled (CK) mouse model and the rat maximal electroshock seizure (MES) model, but other models were confounded by a vehicle effect. Here, a new vehicle (10% cremophor, 20% Captisol in water) was used for oral (PO) dosing in the mouse CK, rat MES, Theiler's murine encephalomyelitis virus (TMEV) and post-kainic acid spontaneous recurrent seizures (KA-SRS) rat model of temporal lobe epilepsy.

Methods: In the MES model, 60 Hz of alternating 150 mA current was delivered to male Sprague Dawley (SD) rats for 0.2 seconds by corneal electrodes. Rats (n = 8/gp) were dosed PO with 30 or 60 mg/kg TRV045 and tested for seizure activity 0.25 to 2 hr postdose to identify the time of peak effect (TPE). An animal was considered "protected" from convulsant activity upon abolition of the hindlimb tonic extensor component of the seizure. A full dose response was then performed with testing at the TPE.

In the CK seizure model, male C57BL/6 mice were fully kindled to reach the criterion of 5 consecutive stage 5 seizures. Mice (n = 8/gp) were dosed PO with 10 or 15 mg/kg TRV045 5-7 days after the last stimulation and tested 1 and 2 hr postdose to determine the TPE. A full dose response was then performed with testing at the TPE to identify an ED50 dose.

In the TMEV model, male C57BL/6 J mice (n = 18-20/gp) were pretreated with TRV045 for 2 days (10 or 30 mg/kg, b.i.d., PO) and then infected with TMEV on Day 3. Daily dosing continued with assessment of seizure severity using a modified Racine scale through Day 7.

In the KA-SRS model, status epilepticus was induced in male SD rats with repeated low-dose kainate treatment. Following a 7-day baseline period, rats (n = 6/gp) received intraperitoneal (IP) doses of TRV045 or vehicle for 5 days. Following a 2-day washout period, animals were crossed over to the opposing treatment arm for a second 5-day period. Seizure burden was calculated as the summation of all Racine scale seizures during treatment divided by the number of treatment days. Seizure freedom was based on an animal having zero seizures from the time of first dose through 12 hr post-final dose.

Results: TRV045 prevented seizures in the rat MES model of generalized tonic-clonic seizures, with a TPE of 0.5 to 1 hr postdose. Tested 0.5 hr postdose, PO dosing of 20, 30, 40, 50 and 60 mg/kg TRV045 prevented generalized seizures in 1, 4, 3, 1 and 0 out of 8 rats, respectively. Tested 1 hr postdose, PO doses of 3, 10, 20 and 40 mg/kg TRV045 prevented generalized seizures in 0, 3, 2, and 0 out of 8 rats, respectively. An ED50 was not identified due to the inverted U-shaped dose response curve.

TRV045 protected mice against chronic secondarily generalized focal seizures in the CK mouse model. In the initial time-course screen, 10 mg/kg TRV045 PO protected 5 out of 8 mice at both 1 and 2 hr postdose, while 15 mg/kg protected 6 out of 8 mice at 1 hr and 4 out of 8 mice at 2 hr postdose, with average seizure scores ranging from 1.25 to 2.5. In the full dose-response study with testing at the TPE 1 hr postdose, 1, 2.5, 10, 15 and 20 mg/kg TRV045 PO protected 0, 4, 5, 6 and 6 out of 8 animals, respectively. TRV045 produced a dose-dependent effect on seizure scores, ranging from 2.5 (2.5 mg/kg) to 1.25 (20 mg/kg), resulting in a calculated ED50 of 5.97 ± 0.47 mg/kg.

TRV045 did not significantly reduce cumulative seizure burden in the TMEV model. Cumulative seizure burden was 88 ± 10% of vehicle in the 30 mg/kg group, with no effect on seizure freedom. Cumulative seizure burden was 90 ± 18% of vehicle in the 10 mg/kg group, with a nonsignificant increase in seizure freedom (7 out of 20 vs 4 out of 20 in the vehicle group).

In the KA-SRS model, 10 mg/kg TRV045 IP significantly reduced mean seizure burden compared to baseline (3.1 ± 0.9 vs 13.4 ± 6.6; $p < 0.05$ Wilcoxon rank sum) but did not significantly improve seizure freedom (3 out of 12 protected vs 2 out of 12 vehicle-treated). A 15 mg/kg dose significantly reduced mean seizure burden compared to the vehicle-treated group (5.0 ± 4.0 vs 9.0 ± 2.7; $p < 0.05$) and significantly improved seizure freedom (6 out of 12 protected vs 1 out of 12 vehicle-treated; $p < 0.05$ Fisher's exact test).

Conclusions: The S1PR1 modulator TRV045 prevented generalized seizures in the CK mouse and rat MES models and reduced seizures in the KA-SRS chronic spontaneous seizure model, indicating that it is a viable candidate for human research in the treatment of epilepsy.

Keywords: Epilepsy, S1P Receptor, Seizure, Preclinical Pharmacology, Pharmacotherapy

Disclosure: Trevena, Inc.: Employee (Self)

P411. Global Electrophysiological Excitatory to Inhibitory Imbalance in Hippocampus and Temporal Cortex in the Continuum of Alzheimer's Disease

Pietro Scaduto, William Russell, Agenor Limon*

University of Texas Medical Branch of Galveston, Galveston, Texas, United States

Background: Individuals at distinct stages of Alzheimer's disease (AD) show abnormal electroencephalographic activity, which has been linked to network hyperexcitability and cognitive decline. However, whether pro excitatory changes at the synaptic level are observed in brain areas affected early in AD, and if they are emergent in mild cognitive impairment (MCI), is not clearly known. Equally important, it is not known whether global synaptic E/I imbalances correlate with the severity of cognitive impairment in AD.

Methods: Using electrophysiology and proteomics of human synapses from the hippocampus and temporal cortex of control, mild cognitive impaired (MCI) and AD individuals we aimed to determine the global synaptic balance in hippocampus and temporal cortex at distinct stages of neuropathology. Electrophysiological synaptic E/I ratios in post-mortem samples from the temporal cortex of individuals with MCI (n = 6) or AD (n = 6) compared to non-demented controls (n = 6), and the

hippocampus (MCI, n = 8; AD n = 11, CTRL = 8) were assessed by microtransplantation of synaptic membranes (MSM). Proteomics of synaptosomes from temporal cortex were analyzed in the context of their electrophysiological responses using Electrophysiologically-anchored Dataset Analysis (EDA).

Results: We found that the higher the amplitude of GABA receptor currents the better the cognitive performance score. A similar association was observed for AMPARs currents. The electrophysiological E/I ratio was significantly higher in the TCx of AD subjects and was negatively associated with the MMSE in the TCx but not in the hippocampus. The synaptoproteome revealed the impact and directionality of protein alterations and neuropathology on the amplitude of synaptic receptors responses and cognitive MMSE scores. By using electrophysiologically anchored analysis of the synapto-proteome in the same individuals, we identified a group of proteins sustaining synaptic function and those related to synaptic toxicity. We also found an uncoupling between the function and expression of proteins for GABAergic signaling in the temporal cortex underling larger E/I and worse cognitive performance in this region

Conclusions: These findings indicate that early shifts of the E/I balance contribute to the loss of cognitive capabilities in the continuum of AD symptomatology.

Keywords: Excitation-Inhibition Balance, Alzheimer's Disease, Postmortem Brain Tissue, Excitatory Synapses, Inhibitory Synaptic Transmission

Disclosure: Nothing to disclose.

P412. Familiarity and Identity Representations in Hippocampal Area CA2

Lara Boyle*, Lorenzo Posani, Sarah Irfan, Steven Siegelbaum, Stefano Fusi

Columbia University, New York, New York, United States

Background: Memory is the basis of all complex social relationships. The hippocampus is critical for remembering interactions with, and information about, other individuals; in other words, it is critical for social recognition memory. Social memory consists of two related processes: the general sense of whether and to what extent one has previously encountered another individual (familiarity) and the specific recall of details or episodes that involve another individual (recollection). To date, the neural mechanisms underlying familiarity and recollection remain unclear, including whether familiarity and recollection depend on the same brain region. Prior studies in rodents have shown that the hippocampal CA2 subregion is of particular importance for social memory as assessed by social novelty detection. However, it is not known how familiarity and identity coding are represented within CA2 and relate to other variables known to be encoded in the hippocampus (space, time). Here, we investigated the neural mechanisms of familiarity and identity coding within hippocampal CA2, including consequences of different neural representation models for memory.

Methods: We injected a Cre-dependent virus into the dorsal CA2 (dCA2) region of Amigo2-Cre male mice to express the genetically encoded calcium indicator GCaMP6f selectively in dCA2 pyramidal neurons. We implanted a grin lens above CA2 to measure calcium events in a large number of dCA2 pyramidal neurons in awake, behaving animals. We imaged behaving mice exposed to different combinations and placements of novel and familiar individuals to explore changes in neural activity during experiments with variable combinations of familiarity, identity, position, and time. These combinations included: novel and familiar individuals in two trials with position swapped (same identity across trials, n = 12), novel and familiar individuals in two trials with position swapped (different identity across trials, n = 5), two novel individuals (n = 11), and two familiar individuals (n = 11). We characterized CA2 neural population representations using a decoding approach (linear support vector machine). In each case, we used data across the two trials to determine the degree that social, spatial, and trial information could be decoded. We then employed a technique called cross condition generalization performance, which gives information about how variables interact in the neural code (the dimensionality of these representations), to determine the relationship (the degree of entanglement) between different variables (familiarity, identity, space, and time) in the neural code.

Results: We found that the degree of familiarity of interacting partners and their locations can be decoded from dCA2 population activity (social decoding performance = 0.88; chance = 0.49 ± 0.02, $p < 0.001$; spatial decoding performance = 0.90; chance = 0.51 ± 0.02; $p < 0.001$). Trial decoding was lower than social or spatial decoding (trial decoding performance = 0.62; chance = 0.54 ± 0.02; $p < 0.001$). Trial decoding also informs the dimensionality of social and spatial coding, with low trial decoding suggesting low dimensionality, and high trial decoding supporting high dimensionality. We calculated the cross-condition generalization performance (CCGP), another marker of dimensionality. CCGP is inversely proportional to the dimensionality of represented variables. We found that familiarity and spatial CCGP were significantly above chance (CCGP = 0.77, chance level = 0.51 ± 0.03 (mean ± SD); $p < 0.001$; spatial CCGP = 0.76; chance level = 0.49 ± 0.03, $p < 0.001$). We then examined CCGP and trial decoding for equally novel and familiar individuals. We found that while novel individuals were represented with low dimensionality, with high CCGP and low trial decoding (Novel identity CCGP = 0.77; chance = 0.50 ± 0.04; $p < 0.001$; Trial decoding = 0.62; chance = 0.49 ± 0.02, $p < 0.001$), familiar individuals were represented in higher dimensions, with relatively low CCGP and high trial decoding (Familiar identity CCGP = 0.62; chance = 0.50 ± 0.04; $p < 0.001$; Trial decoding = 0.69; chance = 0.50 ± 0.032; $p < 0.001$). We generated a geometrical model that recapitulated these experimental findings. In addition, we developed a theoretical argument that captures the advantages of higher dimensional representations for recollection memory.

Conclusions: Our experiments and analyses demonstrate that dCA2 encodes both social familiarity and the social identity of individuals with similar degrees of novelty or familiarity, the latter being a key requisite of recollection. We find that familiarity is coded in a low dimensional space, meaning that as a variable this information is disentangled from other variables (identity, space, time). While novel identities are similarly encoded in a low dimensional framework, familiar identities in contrast are coded in higher dimensional representations, meaning that the variables of identity, space, and time are entangled. The distinct geometrical arrangements of familiar and novel animal representations provide a neural mechanism for how the brain may rapidly distinguish social familiarity from social novelty while storing detailed information of social experiences with familiar animals, thus enabling the same brain structure to encode the two key components of social recognition memory: social familiarity and social recollection.

Keywords: Dorsal Hippocampus, Social Recognition Memory, Recollection and Familiarity

Disclosure: Nothing to disclose.

P413. Correlating the Behavioral Sequelae of Blast Traumatic Brain Injury With Brain Structure and Function in Rats

Moriah McGuier, Crystal Noller, Diana Wallin, Lucas Dwiel, Emily Sullivan, Patrick McCunn, Katelyn Salotto, Armando Braz, John Kanter, Krista DiSano, Paul Holtzheimer, Wilder Doucette*

Geisel School of Medicine, Lebanon, New Hampshire, United States

Background: Nearly 69 million people worldwide are affected by traumatic brain injury (TBI). Military personnel are at high risk of experiencing TBI, especially shockwave-induced blast injury (bTBI), as results from proximity to explosive devices. bTBI increases the risk of developing posttraumatic stress disorder, mood and anxiety disorders, and substance use disorders. Current psychotherapeutic and pharmacologic treatments for these psychiatric consequences provide some symptom relief but may not address the underlying neural dysfunction. The use of pre-clinical models will allow us to identify neural correlates of blast and relate these biomarkers to emergent post-blast behavioral phenotypes. Through this understanding, we hope to guide the implementation of novel interventions (e.g., brain stimulation) to promote recovery from the chronic neuropsychiatric consequences of bTBI. We hypothesize that bTBI induced disruption of communication in the frontal-striatal-limbic system (using measures with translational relevance [e.g., MRI and electrophysiology]) will identify biomarkers of blast injury that relate to behavioral phenotypes.

Methods: 55 male Sprague-Dawley rats were exposed to a blast overpressure injury using a blast tube (ORA, Inc) or were naïve controls. We titrated blast parameters, including varying overpressure targets and interblast intervals, until significant changes in delay discounting task performance were observed ($n = 15$). We then used these parameters (three events ~126kPa [18 psi] with a ~3 min interblast interval [IBI]) on a large cohort ($n = 40$) and assessed an array of behaviors starting one month post-blast: 1) open field test with automated behavioral scoring; 2) sucrose preference; 3) Morris Water Maze (MWM) and 4) context fear conditioning. Following behavioral assessment, a subset of rats (12 bTBI and 12 control) were implanted with electrode arrays targeting the bilateral infralimbic cortex, orbitofrontal cortex, nucleus accumbens (NAc) core and NAc shell for LFP recording. We quantified 216 LFP features (power and coherence) across 6 established frequency ranges (delta through high gamma) and used machine learning to determine if blast and control LFP data could be classified more accurately than permuted datasets. Permute-and-relearn feature importance testing was then used to identify the most important LFP features. Another subset (8 bTBI and 8 control) underwent MRI for resting state functional (rs-fc) and diffusion tensor imaging (DTI) analyses. We used seed (medial frontal and NAc) based analyses to determine if significant connectivity differences existed between groups (blast versus control) and region of interest analyses corresponding to our electrode locations for DTI metrics (e.g., median diffusivity [MD]).

Results: We found that none of the rats in the 24 hour IBI group had significant DDT performance changes from pre-blast to post-blast while roughly half of the rats in the 3 min IBI group made significantly more impulsive choices (paired t-test $p < 0.05$) following blast. No anxiety or movement related differences were detected from the open field metrics but the automated behavioral scoring revealed that blasted rats groomed themselves significantly less (unpaired t-test $p = 0.0035$). Blast injured rats demonstrated higher sucrose preference (two-way ANOVA $p = 0.0182$) than control rats. We did not detect any group differences in the average latency to the platform across training days ($p = 0.1243$) for MWM. Following conditioning to aversive foot shock, no group differences during the 3-minute post shock interval ($p = 0.5106$) were detected but blast injured rats exhibited significantly more freezing (two-way ANOVA $p = 0.0458$) when returned to the conditioned context 24 hours later. Machine learning models revealed that LFP features could classify animals as blast versus control with average model performance (area under the receiver operator characteristic curve [AUROC] = 0.7656), which gives $p = 0.0348$ when compared to the permuted distribution. Feature importance testing indicated reduced low and high gamma power in frontal regions compared to controls. Rs-fc and DTI analyses revealed an array of structural and functional differences between blast injured animals and controls. For example, rs-fc analysis revealed the NAc seed had reduced connectivity to frontal regions and DTI analysis suggests ($p = 0.02$) decreased MD in frontal brain regions of blast injured animals.

Conclusions: Our data aligns with published work showing that blast injury in rodents is associated with a myriad of neurobehavioral consequences, including increased impulsivity and fear. Importantly, we found distinct neural signatures in injured animals that relate to the observed behavioral deficits, supporting a potential role for neural biomarkers in guiding rehabilitation post-injury. Future work will determine how these biomarkers predict recovery and response to treatment, including focal brain stimulation.

Keywords: Traumatic Brain Injury, Neuronal Oscillations, Functional and Structural MRI, Delay Discounting, Contextual Fear

Disclosure: Nothing to disclose.

P414. Activation of Astrocytes in the Paraventricular Nucleus of Thalamus Modulates Social Behaviors

Seungwoo Kang*

Augusta University Medical College of Georgia, Augusta, Georgia, United States

Background: Social behaviors are critical for animals to sustain healthy living patterns. Consequently, deficit or dysfunctional social behaviors are correlated with multiple psychiatric and neurological disorders. The paraventricular nucleus of thalamus (PVT) has been considered a hub brain area controlling context-dependent salience and aversion processing including the social behaviors. Indeed, recent studies suggest that the PVT presents a variety of neural signals according to social condition and the activities are significantly correlated with social behaviors.

Accumulating evidence demonstrate that astrocytes have not only the microstructural supporting role in neural circuit also physically form tripartite synapses with adjacent neurons and directly modulate the neuronal activities via release of gliotransmitters or uptake overflowed neurotransmitters. However, it remains unknown the coordinated activities of astrocytes and neurons in the PVT encode social behaviors.

Methods: To investigate the astrocyte activity-driven changes in the adjacent neuronal activities and behavioral consequences, we chemogenetically activated the astrocytes in the PVT using GFAP promoter-driven expression of hM3Dq, the excitatory designer receptors exclusively activated by designer drugs (DREADDs). First, we used ex vivo and in vivo calcium imaging to examine chemogenetically induced cellular activity in the PVT astrocytes of GFAP-GCaMP6s calcium fluorescence indicator-expressing mice. Then, we recorded electrophysiological changes in the firing and synaptic activity of the PVT neurons. To evaluate the behavioral consequences, we exposed mice in three-chamber social approach tasks. The mice were exposed to the chamber with three different social conditions: without any social factors for habituation and evaluation of place preference, with a strange mouse, or with a strange mouse and a familiar mouse. Social preference was measured as the comparison between the time the experimental animal spent with a stranger mouse and a novel object (sociability) or a familiar mouse (social recognition).

Results: Chemogenetic activation of the PVT astrocytes increased the intensity and frequency of calcium signaling. Interestingly, the astrocyte activation enhanced the firing and the evoked excitatory postsynaptic currents (eEPSCs) in the neurons of the PVT. The changes in eEPSCs induced by the

astrocyte activation were significantly inhibited by astrocyte-specific deletion of a glutamate transporter, GLT1 (as known as EAAT2, slc1a2) via the expression of GFAP-promoter driven Cre recombinase in the PVT of GLT1 floxed mouse. The changes in eEPSCs were also occluded by the pretreatment of DHK, a GLT1-specific blocker, suggesting that GLT1 is, at least partly, required for astrocyte-induced changes in the neuronal activity in the PVT. In social behavioral evaluation, we found that the astrocyte activation significantly reduced the social recognition in the three-chamber social approach test. Importantly, mice lacking GLT1 selectively in the PVT astrocytes mimicked the astrocyte activation-induced impairment in social behaviors.

Conclusions: Together, our observation indicates that the astrocyte activity in the PVT modulates the adjacent neuronal synaptic events via not only the homeostatic passive activities but the direct modulation of the glutamatergic signaling, at least partly, through the GLT1, leading to shape social behaviors. This implies the importance of astrocytes as another layer to modulate social behaviors and the interaction between astrocytes and neurons via glutamatergic signaling could be a potential therapeutic target for related disorders.

Keywords: Social Behavior, Astrocyte, Glutamate Transporter, Paraventricular Nucleus of the Thalamus

Disclosure: Nothing to disclose.

P415. Cognitive Control Predicts Alleviation of OCD Symptoms by Ketamine

Peter van Roessel*, Sepehr Asgari, Booil Jo, Leanne Williams, Carolyn Rodriguez

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

Background: Obsessive-compulsive disorder (OCD) is a common and impairing illness. First-line pharmacologic treatment with serotonin reuptake inhibitors helps most patients, yet benefit may take weeks to accrue, and many patients don't achieve adequate response. Ketamine and other glutamatergic interventions offer great promise as alternative and more rapidly acting treatments. The growing diversity of treatment options, however, increases the need for data to support goals of precision medicine, matching the right treatment to the right patient.

OCD has been associated with alterations in both cognitive and affective processing, assessed using both neuroimaging and behavioral tests. These alterations include deficits in cognitive control and a valence bias reflected in increased threat responsivity and decreased reward responsivity. In secondary analysis of data from a randomized, active-placebo-controlled trial of intravenous ketamine for treatment of OCD, we explored whether validated behavioral measures of cognitive control and valence processing have utility as predictive biomarkers or as clinically meaningful targets of treatment.

Methods: Data were analyzed from N = 45 unmedicated individuals with DSM-5 obsessive-compulsive disorder who participated in a randomized controlled trial exploring the mechanisms of ketamine as a rapid-acting treatment. Participants were randomized 2:1 to receive a single 40 min intravenous infusion of ketamine (0.5 mg/kg) or midazolam (0.045 mg/kg). Clinical OCD symptoms were assessed using the Yale-Brown Obsessive Compulsive Scale (Y-BOCS) at baseline, at post-infusion Day 1, and at post-infusion Day 7. Participants additionally completed a validated, computer-based neurocognitive test battery ('WebNeuro') one day prior to and one day after infusion. Testing included measures of cognitive control (performance on Stroop color-word interference and Go/No-Go tests) and of valence processing (performance on tests of emotion identification and of implicit priming by facial expressions of fear and happiness), quantified as z-scores relative to age-, sex-, and years of education-matched healthy norms. We explored whether baseline performance on cognitive control and valence processing tests moderate symptom alleviation in response to ketamine vs midazolam, and whether pre-post change in test performance is associated with symptom alleviation. MacArthur criteria were used to define potential moderator and mediator variables.

Results: Treatment had a significant effect on OCD symptoms at Day 1 ($\beta = -7.894$, $p = .004$, representing a nearly 8 point mean difference in Y-BOCS change for ketamine vs midazolam) and at Day 7 ($\beta = -6.048$, $p = .005$). In moderation analyses, the interaction between a measure of Stroop interference by response speed and treatment had a significant effect on the change in Y-BOCS at Day 1 ($\beta = -4.0618$, Cohen's $f2 = 0.14$, $p = .04$), but not at Day 7. That is, the effect of treatment at Day 1 was considerably larger for those with faster normalized performance on a color-naming relative to color word-reading task. Performance on the Go/No-Go and valence processing tests did not moderate the treatment effect on outcomes at Day 1 or Day 7. In mediation analyses, treatment did not have a significant effect on pre-post change in cognitive control or valence processing; these measures did not fulfill criteria as potential mediators of Y-BOCS change. Pre-post change in Stroop interference by response speed did, however, correlate with change in Y-BOCS at Day 1 ($r = 0.36$, $p = .03$).

Conclusions: Ketamine robustly reduced symptoms of OCD at Day 1 and Day 7 post-infusion, compared to the active-placebo midazolam. Our data suggest that the short-term effect of ketamine vs midazolam on Y-BOCS may be influenced by baseline inhibitory cognitive control, such that greater control (less Stroop interference for color-naming) predicted greater benefit from ketamine vs midazolam. Our data may accord with published findings suggesting that Stroop task performance moderates the effectiveness of other OCD treatment modalities (e.g., cognitive behavioral therapy vs fluoxetine).

Keywords: Obsessive-Compulsive Disorder, Ketamine, Neurocognitive Assessment, Moderators, RCT

Disclosure: Nothing to disclose.

P416. Efficacy of Ketamine in Unmedicated Adults With Obsessive-Compulsive Disorder: A Randomized Controlled Trial

Carolyn Rodriguez*, Chi-Ming Chen, Gary Glover, Booil Jo, Daniel Spielman, Leanne Williams, Peter van Roessel, Charles DeBattista, Pamela Flood, Alan Ringold, Max Wintermark, Kelley Anderson, Omer Linkovski, Anthony Lombardi, Andrea Millen, Anthony Pinto, Hannah Raila, Keara Valentine, Maria Filippou-Frye, Jessica Hawkins, Elizabeth McCarthy, Pavithra Mukunda, Andrea Varias, Jordan Wilson, Brianna Wright

Stanford University, Stanford, California, United States

Background: Developing novel, robust Obsessive Compulsive Disorder (OCD) treatments is an urgent public health need, given OCD typically starts in childhood, leads to lifelong morbidity, and costs the economy $2.1 billion (direct costs) and $6.2 billion (indirect costs such as lost productivity) annually. OCD is characterized by an inability to inhibit intrusive thoughts (obsessions) and repetitive behaviors (compulsions). Serotonin reuptake inhibitor (SRI) treatment of OCD exhibits a long lag time (2-3 months) before clinical benefit, and this benefit is typically only partial. Identifying effective, fast-acting treatments will help reduce OCD morbidity and its life effects. We previously reported the rapid OCD symptom reduction of ketamine, a glutamate N-methyl-d-aspartate (NMDA) receptor antagonist, versus saline infusions in a proof-of-concept crossover trial (n = 15) in

unmedicated adults with OCD. Building on this initial finding, we evaluated the efficacy of ketamine in a larger group of unmedicated OCD patients with improved control conditions (active placebo control condition).

Methods: This was a randomized controlled trial of a single infusion of ketamine compared to an active placebo condition (midazolam, an anesthetic). With institutional review board approval, unmedicated adult patients (age 18-65) with OCD were randomly assigned under double-blind conditions to receive a single intravenous infusion of ketamine (0.5 mg/kg) or midazolam (0.045 mg/kg) in a 2:1 ratio (total n = 45). Participants met DSM-5 criteria for OCD with at least moderate symptoms (Yale-Brown Obsessive Compulsive Rating Scale score of at least 16). Exclusion criteria included severe depression (17-item Hamilton Depression Rating Scale was less or equal to 18 to enter the study) and comorbid psychiatric or medical conditions that made participation unsafe. The primary outcome was change in OCD severity 1 week after drug administration, as assessed by the Y-BOCS. Duration of effect was explored with weekly Y-BOCS up to 4 weeks post-infusion. We focused on estimating intention to treat effects based on longitudinal mixed effects modeling. For both moderator and mediator investigation, we employed the MacArthur approach embedded in mixed effects modeling.

Results: Regarding the primary outcome, the ketamine group had significantly greater improvement in Y-BOCS score than the midazolam group 1 week after treatment (Cohen's $d = 1.25$, $p < 0.001$). The effects from a single intravenous infusion of ketamine persist up to 3 weeks post-infusion (Cohen's $d = 0.59$, $p = 0.007$), gradually reducing each week and then becoming insignificant by Week 4. We examined age, sex, and race as potential moderators of treatment effects, although none were identified as significant moderators. We also examined change in dissociation as a potential mediator of treatment effect, although it did not turn out to be a significant mediator.

Conclusions: To our knowledge, this is the largest clinical trial to date of ketamine in unmedicated OCD patients. Ketamine demonstrated rapid and durable OCD symptom improvement compared to the active control condition. By using an optimized active placebo design to control for nonspecific anesthetic effects, this study provides new supporting evidence for the specific OCD therapeutic effects of ketamine.

Keywords: Ketamine, OCD, Midazolam

Disclosure: American Psychiatric Association Publishing: Other Financial or Material Support (Self), Biohaven Pharmaceuticals, Osmind: Consultant (Self), Biohaven Pharmaceuticals: Contracted Research (Self)

P417. Investigating the Role of Medial Orbitofrontal Cortex (mOFC) in Deterministic and Probabilistic Negative Reinforcement

Brittany Chamberlain*, Matthew Geramita, Susanne Ahmari

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: Obsessive compulsive disorder (OCD) is a chronic, severe psychiatric illness that is characterized by obsessions – recurrent intrusive thoughts and images – and compulsions – repetitive behaviors performed to relieve anxiety associated with obsessive thoughts. Negative reinforcement-based theories of OCD suggest that the manifestation and maintenance of compulsions may be driven by this temporary relief from obsession-evoked anxiety. A recent clinical study by our group (Panny et al., under review, Biological Psychiatry) found heightened activity in the medial orbitofrontal cortex (mOFC) when OCD patients removed compulsion-related images in a novel negative reinforcement paradigm. To further dissect the cellular responses contributing to these bulk regional changes in mOFC activity, we measured neural activity via single-photon calcium imaging in wild-type mice during a negative reinforcement task in this exploratory study.

Methods: C57Bl6/J mice (n = 6; 4 male, 2 female) were trained on a novel negative reinforcement task. During the task, a light cue predicted a foot shock, and mice could avoid shocks with 100% probability by pressing a lever within the 20 sec cue period ("avoid response"). If mice did not press the lever during the cue period, a series of shocks commenced. Mice could escape remaining foot shocks by lever pressing during this period ("escape response"). Mice performed 50 trials per day for 7 days. This was followed by 5 days of probabilistic reinforcement of responses, in which lever pressing led to shock avoidance on 50% of trials. Prior to training, mice were injected with virus encoding GCaMP6f (AAV5-CaMKII-GCaMP6f-WPRE-SV40, titer 2.2×10^12) into mOFC and implanted with gradient-index (GRIN) lenses to visualize mOFC activity using miniature microscopes (Inscopix). Single-cell calcium activity was extracted using CNMF-e algorithm, converted to ΔF/F, and time-locked to several timepoints within each trial (i.e. presentation of avoidance cue, lever extension, lever press, shock onset). Calcium transients aligned to behavioral events of interest that exceeded >1 SD from null distribution were considered significant.

Results: Mice quickly learned to avoid foot shocks, showing significantly more avoidance responses (59.3%) than either escape responses (29.3%) or trial failures (11.3%) beginning on the first day of training (avoid vs. escape: $p < .05$; avoid vs. failure: $p < .005$). By day 7 of training, mice avoided 99.8% of potential shocks, performing an avoid response on 97.3% of trials. Calcium imaging on day 1 of training demonstrated that 13.3% of neurons (69/518) were activated at onset of lever pressing to initiate an avoidance response; this effect was maintained over time, with 15.3% of neurons (107/699) activated at onset of avoidance responses on day 7. Ongoing analyses of cells tracked over days will determine whether activity of individual mOFC neurons changes as 1) avoidance behaviors become well-learned and maintained over time and 2) after changing from deterministic to probabilistic negative reinforcement.

Conclusions: In OCD, compulsive behaviors are reinforced by the temporary relief provided from anxiety brought on by intrusive thoughts and images. Preliminary findings from our lab show that patients with OCD display heightened activity in mOFC in response to removal of compulsion-related images, suggesting this may be a key locus of negative reinforcement in OCD. Here, we developed a novel task which leads to rapid acquisition of active avoidance responses in mice and coupled it with single-photon in vivo calcium imaging in mOFC. Preliminary analyses show that a subset of neurons in mOFC are selectively responsive during engagement in an active avoidance behavior. Further analyses will establish how mOFC responses are modulated throughout negative reinforcement training and following changes in task contingencies.

Keywords: Active Avoidance, Negative Reinforcement, Medial Orbitofrontal Cortex, Obsessive-Compulsive Disorder (OCD), In Vivo Calcium Imaging

Disclosure: Nothing to disclose.

P418. Precision Functional Mapping in Obsessive-Compulsive Disorder Using Dense Sampling Scanning

Matilde Vaghi*, Sunjae Shim, Jaime Ali Rios, Patrick Bissett, Pavithra Mukunda, Carolyn Rodriguez, Russell Poldrack

Stanford University, Stanford, California, United States

Background: One of the roadblocks in translating imaging research findings into clinical practice biomarkers is represented by shortcomings of traditional approaches. These generally entail analysis of limited data from each subject. Accordingly, inferences are based on cross-subjects averages which might obscure patterns of brain organization specific to each individual. Additionally, by focusing on an individual time point there is limited understanding of whether observed effects are stable over time or state dependent.

Here, we used a recently pioneered 'dense sampling' scanning acquisition where imaging data are collected on the same subject repeatedly over time, resulting in more data per individual. This experimental design enables conclusions to be drawn at the individual level and to capture brain fluctuations in the intermediate timescale of weeks to months, which might bear relevance to fluctuations in symptoms. We examined magnitude and anatomical distributions of network variability across subjects and sessions from seven high-quality, highly sampled patients with Obsessive Compulsive Disorder (OCD). Imaging data were collected while subjects performed cognitive tasks as well as during resting-state, but results presented here pertain to resting-state only.

Methods: Data were collected from 7 patients with a diagnosis of OCD (4 males; age: 22-48). For each subject, data were acquired on 10 sessions, each on a separate day, approximately one week apart. To minimize within-subject variability due to external factors, scans were performed at fixed times of the day, with rare variations due to participant or scanner's scheduling constraints. Each session entailed psychiatric evaluation as well as functional MRI. Overall, this resulted in approximately 6 h (3 h resting-state) of functional imaging per subject. MRI data were acquired on a research-dedicated GE 3 T MRI scanner using a 32-channel head coil. All functional imaging scans were performed using a multi-echo, multi-band, gradient-echo EPI sequence. Additionally, a T1 and a T2-weighted image were obtained for each subject. Imaging data were pre-processed via fMRIPrep 22.0.0 and denoised with Tedana. Functional connectivity (FC) was measured via time-series correlations among cortical regions parcellated using the Yeo's 17 networks template. Accordingly, a FC connectivity matrix was obtained for each subject and session. Effects were calculated per subject and were compared with one another using paired two-tailed t tests, with p values FDR corrected for the number of comparisons.

Results: Group and individual effects on network similarity were investigated by calculating correlations among FC connectivity matrixes. To quantify group effects, correlations between FC connectivity matrixes from different individuals and sessions were inspected. This analysis indicated a shared common basic structure, as functional networks from different individuals and sessions showed substantial similarity (mean Spearman's correlation = 0.76). However, networks from the same individual and different sessions were even more like each other, with an additional effect of mean Spearman's correlation = 0.86 over the group effect, demonstrating a large influence of individual identity on functional networks. We also investigated within-subject variability across sessions by computing, for each subject, the standard deviation of the correlation between each parcel-pair across all 10 sessions. Average variability across all correlations showed a pattern of relatively larger variability in visual, somatomotor and dorsal attentional regions, which is in line with analogous studies in healthy subjects. However, in contrast to studies in healthy subjects, variability in these networks was not significantly different from the one observed in executive control areas, which have been traditionally implicated in OCD.

Conclusions: We find that, in patients with OCD, functional networks are dominated by common organizational principles as well as prominent individual features. These results mirror previous investigations in healthy subjects and extend it to patients with OCD highlighting the importance of individualized approaches for studying properties of brain organization. In contrast to previous reports in healthy subjects, variability was similar in processing and executive control regions, setting the stage for relating longitudinal dynamics of brain functions to behavioral and psychiatric symptoms variability.

Keywords: Obsessive-Compulsive Disorder (OCD), Precision Psychiatry, Functional Connectivity

Disclosure: Nothing to disclose.

P419. Resting-State Functional Connectivity Signatures of Obsessive-Compulsive Symptom Profiles

Tracey Shi*, Seonjoo Lee, Nikolaus Kriegeskorte, David Pagliaccio, H. Blair Simpson, Rachel Marsh

Columbia University, New York, New York, United States

Background: Obsessive-compulsive disorder (OCD) is a disabling illness that exhibits bimodal timing of onset, with up to half of cases beginning in childhood. Pediatric subclinical obsessive compulsive (OC) symptoms often increase risk for the development of OCD, but the neural bases and longitudinal trajectory of OC symptoms remain poorly characterized. Capitalizing on the Adolescent Brain Cognitive Development (ABCD) study to perform a well-powered whole-brain analysis, herein we identify "profiles," or groups of OC symptoms with similar neural underpinnings and characterize resting state functional connectivity (rsFC) patterns that correlate with these symptom profiles across the severity spectrum.

Methods: ABCD data was divided into matched training and test subsamples. Participants completed rsFC scans, CBCL, and KSADS. Eight individual items from the CBCL (CBCL-OC Symptom scale) were used as clinical data for the primary analyses. RsFC data were preprocessed, and connectivity matrices were calculated by the DCAN Lab. Participants with at least 10 minutes of motion-free data were included in analyses (training: n = 2,846; ages 8-11; 1,495 female; test: held out from current work). Using only the training subsample, age, sex, race, and head motion were regressed out of rsFC and clinical data ("features"). 501 rsFC features were selected for elastic net canonical correlation analysis (eCCA) based on stable correlations with clinical features across 100 subsamples. ECCA hyperparameters were optimized using cross-validation, eCCA was performed, and significance was assessed via permutation testing. For post-hoc validation, the canonical variates were t-tested for group differences across participants with and without KSADS-diagnosed OCD.

Results: ECCA revealed 8 significant pairs of canonical variates ("profiles"). Canonical variate 1 ($r = .47$, $p = .01$) represented a clinical profile with both obsessions and compulsions, in addition to general symptoms like worries and fears. RsFC dysconnectivity centered around the dorsolateral prefrontal cortex (dlPFC), with symptoms associated with more positive connectivity between task-positive and negative regions and weaker positive connectivity within the task-negative default mode network.

Canonical variate 2 ($r = .39$, $p = .01$) corresponded to obsessions, perfectionism, and worries, but not compulsions. This profile was associated with rsFC features involving motor and sensory areas, including weaker negative connectivity with the insula and weaker positive connectivity within sensory and motor networks.

The 3rd ($r = .33$, $p = .01$) profile focused on perfectionism and fear of doing something bad, specifically without obsessions. As with the first profile, the rsFCs centered around the dlPFC. However, the connections were in the opposite direction: stronger symptoms in this profile were associated with less positive connectivity between task-positive and negative regions and

stronger positive or weaker negative connectivity within the default mode network.

Profile 4 ($r = .36$, $p = .01$) was predominately compulsions without general strange behavior. This was most associated with connectivity involving the frontal eye field in the frontoparietal network, including more positive connectivity with task-negative regions and more negative or weaker positive connectivity with task-positive regions.

In post-hoc analyses, the rsFC patterns for the first 3 profiles significantly differed between participants with and without KSADS-diagnosed OCD ($p < .03$ after Bonferroni correction). Specifically, OCD diagnoses were associated with a rsFC pattern indicating higher OC symptoms according to CBCL scores.

Profiles 5-8 were statistically significant ($r \sim .30$, $p < .02$) but not discussed due to space limitations.

Conclusions: We identified 8 significant "profiles," or sets of OC symptoms and associated patterns of rsFC dysconnectivity. The first 3, derived from quantitative relationships with subclinical to clinical OC symptoms, had clinical validity in that they distinguished children with OCD diagnoses from those without. One possible interpretation of these findings is that profile 1 may represent a more traditional presentation of OC symptoms including both obsessions and compulsions, while profiles 2-4 represent atypical presentations and/or related disorders such as obsessions-predominant, general fears or anxiety, and compulsions-predominant, respectively. The distinct rsFC patterns associated with each clinical profile suggests different underlying network disturbances giving rise to different symptoms, possibly denoting heterogeneity that could predict longitudinal symptom course or responsiveness to interventions or treatment. The rsFC patterns for profiles 1 and 3 center around the dlPFC, a target for transcranial magnetic stimulation for OCD due to its rich connectivity with the cortico-striatal loops that are altered in OCD. Furthermore, rsFC patterns detected herein corroborate prior work showing that an altered balance between task-positive and negative networks may be central to OCD pathophysiology. Future work will include publicly preregistering the findings from this training subsample, assessing replication of the canonical variates in the matched test subsample (held out from present analyses), and evaluating whether these profiles predict longitudinal progression of OC symptoms or response to OCD treatment in a separate sample of participants.

Keywords: Resting State fMRI, Obsessive Compulsive Symptoms, Obsessive Compulsive Disorder

Disclosure: Nothing to disclose.

P420. Imaging Transcriptomics in Obsessive-Compulsive Disorder

Leonardo Saraiva*, João Sato, Roseli Shavitt, Euripedes Miguel, Carolina Cappi

Institute of Psychiatry of the Sao Paulo University Medical School, Sao Paulo, Brazil

Background: Robust genetic and neuroimaging studies have implicated genetic and neural alterations, respectively, in OCD. However, few studies have performed investigations integrating genetics and neuroimaging in the disorder. In this respect, imaging transcriptomics is a pioneering field that explores gene expression signatures underlying imaging-derived phenotypes. This association is performed by using the neurotypical brain-wide gene expression data from the Allen Human Brain Atlas (AHBA), which was mapped in the stereotaxic space. The morphometric similarity network (MSN) is a recently proposed methodology that captures anatomical connectivity between cortical regions with similar cytoarchitecture and gene expression profiles. Imaging transcriptomics studies uncovering genetic and molecular factors underlying abnormalities in MSN characteristics have been performed in major depression, schizophrenia, and neurodevelopmental disorders, but not in OCD. Therefore, this study is the first imaging transcriptomics study using the MSN in OCD. Its central aim is to probe the gene expression signatures, and its biological correlates, underlying neural abnormalities associated with the disorder.

Methods: T1-weighted MRIs of 116 OCD cases and 74 healthy controls were used in this study. These MRIs were processed using the Freesurfer standard automated processing pipeline. A cortical parcellation subdividing the Desikan-Killiany atlas into 308 regions of approximately equal surface area was computed in the native surfaces of the MRIs. Intra-individual MSNs were computed by calculating pairwise Pearson correlations coefficients between vectors of z-scored MRI features (surface area, gray matter volume, cortical thickness, intrinsic curvature and mean curvature) for each pair of cortical regions. Intra-individual regional morphometric similarity (MS) measures were computed for each region by averaging its correlation coefficients with all other regions. OCD case-control differences in regional MS were computed by using linear regression, with sex and age as covariates. Regional brain gene expression data from the AHBA were processed using the abagen toolbox. Partial least squares (PLS) regression was performed to assess the contribution of regional brain gene expression, as measured by the AHBA, to the observed OCD case-control differences in regional MS for the left hemisphere. It should be noted that only the left hemisphere was included in this analysis since the right hemisphere is undersampled in the AHBA dataset. The PLS first component (PLS1) thus represents the linear combination of weighted brain gene expression measures whose spatial distribution mostly contributes to the spatial distribution of observed OCD case-control differences in regional MS for the left hemisphere. Permutation testing was used to assess the statistical significance of the variance explained by the PLS1. Bootstrapping was used for the estimation of z-scores for the PLS1 gene weights. Genes with significant positive and negative contributions to the PLS1 were defined as having z-scores > 3 (PLS1 +) and < -3 (PLS1-), respectively. The R package gProfileR was used to perform enrichment analysis for the PLS1 + and PLS1- genes. Finally, a OCD case-control differential gene expression (DGE) analysis was performed in RNA-sequenced peripheral blood samples of 19 OCD cases and 19 healthy controls. Pairwise Spearman correlation coefficients were computed between the PLS1 + and PLS1- genes z-scores and their respective logarithms of folds changes (logFC) obtained in DGE analyses.

Results: Statistically significant (not corrected for multiple testing) OCD case-control differences in regional MS were found for cortical regions in both hemispheres. In the left hemisphere, these regions include frontal lobe regions (parsopercularis [part1], rostralmiddlefrontal [part4], and precuneus [part6] cortices) and parietal lobe regions (superiorparietal [part5], superiorparietal [part9], and supramarginal [part2] cortices). In the right hemisphere, these regions include frontal lobe regions (fusiform [part4], and inferiorparietal [part3] cortices) and parietal lobe regions (postcentral [part7], and superiorparietal [part1] cortices). The PLS1 explained 16.22% of the variance in the spatial distribution of the observed OCD case-control differences in regional MS, which was statistically significant ($p = 0.035$) in permutation testing. After the estimation of z-scores for the PLS1 gene weights by bootstrapping, 461 PLS1 + genes and 472 PLS1- genes were identified. The PLS1 + genes were mostly enriched for

cerebral cortex in the Human Protein Atlas (HPA) database and for regulation of cellular process in the Gene Ontology Biological Process (GO:BP) database. The PLS1 + genes were mostly enriched for cerebral cortex in the HPA database and for generation of precursor metabolites and energy in the GO:BP database. Finally, the z-scores of the PLS1 + and PLS1- genes were negatively correlated (rho = -0.15 and -0.042, respectively) with their respective logFC obtained in the OCD case-control DGE analysis in peripheral blood. Only the PLS1 + genes correlation was statistically significant ($p = 0.007$).

Conclusions: This study provide compelling evidence that regional brain gene expression profiles contribute to neural abnormalities associated with OCD. Moreover, this study suggests that these brain gene expression profiles are reproduced in peripheral blood.

Keywords: Gene Expression, Imaging-Genetics, Obsessive-Compulsive and Related Disorders, Transcriptomics, Brain Transcription

Disclosure: Nothing to disclose.

P421. Recent Advances in the Genetic Architecture of OCD: Genetic Maternal Effect, Ultrarare, Rare, and Common Variation Contribute to Risk

Behrang Mahjani*, Seulgi Jung, Carolina Cappi, Marina Natividad Avila, Lily Cohen, NORDiC Consortium, EGOS Consortium, James Crowley, Kathryn Roeder, Bernie Devlin, Joseph Buxbaum, Dorothy Grice

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

Background: Obsessive-compulsive disorder (OCD) is a multifactorial disorder, meaning that the risk of OCD is influenced by multiple genetic and other factors. The heritability of OCD, historically estimated by the analysis of twin and family studies, is reported to be 35-50%. Heritability can also be estimated from individuals drawn from a population who have no obvious familial relationships, as long as they have been characterized for genetic variation across their genomes. Usually, this genetic characterization employs genotypes of single nucleotide polymorphisms (SNPs) for which alleles are common in the population. When the heritability of OCD is computed in this manner, estimates range from 25-43%, which mostly arise from covariance due to common genetic variants. Among the environmental factors, different studies have linked maternal conditions before and during pregnancy to the risk of OCD. Such factors could represent maternal effect. Maternal effect is defined as a causal effect of the mother's phenotype on the phenotype of the offspring, and it can be partitioned into genetic maternal effect (maternal genetic nurture) and environmental maternal effect. Genetic maternal effect occurs when the genotype impacting the phenotype of the mother influences the phenotype of the offspring, usually independent of the child genotype. Environmental maternal effect occurs when the environment impacting the phenotype of the mother (independent of genotype) influences the phenotype of the offspring.

Despite these findings, the contribution of inherited genetic variation across the allelic frequency spectrum and role of maternal effects in the risk of OCD remains uncertain and worthy of further study, as it impacts both our understanding of processes underlying OCD risk architecture and rational study design. In our studies, we partitioned the risk architecture of OCD by a common set of latent genetic and environmental factors (e.g., maternal effects). In addition, we identified rare potentially damaging structural variations (e.g., potentially damaging copy number variation; pdCNV) and significant OCD risk genes impacted by rare variation.

Methods: To study the genetic risk architecture of OCD, we studied family data for 822,843 individuals in Sweden [7,184 (0.9%) with OCD], as well as SNP data from a sample of 2,090 Swedish-born individuals diagnosed with OCD and 4,567 controls. Using genotypes of these SNPs to estimate distant familial relationships among individuals, we estimated heritability of OCD, both overall and partitioned according to minor allele frequency (MAF) bins. We used a subsample of these data to evaluate the distribution of pdCNV in OCD, examining associations between pdCNV and the phenotypes of probands, including a consideration of early- vs. late-diagnoses. We used analytical approaches to estimate the number of genes that could increase risk for OCD through de novo mutations and used this estimate for power calculations for sequencing studies. In ongoing studies, these Swedish, and additional, samples are being sequenced for rare association studies.

Results: Using family data, we observed that 35-40% of the liability for OCD is due to direct genetics (heritability), and 7-7.6% due to genetic maternal effect. We observed evidence for substantial assortative mating among individuals with OCD. Additionally, our results demonstrated associations between parental age and maternal smoking during pregnancy with risk of OCD. Using SNP data, we estimated narrow-sense SNP-heritability of 29% (SE = 4%). Contrary to earlier studies, however, SNPs with MAF between 0.01 and 0.05 accounted for 10% of heritability and estimated heritability per bin roughly follows expectations based on the infinitesimal model (where risk is influenced by a large number of loci distributed across the genome and across MAF bins). Nine percent of OCD probands carried a pdCNV, showing that 1 in 11 OCD cases may have a genetic diagnosis from CNV. The most frequent pdCNV found was at the 16p13.11 region. There was no significant difference in pdCNV frequency between early- vs. late-diagnosed OCD probands. Using published data, we estimate that deleterious mutations in about 200 distinct genes can increase risk for OCD, motivating high-throughput sequencing for gene discovery in OCD. Sequencing of 8000 samples is ongoing.

Conclusions: Our results provide new insights into the risk architecture of OCD. We showed that maternal effect, combined with the effects of rare and common variants, shape a large portion of the risk architecture of OCD. We observed that genetic variation across a very broad allelic frequency spectrum influences the risk of OCD. Studies by other groups have shown that ultrarare de novo mutations are also part of OCD risk and we expect to observe this in our ongoing sequencing studies.

Keywords: Obsessive Compulsive Disorder, Heritability, Genetics, Rare Genetic Variation, Copy Number Variants

Disclosure: Nothing to disclose.

P422. De Novo Mutations in Conserved Genes Contribute to OCD Risk

Seulgi Jung, Carolina Cappi, Marina Natividad Avila, NORDIC Consortium, EGOS Consortium, Kathryn Roeder, James Crowley, Behrang Mahjani, Joseph Buxbaum, Dorothy Grice*

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

Background: OCD is a neurodevelopmental psychiatric disorder that affects ~3% of the population. For years, our understanding of the genetic architecture and risk factors for OCD has lagged compared to other major psychiatric disorders. However, as OCD genetic cohorts and sample sizes increase, we have the opportunity to expand our knowledge regarding OCD risk

architecture. Early studies provide convincing evidence for the importance of genetic risk factors in OCD. In family studies, the odds of OCD in case versus control relatives were 4.6 - 5.0 in first-degree relatives, 1.5 – 2.3 in second-degree relatives and 1.4 in third-degree relatives. The heritability of OCD has been estimated at 27-56% in family-based studies and SNP-based heritability at 25-43%. While earlier studies suggested that inherited risk variation was skewed toward the most common variants, our group recently showed that variants across the allelic spectrum all contribute meaningfully to the heritability of OCD. Early and recent studies of copy number variation (CNV) supported a role for rare de novo mutations in OCD. Taken together, these findings suggest that additional inherited and rare variants contribute to risk for OCD. Indeed, whole-exome sequence (WES) studies using OCD cases support the role for rare single nucleotide variation (SNV) in risk for OCD, including de novo mutations. Cappi et al. observed that rates of de novo mutations, likely protein-truncating variation (PTV), are significantly elevated in OCD trios. Similarly, Halvorsen et al. observed a 1.3-fold increase in damaging de novo variants in OCD cases relative to controls, including a 2.6-fold increase in de novo PTV in constrained genes. To identify genes that increase risk for OCD when harboring rare deleterious mutations, we are sequencing 8000 samples, while integrating data from existing and emerging OCD WES studies. Currently, we are analyzing published and unpublished data from OCD cases, with parental or ancestry-matched controls.

Methods: To date, we have new WES data including 160 cases from Mount Sinai and 480 Swedish OCD cases, together with over 2,800 ancestry-matched controls. We also analyzed previously published WES data. For calling SNV and indel variants from WES data, we used the current Genome Analysis Toolkit (GATK) protocol. We performed quality control steps, including checking the accuracy of the reported pedigree information (samples with a discrepancy are dropped) using Hail (an open-source framework for scalable genetic data analysis). Variants were annotated with the Variant Effect Predictor (VEP), prioritizing coding canonical transcripts. We then performed genetic analyses using the Transmitted and De novo Association (TADA) model for gene discovery.

Results: All genes were ranked by P value, and constraint scores were noted, including constraint for PTV and missense variation in the TADA analysis. The top 10 genes were CHD8, INO80, SCUBE1, ZMYM2, ADIPOR2, EIF3G, TP53BP2, KMT2B, PITPNM2, and TRRAP. Three potentially novel OCD genes - EIF3G, KMT2B, and TRRAP, are already known neurodevelopmental genes. (1) EIF3G, with deleterious missense variation in Cappi et al. and in a Mount Sinai sample, is also a likely autism risk gene. (2) Cappi et al report a de novo PTV in KMT2B, and we observe 1 missense with MPC > 2 in a Swedish case. Interestingly, a de novo missense mutation in KMT2B was also observed in a Chinese case (Lin et al.). KMT2B codes for lysine methyltransferase 2B. Interstitial deletions or pathogenic mutations of KMT2B cause a dystonia syndrome, which onsets in the first or second decade of life and can include psychiatric comorbidities such as mild intellectual disability (ID), attention-deficit/hyperactivity disorder (ADHD), anxiety, and OCD. (3) TRRAP encodes a large multidomain protein of the phosphoinositide 3-kinase-related kinases family with de novo mutations reported in autism, ID, schizophrenia, and early-onset psychosis with OCD. Additionally, based on parameters such as de novo mutation rates, and relative risk ratios estimated from the data underlying these studies, we estimate that roughly 200 OCD genes can ultimately be discovered using these approaches.

Conclusions: As seen in ID and autism, de novo mutations in OCD are strongly enriched in highly conserved genes. This represents less than 10% of all genes, when comparing individuals with OCD to individuals without a known psychiatric phenotype. Leveraging this, we are beginning to discover genes that increase risk for OCD, when these genes are impacted by rare deleterious variation. We are also seeing overlap with known ASD and ID risk genes, as well as interesting associations with known syndromal genes.

Keywords: OCD, Genetics, Rare Variation

Disclosure: Nothing to disclose.

P423. Interaction of Excitatory Neuromodulation and Perceptual Modification of Self-Images in Body Dysmorphic Disorder

Wan-wa Wong, Rangaprakash Deshpande, Reza Tadayon-Nejad, Joel Diaz, Andrew Leuchter, Gerhard Hellemann, Jamie Feusner*

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

Background: Body dysmorphic disorder (BDD) is marked by preoccupation with misperceived appearance flaws, which they believe render them ugly and disfigured. Disturbances of visual information processing in BDD seem to be core neurobiological contributors to the psychopathological feature of perceptual appearance distortions. Previous neuroimaging studies have found abnormally reduced dorsal visual stream (DVS) activity in BDD when viewing filtered images that conveyed configural/holistic information, contributing to a model of imbalances in global vs. local visual processing. Some evidence has shown that magnocellular pathways in the DVS are tuned to rapid presentation of images, thereby allowing global/holistic, but not local/detailed, visual processing. Moreover, ventral visual stream (VVS) regions seem to reduce activation magnitude with higher stimulus frequency/shorter stimulus duration. Accordingly, we investigated if DVS/VVS systems in individuals with BDD could be altered during rapid face presentation. In addition, we tested if intermittent theta-burst stimulation (iTBS), a form of repetitive transcranial magnetic stimulation, could further enhance the effects of rapid face presentation, as quantified through dynamic effective connectivity (DEC) modeling.

Methods: Fourteen unmedicated adults with BDD, were randomly assigned to receive active (n = 7) or sham (n = 7) iTBS. Stimulation targets were left and right lateral parietal cortices, (corresponding to CP3 and CP4 respectively, on the EEG 10-10 system). Stimulation was applied at 100% active motor threshold (AMT) for the active group and 10% AMT for sham. We then immediately acquired fMRI data from participants while they viewed photos of their own faces for short (125 ms, 250 ms, 500 ms) and long (3000 ms) durations. As an additional comparison, given the possibility of sham stimulation effects, the data were also compared to the data collected separately from 38 BDD participants and 29 healthy controls during an identical task of face viewing, but without iTBS beforehand. The BDD participants met DSM-5 criteria for BDD with face concerns.

Fourteen regions of interest (ROIs) in DVS and VVS were selected: 2 ROIs in primary visual cortex (V1) [bilateral calcarine], 6 ROIs in VVS [bilateral inferior occipital gyrus (IOG), fusiform gyrus (FG), and inferior temporal gyrus (ITG)], and 6 ROIs in DVS [bilateral superior occipital gyrus (SOG), inferior parietal gyrus (IPG), and superior parietal gyrus (SPG)]. Hemodynamic deconvolution was then performed on the timeseries extracted from these ROIs to minimize intra-subject hemodynamic response function variability, and to improve estimation of effective connectivity. DEC, a dynamic measure of directional connectivity between pairs of ROIs, was computed at each time point using Kalman-filter based time-varying Granger causality (GC). Twelve intra-hemispheric connections were chosen and divided into 4 categories: 1) VVS Lower (Calcarine to IOG), 2) VVS Higher (IOG to FG; IOG to ITG), 3) DVS Lower (Calcarine to SOG), and 4) DVS Higher (SOG to IPG; SOG

to SPG). Linear mixed model was used to analyze the data (fixed factors: group [BDDActive_iTBS, BDDSham_iTBS, BDDNo_iTBS or HCNo_iTBS], duration [125 ms, 250 ms, 500 ms or 3000 ms], category [VVS Lower, VVS Higher, DVS Lower or DVS Higher]; random factor: participant).

Results: For positive GC values, there was significant three-way interaction between group, duration and category from tests of fixed effects ($F[27, 135614]=1.89$, $p = 0.004$). For DVSHigher, the active group exhibited stronger DEC than the BDD without iTBS across the four durations, and they achieved similar levels as the controls.

Conclusions: In this proof-of-concept study, we explored the effects of interactions between excitatory neuromodulation and rapid face presentation on dorsal visual stream and ventral visual stream connectivity in BDD. Excitatory neuromodulation induced by iTBS enhanced dynamic connectivity for DVSHigher, achieving similar levels as the healthy controls. These results, showing target engagement and modulation, have implications for designing novel perceptual retraining treatments to remediate perceptual distortions of appearance in those with BDD.

Keywords: Body Dysmorphic Disorder, Repetitive Transcranial Magnetic Stimulation (rTMS), Visual Perception, Effective Connectivity, Perceptual Distortion of Appearance

Disclosure: Nothing to disclose.

P424. The Psychosocial and Educational Burden of Obsessive-Compulsive Disorder in Youth

McKenzie Schuyler*, Bowie Duncan, Daniel Geller, Amitai Abramovitch

Massachusetts General Hospital, Boston, Massachusetts, United States

Background: Obsessive-compulsive disorder (OCD) is associated with significant multi-domain impairment. Indeed, OCD is associated with poorer long-term educational attainment, vocational problems, and cognitive dysfunction. Compared to adults, youth with OCD exhibit relatively better cognitive functioning. Nevertheless, they may struggle more in family and peer relationships and may require more assistance in school settings compared to healthy controls. Only a few studies have directly examined these issues. Furthermore, research investigating the impact of comorbidities on psychosocial functioning in pediatric OCD is limited. The goal of the present study was to directly assess psychosocial functioning in a large, well-characterized sample of children and adolescents with OCD.

Methods: A sample of 100 children and adolescents with OCD (Mage = 11.42; 42.0% male) and 138 non-OCD controls (Mage = 11.45; 42.8% male) participated in the study. Participants were administered a battery of measures including the Kiddie Schedule for Affective Disorders and Schizophrenia (KSADS) to assess diagnostic status and comorbidities, the Children's Yale-Brown Obsessive Compulsive Scale (CY-BOCS) to assess OC symptom severity, the Social Adjustment Inventory for Children and Adolescents (SAICA) to assess psychosocial adjustment, and the Family Environment Scale (FES) to assess family functioning. In addition, information regarding repeated grades, special placement classes and extra help/tutoring was obtained from teachers and parents. We conducted independent samples t-tests to examine group differences in psychosocial, family, and school functioning variables.

Results: Compared to controls, youth with OCD had significantly higher levels of school behavior problems ($p = .001$; Cohen's $d = .43$), problems with spare time activities ($p = .000$; $d = .89$, problems with peer activities ($p = .000$; $d = .71$), and problems with parents ($p = .001$; $d = .49$). Compared to controls, families of youth with OCD also had significantly elevated current and past family conflict scores ($p = .035$, $d = .28$ for both), and reduced current and past family cohesion scores ($p = .000$, $d = .51$; $p = .018$, $d = .32$). There were no differences in the family expression subscale. Significantly more youth with OCD attended special classes (0.7% vs. 4.3%, $p < .0001$) and received extra help (10.2% vs. 64%, $p < .0001$) compared to controls. There were no differences in rates of repeated grades. OCD severity was significantly correlated with attending special classes and with several SAICA subscales. However, it was not correlated with FES subscales. Finally, comorbidity with attention deficit hyperactivity disorder, conduct disorder, and oppositional defiant disorder resulted in a different pattern of difficulty, primarily in the domain of school behavior problems.

Conclusions: Although pediatric OCD is not associated with substantial cognitive impairments, this population receives more extra help in school, exhibits a higher frequency of enrolling in special classes, and displays significantly more psychosocial and family problems than controls. In addition, there is a differential impact of several comorbid disorders on psychosocial problems in this population. These results highlight the need for careful screening for OCD in this age group, including assessment of comorbid conditions. More research is needed to examine how reassurance-seeking versus measurable academic deficits contribute to help-seeking in this population.

Keywords: Obsessive-Compulsive Disorder (OCD), Pediatric, Family Study

Disclosure: Nothing to disclose.

P425. Tripping Mice and Stoned Fish: Head Twitch Response (HTR) and Behavioral Phenotypic Evidence of Effect Differences Between Synthetic Psilocybin and Psychedelic Mushroom Extract

Leonard Lerer*, Alexander Botvinnik, Katherine Spear, Orr Shahar, Patryk Lipski, Haley Calderon, Karin Blakolmer, Tzuri Lifschytz, Bernard Lerer

Back of the Yards Algae Sciences, Chicago, Illinois, United States

Background: Anecdotal reports suggest that the behavioral and pharmacological effects of psilocybin-containing, "full spectrum" psychedelic mushroom extract (FSME) differ from those of chemical psilocybin (PSIL) in their nature and intensity. A limited number of rodent studies have compared synthetic psilocybin (or psilocin) with crude psychedelic mushroom extract. Furthermore, psychedelic mushrooms contain intermediate products of the psilocybin biosynthetic pathway such as baeocystin, norbaeocystin and aeruginascin that may influence the nature of the effect of psilocybin ("entourage effect") along with other components such as harmines with monoamine oxidase inhibiting properties. In the current study, we compared the effect of PSIL to that of FSME on the mouse head twitch response (HTR), which is correlated with psychedelic effects in humans, on a rodent screening test for antidepressant effect and in a behavioral phenotypic zebrafish model.

Methods: Male C57Bl/6j mice were used in all head twitch experiments. PSIL (98.75% purity) was provided by Usona Institute. FSME, a methanol extract of Psilocybe cubensis with a psilocybin content of 1.5%, was provided by BYAS-PEB. Drug doses were calculated so that equal injection volumes of PSIL and FSME contained equal concentrations of psilocybin on a mg per kg basis. Control mice received vehicle (VEH) injections (0.9% NaCl solution). HTR was measured over 20 minutes in a magnetometer-based system using ear clip magnets. The tail suspension test (TST) was conducted using a Noldus Ethovision system by observers blind to treatment status, 48 hours after

drug administration. Individual male zebrafish (Danio rerio) were used in an open arena, behavioral phenotyping experiment. The drug dose, 3 mg/L of PSIL and FSME was administered in a beaker containing 200 ml of water over 10 minutes. Control fish were placed in a beaker containing 200 ml of water for 10 minutes. The fish were then placed in an 50x50x4cm arena and video-tracked using idTracker software, with trajectories recorded for 20 minutes immediately after treatment and for 20 minutes at 80 minutes after treatment.

Results: FSME induced a significantly greater number of head twitches over 20 minutes at a psilocybin dose of 4.4 mg/kg than PSIL at the same dose ($F = 4.41$, df 1,21, $p = 0.04$; FSME $n = 11$, PSIL $n = 12$). The difference was evident over the entire time course. On the tail suspension test (TST) conducted 48 hours after i.p. administration of PSIL (4.4 mg/kg), FSME (psilocybin content 4.4. mg/kg) or VEH, both PSIL (209.3 ± 93.6 sec, $n = 11$, $p = 0.01$) and FSME (198.14 ± 61.3 sec, $n = 10$, $p = 0.0006$) showed significantly less inactivity than VEH (296.0 ± 33.8 sec, $n = 11$). On the highly active measure of the TST, FSME (11.38 ± 10.13 sec, $n = 10$) induced significantly more activity than both VEH (0.04 ± 0.12, $n = 11$, $p = 0.0007$) and PSIL (0.53 ± 1.04 sec, $n = 11$, $p = 0.001$). In the zebrafish experiment, during the first 20 minute recording period, 2D spatiotemporal reconstructions of the zebrafish swim paths demonstrated clearly visible differences in swimming patterns including velocity, distance swum, average distance to perimeter and middle and changes in direction, between the control and the FSME/PSIL groups. There were also a number of clear differences in swimming patterns between the FSME and PSIL groups, especially related to the mean time spent in the corners of the arena ($P < 0.05$; FSME/PSIL $n = 9$). At 80 minutes, the swimming pattern of the PSIL group closely resembled that of the control group, whereas the FSME group continued to show a similar, slightly attenuated swimming pattern to that observed in the initial 20-minute recording period.

Conclusions: A prior study by Zhuk et al (2015) suggested that mushroom extract has greater potency in inducing HTR than psilocin (the active metabolite of psilocybin). Our findings in mice are in accordance with this observation. We have further shown that on the TST, a screening test for antidepressant potential, FSME induces a stronger effect than PSIL when the same dose of psilocybin is administered with both preparations. Furthermore, this work provides evidence of a robust and measurable zebrafish response to PSIL and FSME. The more sustained effect of FSME may be indicative of an "entourage effect". Further studies are indicated to elucidate a possible therapeutic advantages of full spectrum psychedelic mushroom extract as compared to chemical psilocybin and to identify the entourage molecules that contribute to this effect. This work also raises a tantalizing and important challenge related to the possible development of a zebrafish behavioral model equivalent to the mouse HTR test.

Keywords: Psilocybin, Psychedelics, Entourage Effect, Head Twitch Response, Tail Suspension Test

Disclosure: Back of the Yards Algae Sciences: Founder (Self)

P426. Translational Implications of Marble Burying in ICR Mice for the Anti-Obsessional Effects of Psilocybin

Sandeep Singh, Alexander Botvinnik, Orr Shahar, Gilly Wolf, Amit Lotan, Bernard Lerer*, Tzuri Lifschytz

Hadassah-Hebrew University Medical Center, Jerusalem, Israel

Background: Initial clinical findings, supported by preclinical studies employing behavioral paradigms such as marble-burying, suggest that the psychedelic tryptamine, psilocybin, may be effective in treating obsessive-compulsive disorder (OCD) The aims of this study were to further evaluate the role of 5-HT2A receptors in the effect of psilocybin on marble-burying, to explore the role of 5-HT1A receptors in this effect and to examine potential use the 5-HT1A receptor partial agonist, buspirone, as a concurrent treatment for OCD with psilocybin

Methods: Male ICR mice were administered psilocybin 4.4 mg/kg, escitalopram 5 mg/kg; the 5-HT1A agonist, 8-OH-DPAT 2 mg/kg; the 5-HT2A antagonist, M100907 (volanserin) 2 mg/kg; the 5-HT1A partial agonist, buspirone, 5 mg/kg; or the 5-HT1A antagonist, WAY100635 2 mg/kg; or combinations. Drugs were administered intraperitoneally, and the mice were tested on the marble burying test (MBT) for 30 minutes after treatment. Head witch response (HTR) induced by psilocybin alone or in combination with buspirone, was examined in a magnetometer-based assay.

Results: 1) Both psilocybin ($p < 0.01$) and the positive control, escitalopram ($p < 0.01$), significantly reduced marble-burying. The effect of psilocybin was not attenuated by the 5-HT2A antagonist, M100907. The 5-HT1A agonist, 8-OH-DPAT, reduced marble-burying ($p < 0.01$) as did the 5-HT1A partial agonist, buspirone ($p < 0.01$). The effect of 8-OH-DPAT was additive to that of psilocybin ($p < 0.01$) but that of buspirone was not. The 5-HT1A antagonist, WAY100635, attenuated the effect of 8-OH-DPAT and buspirone on marble burying but not the effect of psilocybin. 2) Psilocybin injections over 3.5 hours had no effect on marble-burying and the effect of bolus injection was not persistent. 3) Co-administration of buspirone with psilocybin blocked the effect of psilocybin on HTR but not its effect on marble burying.

Conclusions: Neither 5-HT2A nor 5-HT1A receptors are pivotally implicated in the effect of psilocybin on marble-burying. Co-administration with buspirone may block the psychedelic effects of psilocybin without impeding its anti-obsessional effects. Concurrent treatment of OCD with buspirone and psilocybin is a feasible strategy to achieve anti-obsessional effects while avoiding or minimizing the psychedelic trip.

(Supported in part by Back of the Yards algae sciences and Parow Entheobiosciences)

Keywords: Psychedelics, Obsessive-Compulsive Disorder (OCD), Psilocybin, Buspirone, 5-HT1A Receptors

Disclosure: Back of the Yards Algae Sciences: Contracted Research (Self), Parow Entheobiosciences: Contracted Research (Self), Negev Capital: Consultant (Self)

P427. Translating the Persistent Avoidance Circuitry: Implications for Obsessive Compulsive Disorder

Lucas Remoaldo Trambaiolli*, Freddyson J. Martínez-Rivera, Wei Tang, Mary Phillips, Gregory Quirk, Suzanne Haber

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

Background: The caudal area 47/12 of the ventrolateral prefrontal cortex (vlPFC) was recently described as a node of the salience network in humans and nonhuman primates (NHP) (Trambaiolli et al. 2022 eLife). This region supports stimulus-outcome predictions associated with salient stimuli and prepares appropriate responses later selected by the anterior cingulate cortex (ACC).

In the rodent persistent avoidance circuit, the agranular insular/lateral orbital (AI/LO) cortex has excitatory inputs to the rostral prelimbic (rPL) cortex (Martínez-Rivera et al. 2022 Biol. Psych.). These neurons project to the ventral striatum (VS) to control avoidance expression. Hypoactivity in AI/LO decreases excitatory drive from rPL to the VS, increasing VS susceptibility to basolateral amygdala (BLA) excitatory inputs. This drives avoidance expression.

Herein, we hypothesize that (i) the primate caudal 47/12 includes the AI/LO region identified in rats and (ii) a similar avoidance circuit can be identified in NHP using anatomical tract-tracer data starting from caudal 47/12.

Methods: We injected bidirectional tracers in the vlPFC area 47/12 (four injection sites) and the ACC (two injections in area 32 and three in area 24), anterograde tracers in the BLA (two injections), and retrograde tracers in the VS (two injections).

We used bright-field microscopy to identify which sublocations in the vlPFC and ACC are highly interconnected. We quantified the number of cells labeled in 23 cytoarchitectonic regions in the frontal cortex and amygdala. The connectivity strength between each cortical area and the injection location was measured as the ratio between the number of cells in this area by the total number of labeled cells.

We used dark-field microscopy to delineate axonal projection zones between all nodes of the avoidance circuit (vlPFC, ACC, VS, and BLA), starting from the locations identified in the previous step. Finally, we validated the connectivity strength of projections from vlPFC, ACC, and BLA to the VS using the same retrograde approach described before.

Results: Area 24 was the ACC portion with the strongest projections to the vlPFC, showing a rostro-caudal gradient peaking at the caudal-most part of 47/12. The strength of connections from the vlPFC to the ACC peaked in the pregenual-genu portion of area 24 (rostral 24). Dark-field analysis validates the bidirectional projections between caudal 47/12 and rostral 24, with dense axon terminal fields in both locations.

Rostral area 24 has dense projections to the VS (core extending to the shell) and BLA. Other ACC regions and the caudal 47/12 present weak or no projections to these regions. The BLA projects densely to the VS (mainly the shell), vlPFC, and ACC. Importantly, BLA and rostral 24 projections overlap in the border between the VS shell and core. Retrograde injections in the VS validate these patterns.

Conclusions: These findings suggest that the anatomical connections relevant to the persistent avoidance circuitry in rodents are also present in NHP. The NHP caudal 47/12 likely includes this circuit's AI/LO node, and rostral 24 is thought to be homologous to the rPL. Caudal 47/12 has weak-to-absent projections to the subcortical regions of interest, focusing its control over the circuit on interactions with the rostral ACC.

Abnormal computations in the caudal 47/12 could lead to pathological behaviors, such as the persistent avoidance observed in patients with obsessive-compulsive disorder (OCD). Importantly, caudal 47/12 receives dense projections from the BLA, which may cause biased attention to negative stimuli and impaired response preparation. This is in line with recent studies reporting that OCD patients have hyperconnectivity between the caudal 47/12 and the amygdala compared to healthy controls (Thorsen et al. 2020 Neuroim. Clin.). This connection could also be associated with the hypoactivity in AI/LO observed in avoidant rodents (Martínez-Rivera et al. 2022 Biol. Psych.) or in caudal 47/12 from OCD patients (Chase et al. 2020 Neuroim. Clin.).

The VS is a downstream region where avoidance behaviors are triggered or stopped. Rostral 24 and BLA converge to the same VS location (border between the shell and core). This region is a common target for deep-brain stimulation in patients with OCD, with rodent models suggesting its effectiveness is due to enhancing fear extinction (Rodriguez-Romaguera et al. 2020 PNAS). Given its role in the avoidance circuit and the homologies between NHPs and humans, caudal 47/12 can be an additional target within this circuit for treating these symptoms in OCD.

Keywords: Ventrolateral Prefrontal Cortex, Persistent Avoidance, Obsessive Compulsive Disorder, Anterior Cingulate Cortex (ACC), Ventral Striatum

Disclosure: Nothing to disclose.

P428. Dorsal Striatum Modulates the Expression of Uncertain, but Not Certain, Value-Based Behaviors

Matthew Geramita*, Susanne Ahmari, Eric Yttri

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

Background: Patients with obsessive compulsive disorder (OCD) display deficits in making decisions based on both value and certainty. The dorsomedial striatum (DMS) likely plays a critical role in these disruptions as the DMS encodes both certainty and value and displays abnormal activity in patients with OCD. Yet it is unknown how DMS representations of value and uncertainty contribute to behavior. To answer this question, we recorded and manipulated DMS activity during a Pavlovian conditioning task in which value and uncertainty were modulated.

Methods: Mice ($n = 10$; 5 female) expressing either channelrhodopsin-2 in either direct or indirect pathway medium spiny neurons (dMSNs or iMSNs) were implanted with bilateral optical fibers in the DMS (anteroposterior: +0.7 mm, mediolateral: +/-1.5 mm, dorsoventral: −2.5 mm) and trained to perform a head-fixed auditory Pavlovian conditioning task in which three cue types predicted reward on 0%, 50% or 100% of trials. To manipulate activity, dMSNs or iMSNs were briefly stimulated bilaterally (473 nm; 2 mW; 15 Hz for 1 s) during either the cue, delay, or outcome period. In a separate cohort of animals ($n = 3$; 1 female), MSN activity was recorded using Neuropixels electrodes.

Results: Mice learned the value of each auditory stimulus as the rate of anticipatory licking during the cue and delay period correlated with reward probability (0%: 1.2 ± 0.1 Hz; 50%: 4.4 ± 0.2 Hz; 100% 5.1 ± 0.1 Hz; One-way ANOVA, $p < 10\text{-}32$). In a population of 343 putative MSNs, 51% were modulated equally by all three cues (ie salience), 40% were modulated by the value of the cue, and 8% were modulated by the certainty of the cue. Unsupervised clustering of activity during the cue and delay periods indicated four distinct response patterns – two that were positively modulated and two that were negatively modulated by cues. Value, salience, and certainty neurons were distributed equally across all four clusters. Interestingly, cue activity from neurons belonging to the two excitatory clusters was negatively correlated with licking for 50% cues (Cluster 1: $r = -0.091 \pm 0.01$, $p < 10\text{-}8$; Cluster 2: $r = -0.085 \pm 0.01$; $p < 10\text{-}11$), but not 0% or 100% cues. These data suggest that a subpopulation of DMS MSNs negatively modulated anticipatory licking during uncertain cues regardless of whether neurons encoded information about cue value, salience, or certainty. Cell-type specific modulation of MSNs corroborated in vivo data. Photostimulation of either dMSNs or iMSNs during the cue period of the 50% cue decreased anticipatory licking (No stim: 4.4 ± 0.3 Hz; vs Stim: 2.8 ± 0.3 Hz; t-test, $p < 0.001$). Surprisingly, there was no change in anticipatory licking when dMSNs or iMSNs were stimulated during the 100% or 0% cue. Additionally, photostimulation of either population during the delay or outcome period of any cue type did not alter licking.

Conclusions: The DMS plays an important role in decision-making in a variety of contexts yet how value and certainty regulate value-based behavior is unknown. Here, using in vivo electrophysiology and optogenetics, we find that both DMS dMSN and iMSN activity inhibits anticipatory licking during uncertain, but not certain, cues. These findings suggest that the certainty of reward gates the influence of DMS activity on behavior.

Keywords: Dorsal Striatum, Uncertainty, Value-Based Decision-Making

Disclosure: Nothing to disclose.

P429. Approach to Use Data of Diverse Ancestries to Uncover Generalizable Genotype Driven Fetal- and Adult-Specific Brain Transcriptomic Mechanisms in Psychiatric Disorders

Aarti Jajoo, Christos Chatzinakos, Cindy Wen, Tade Souaiaia, Tim Bigdeli, Michael Gandal, Nikolaos Daskalakis*

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

Background: Brain transcriptomic imputation utilizes genotype-expression reference panels to build predictive models of genetically regulated gene expression (GReX). GReX models can be applied to GWAS to conduct transcriptome-wide association studies (TWAS) to prioritize gene-trait associations (GTAs) with functional importance. However, reference panels and GWAS studies have been biased toward European Ancestry (EA) samples, posing limitations for the identification of generalizable GTAs. To overcome this challenge, we propose meta-analyzing TWAS results across the ancestries by applying ancestry-specific models to the respective ancestry GWAS.

Methods: To this effect, we trained ancestry-specific GReX models, of Admixed African Ancestry (AA) and EA, separately for healthy adult DLPFC tissue (NAA = 165, NEA = 453 subjects) and fetal brain tissue (NAA = 164, NEA = 292).

Results: Fetal models predict fewer genes and have lower R2 model performance for overlapping genes compared to same ancestry adult models (R2 = 0.17 for EA_adult vs R2 = .11 for EA_fetal, P < e-16). Within each tissue type, R2 was comparable between ancestries, but AA models predicted 45% fewer genes compared to the same tissue EA models likely due to less power. EA models compared to AA models performed worse in the AA test set, while AA models performed comparably in EA test set. We applied these models to GWAS of bipolar disorder (BD), major depressive disorder (MDD), post-traumatic stress disorder (PTSD), and schizophrenia (SCZ), using at least one GWAS for each ancestry within each disorder. The average correlation of GTA z-scores was Rho=0.49 between adult and fetal analyses. Fetal tissue yielded a higher percentage of GTAs for SCZ which was expected because of neurodevelopmental origins, but surprisingly this was true for PTSD which has a later age of onset and lower heritability. The common GTAs from fetal and adult analyses tended to cluster in particular genomic regions (e.g, 17q21.31 in PTSD). Furthermore, the general observation is that bi-ancestry TWAS meta-analysis using just EA GReX models yields the least percentage of Bonferroni significant GTAs while using ancestry-specific GReX models for respective ancestry GWAS yields the highest percentage (e.g., 2x increase in SCZ, 30x increase in PTSD).

Conclusions: Our work shows that fetal and adult brain tissue-based GTAs reveal shared and distinct genetic underpinnings of psychiatric disorders that operate in multiple stages, and that bi-ancestral TWAS is more beneficial when ancestry-specific GReX models can be applied to respective ancestry-specific GWAS.

Keywords: Genetic Ancestry, Gene Expression, Fetal Brain, Adult Brain

Disclosure: Nothing to disclose.

P430. Chemogenetic Inhibition of VTA Gaba Neurons Lead to Persistent but Sexually Diergic Changes in Reactivity to Ethanol

Laverne Melón*, Han Bin Kwon, Isabella Ahearn, Bilge Buyukdemirtas

Wesleyan University, Middletown, Connecticut, United States

Background: Sensitivity to alcohol's stimulant effects predicts diagnosis with AUD and symptom severity (King et al., 2021). Preclinical models of repeated alcohol exposure that sensitizes this stimulant effect are useful tools for understanding maladaptive plasticity underlying the transition from use to disordered use. Moreover, sex differences in these preclinical models may be leveraged to better dissect which biological systems altered by alcohol sensitization are relevant for specific drug's consequences. To this end, we used a chemogenetic approach to dissect a role for GABAergic inhibition in the VTA in the expression of sensitization and the sex-specific reduction in preference for social reward that follows alcohol-induced sensitization.

Methods: Male and female C57BL6/J mice (5-8/group) were administered alcohol (2.0 g/kg) repeatedly for 14 days, with locomotor activity assessed on the first and final days of exposure. Animals were given an opportunity to interact with a sexually-immature juvenile for 5 minutes during early (1 day following last exposure) or protracted (14 days following last exposure) withdrawal. Brains and bloods were harvested 30 minutes following this social interaction opportunity.

To probe the role that GABAergic inhibition in the ventral tegmental area plays in this sex difference, male and female VGAT-cre mice (7-9/group) received 250 nL of the Gi-DREADD virus (pAAV-hSyn-DIO-hM4D(Gi)-mCherry) or the sham mCherry virus (pAAV-hSyn-DIO-mCherry) into the VTA. Animals received a single administration of CNO (1.0 mg/kg, ip) and were tested 7 or 14 days later for their reactivity to alcohol (2.0 g/kg).

Results: Both males and females demonstrated a sensitized response to ethanol at this dose, though the effect was more pronounced for females. Although both groups showed no change in preference for social interaction during early withdrawal from this sensitization protocol, females show a significant reduction in social preference two weeks after their final exposure to ethanol. We found that a singular inhibition of VTA GABA neurons induced heightened baseline activity seven days later in both males and females. However, two weeks following this acute chemogenetic inhibition of VTA GABA neurons only males display significantly greater locomotor response to ethanol (p = 0.014). Current analysis of the effect that this manipulation has on sensitivity to social interaction is currently underway.

Conclusions: The results demonstrate that acute inhibition of VTA GABAergic neurons has a sexually diergic effect on later reactivity to ethanol and this may underlie sex-specific plasticity in behaviors following exposure to the drug.

Keywords: Alcohol, Sensitization, GABA, VTA, Sex Difference

Disclosure: Sage Therapeutics: Grant (Self)

P431. A Human Laboratory Study on the Effects of a Novel Ghrelin Receptor Inverse Agonist Competitive Antagonist on Alcohol and Food-Related Behaviors in Individuals With Alcohol Use Disorder

Monica Faulkner*, Mehdi Farokhnia, Lisa Farinelli, Sara Deschaine, Brittney Browning, Fatemeh Akhlaghi, Lorenzo Leggio

National Institute on Drug Abuse, Baltimore, Maryland, United States

Background: Ghrelin is a 28 -amino acid peptide that acts as the endogenous ligand for the growth hormone secretagogue receptor (GHS-R1a). In preclinical studies, ghrelin has also been found to increase ethanol intake by modulating the neural mechanism of reward processing. Clinical studies have also demonstrated the role of ghrelin in alcohol use behaviors by showing that ghrelin levels are suppressed by both oral and intravenous acute alcohol administration and that baseline ghrelin levels are positively correlated with craving for alcohol. In human

laboratory studies, intravenous ghrelin administration significantly increased cue alcohol craving, and self-administration of alcohol and influenced increases in neural activation of the amygdala and medial orbitofrontal cortex during an alcohol related incentive delay task in non-treatment seeking individuals with AUD.

Recently, we investigated of the safety and tolerability of a novel ghrelin receptor inverse agonist/competitive antagonist, PF-5190457, when it is co-administered with alcohol. While our focus was on safety, we conducted a preliminary investigation on the drug's effect on alcohol cue elicited craving in a small subset of the study and found it reduced alcohol cue elicited craving in a bar like laboratory setting. Based on these preliminary findings, we expanded our investigations on PF-5190457 to specifically assess the drug's effects on alcohol- and food-related behaviors in an inpatient sample of treatment-seeking individuals with AUD.

Methods: The study was a within subjects, double blinded, placebo controlled clinical trial, comparing the effects of PF-5190457 100 mg b.i.d. up to steady state on alcohol/food-related behaviors. Participants were twenty-nine treatment-seeking, detoxified individuals with AUD (M = 21, F = 8). Participants completed 2 behavioral paradigms to 1.) assess whether the drug would reduce alcohol and food cue-elicited craving assessed in a "bar-like" laboratory and 2.) to assess whether the drug would reduce food choices in a "virtual buffet" conducted in a virtual reality environment. To assess alcohol and food cue-elicited craving, participants were exposed to their preferred alcohol-containing beverage and snack in a bar like laboratory. The procedure began with a baseline assessment of alcohol craving, measured by the Alcohol Urge Questionnaire (AUQ), and food craving, measured by the General Food Craving Questionnaire-State (GFCQ-S) outside of the bar like laboratory. After the baseline assessments, participants were escorted into the bar and exposed to alcohol, food, and neutral (water) cues. There was 1 exposure to the neutral cue (water), 1 exposure to the food cue, (their preferred snack), and 2 exposures to the alcohol cue, (their preferred alcohol beverage). After each exposure, participants completed the Alcohol Urge Questionnaire (AUQ) to assess craving for alcohol, General Food Craving Questionnaire-State (GFCQ-S) to assess craving for food, and one additional questionnaire after each alcohol exposure, the Alcohol Attention Scale (AAS), assess their attention to the alcohol-containing beverage.

To assess food choice behavior, participants engaged in a virtual lunch buffet where they chose as many and as much of the virtual food and beverages during one trip to the buffet as they would normally choose during lunch and then completed questionnaires to assesses the current state of alcohol craving (AUQ), food craving (GFCQ-S), and affective mood state (POMS). Participant's food choice behavior was assessed by calculating the total calories selected during the virtual lunch buffet.

Repeated measures ANOVAs were used to analyze the results of the outcomes of the study and all analysis controlled for body mass index (BMI), acyl-ghrelin levels, and age on day 1 of the study as well as gender and experimental condition order.

Results: The drug did not reduce alcohol cue-elicited craving as measured by the AUQ ($F_{1,5} = .11$, $p = .738$) and there was no drug × time interaction ($F_{1,5} = .72$, $p = 0.607$). There was a significant effect of the drug on attention to alcohol, with participants reporting paying less attention to the smell of alcohol ($F_{1,31} = 4.88$, $p = 0.035$) and having to exert less effort to stop thinking about drinking the alcohol ($F_{1, 46} = 5.94$ $p = 0.02$).

There was also a significant effect of the drug on food choice behavior, with participants selecting fewer calories during the virtual lunch compared to the placebo condition ($F_{1,29.6} = 4.49$, $p = .043$.

Conclusions: We found that the novel ghrelin receptor inverse agonist PF-5190457 had a significant effect on food choice behavior during a virtual lunch buffet and an effect on attention to alcohol cue during a cue reactivity paradigm in a bar-like laboratory. However, there was no effect of the drug on alcohol or food cue elicited craving in the bar-like laboratory. These preliminary findings suggest that ghrelin may play a role in the attention to reward related cues and stimuli but not craving in detoxified individuals with AUD. Future investigations should explore different stages of the AUD cycle to assess if ghrelin antagonism has similar effects across all stages.

Keywords: Alcohol Use Disorder - Treatment, Clinical Trial, Ghrelin

Disclosure: Nothing to disclose.

P432. Molecular Characterization of the Mouse Lateral Septum

Lionel Rodriguez*, Matthew Nguyen Tran, Elizabeth Pattie, Heena Divecha, Leonardo Collado-Torres, Stephanie Cereceo Page, Keri Martinowich

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

Background: The lateral septum (LS) is a midline, basal forebrain region that is surrounded by the lateral ventricles. The LS is a sensory and affective integrator that utilizes environmental stimuli to modulate behavior. In the rodent, LS circuitry plays roles in reward and fear learning, stress responses, context-dependent feeding, and various social behaviors, including aggression and response to social novelty. In line with these functions, the LS is connected, often reciprocally, to regions such as the ventral tegmental area, the dorsal raphe, the amygdala, various hippocampal sub-regions, and many cortical regions. While it is known that the LS contains diverse populations of GABAergic cells, which express combinations of neurotensin, somatostatin, and calbindin but are devoid of parvalbumin, the overall chemoarchitecture of the LS is understudied. To better understand and characterize cellular composition in the LS, we used single-nucleus RNA-sequencing (snRNA-seq) to generate a molecular atlas of LS cell types.

Methods: Here, we used the 10X Genomics 3' single cell platform to generate molecularly defined LS cell-types using snRNA-seq. For each sample, LS tissue from 2 naive mice of the same sex were pooled and processed together [N = 4 total samples (n = 8 mice,4 female, 4 male)]. Brains were extracted, flash-frozen, and sectioned into two 1 millimeter coronal brain slabs encompassing the span of the LS across the anterior- posterior axis. The LS was dissected from each slab, and nuclei were isolated using the "Frankenstein" nuclei isolation protocol developed by Martelotto et al. (2020). Samples were sorted on Bio-Rad S3e Cell Sorter, with 9000 nuclei sorted directly into reverse transcription reagents from the 10x Genomics Single Cell 3' Reagents v3.1 kit. 10x Chromium was performed and libraries prepared according to the manufacturer;s protocol, and then sequenced on an Illumina NovaSeq 6000. FASTQ data generated from sample libraries were aligned to the mouse genome using cellranger (version 6.1) utilizing the –include-introns flag to account for the nuclear transcriptome settings. Using the Bioconductor suite of single-cell analysis packages by Amezquita, et al. (2021), nuclei calling was performed using emptyDrops on the raw feature-barcode matrices. Specifically, we used a sample-driven threshold to call nuclei as the default threshold failed and called ~10-fold greater nuclei than were sorted. Next, we performed mitochondrial rate adaptive thresholding using a 3x median absolute deviation from the median to threshold and filtered called nuclei from each sample. Feature selection was computed using deviance residuals and the total deviance was used to calculate the top 2000 highly deviant genes. In this feature space, PCA was conducted as by Townes et al (2019). Using the top 50 PCs, we performed graph-based clustering, using k = 20 nearest neighbors and the walktrap community detection algorithm yielded 35 preliminary clusters. We annotated clusters based on expression of

the mouse orthologs of broad cell class markers from Tran, Maynard et al (2021). For cluster marker detection we employed the approach outlined in Tran, Maynard et al (2021) to characterize strict pairwise test-significant markers per cluster, in addition to cluster-enriched markers. The identified gene markers that were uniquely expressed by each cluster were surveyed using Allen Brain's Genome-wide atlas of in-situ hybridization (ISH) data to assign clusters to regions by exploring marker spatial distributions.

Results: We identified 33 cell-type clusters, 24 of which were neuronal. Within the neuronal populations 18 were from the septum and six were from other, adjacent regions (the striatum (four), thalamus (one), and islands of Calleja (one)). The septal populations were subclustered and further classified. Nine GABAergic clusters were annotated as LS including a distinct GABAergic population that lined the ventricle. Three glutamatergic clusters were annotated as medial septum including a small cholinergic population. Three glutamatergic clusters were annotated as a mix of neurons from the dorsocaudal septum where the tenia tecta, induseum griseum and septohippocampal nucleus meet. Two GABAergic clusters were annotated as septal broadly as their cluster markers were not confined to a previously identified septal sub-division. One glutamatergic cluster was annotated as the triangular nucleus of the septum. A curated set of genes, identified from previous studies of the chemoarchitecture of the LS, were used to characterize the LS-specific clusters. These genes included receptors for neuromodulators including dopamine, norepinephrine and serotonin as well as receptors for neuropeptides such as oxytocin, vasopressin and corticotropin releasing hormone. We then identified the cellular expression patterns of these markers in our annotated clusters. Finally, we generated an interactive web browser to allow exploration of these data for the neuroscience community.

Conclusions: Together, these data create a molecular atlas of septal cell types. We describe cell-specific gene expression patterns, and identified novel LS neuronal cell-types. This resource provides cell-type-specific information that will be useful in better understanding the biology of the LS, and which cell types may underlie behaviors that are mediated by the LS.

Keywords: Lateral Septum, Single-Nucleus RNA Sequencing, GABA Neuron, Septum

Disclosure: Nothing to disclose.

P433. Gender Differences in Adult ADHD Symptoms: A Network Analysis of Real-World Data

Kira Griffiths, Gunjan Batra, Li Tong Low, Emily Palmer, Miguel Renteria, Rashmi Patel, Scott Kollins*

Holmusk, New York, New York, United States

Background: Symptoms of ADHD can be classified as inattentive (IA) or hyperactive/impulsive (H/I). However, significant heterogeneity in the clinical presentation of ADHD has been reported across development and between genders. Network analysis offers a more detailed understanding of symptom structure compared to traditional factor analytic approaches. Electronic health record (EHR) data captures real-world symptom presentation in large and representative cohorts. The current study applied network analysis to real-world symptom data from adults with ADHD. This study aimed to explore and compare ADHD symptom networks in adult males and females.

Methods: Symptoms recorded by mental healthcare professionals as part of clinical assessment were obtained from a de-identified EHR dataset from 25 US mental healthcare systems (WCG Institutional Review Board Ref: WCG-IRB 1-1470336-1).The cohort included adult patients (>18 years) with a diagnosis of ADHD (ICD9/10: 314.00, 314.01, F90.0, F90.1, F90.2, F90.8 and F90.9) and symptom data within the first six months of diagnosis. Symptoms were defined according to the 18 symptoms outlined in the DSM-5. Networks for males and females were constructed separately, with each symptom reflecting a unique node within a network. Symptoms were entered as binary variables indicated by their presence or absence within individual patients. Analyses were conducted using R version 4.1.3, Bootstrap package 1.5 and Isingfit package v0.3.1. Networks were estimated using the eLASSO procedure and then visualized. In addition to the network graphs the centrality measures of betweenness, closeness and strength were calculated. In networks of psychiatric symptoms, centrality measures allow the identification of symptoms of high importance within a network and may therefore highlight key symptoms to target for intervention.

Results: Data were available for 2,398 patients (53% female) with a diagnosis of ADHD. Networks clustered into H/I and IA symptoms. In adult males, hyperactive/impulsive symptoms form a relatively tight network, whereas inattentive symptoms are less central. H/I and IA symptom clusters in females were relatively more dispersed than in males. In terms of strength, the top three unique symptoms for males and females both included two symptoms from the H/I cluster and one from the IA cluster. However, these differed by gender with the strongest symptoms associations in males being excessive/inappropriate movement, being "on the go" and failure of close attention. Whereas for females' symptoms of difficulty remaining seated, frequently interrupting and being easily distracted had the highest strength in the network. In terms of closeness, the most central symptom was in the IA cluster for females (does not seem to listen) and the H/I cluster for males (difficulty waiting their turn). H/I symptoms were the most central in terms of betweenness for males and females, both of which reflected inappropriate movement.

Conclusions: Clustering of H/I and IA symptoms is generally consistent with the current conceptualisation of ADHD. Differences in unique symptoms identified as most central in each network add further evidence that symptoms may vary in terms of clinical relevance for males and females, which may impact the identification, diagnosis, and treatment of ADHD between genders. These differing networks may also be important for identifying unique treatment targets for males and females. In addition, the general approach to characterizing networks could also be important for more precisely defining ADHD symptoms in other subgroups (eg., different racial/ethnic groups, different developmental levels).

Keywords: ADHD, Gender Differences, Network-Analysis

Disclosure: Holmusk: Employee (Self), Holmusk: Stock / Equity (Self)

P434. An Experimental Program to Determine the Effects of Viloxazine on Cortical Serotonin Neurotransmission at Doses Relevant for ADHD Treatment

Jami Earnest*, Jennie Garcia-Olivares, Brittney Yegla, Vladimir Maletic, Chungping Yu

Supernus Pharmaceuticals Inc, Crownsville, Maryland, United States

Background: FDA-approved ADHD treatments are presumed to work by augmenting dopamine (DA) and norepinephrine (NE) signaling in the brain. The focus on these neurotransmitter systems stems largely from the pharmacology of effective treatments. However, the breadth of symptoms and comorbidities in ADHD, emerging genetic and imaging studies, and the demonstrated robust efficacy of viloxazine ER (despite its moderate ability to inhibit norepinephrine transporters (NET)) all suggest a more complex neuropathology may be involved. In a prior microdialysis study, we found viloxazine augments serotonin

(5-HT) in the prefrontal cortex (PFC) in addition to increasing NE and DA. Similarly, an early in vitro functional activity study suggested viloxazine can act as a partial agonist at 5-HT2C receptors and antagonist at 5-HT7 and 5-HT2B receptors. However, it was unclear to what extent these effects occurred at clinically relevant concentrations. To further understand these serotonergic effects, we initiated a series of experiments to build upon this earlier work. Our objectives are three-fold: 1) Can we confirm and better elucidate the previously observed serotonergic effects of viloxazine and determine whether they occur at clinically relevant concentrations? 2) Are these effects observed in species with close physiology to humans?

Methods: To answer objective 1, viloxazine was assessed in in vitro binding competition assays for 5-HT2C, 5-HT2A., and 5-HT7 receptors using the specific radioligands [3H]-mesulergine, [3H]-ketanserin and [3H]-LSD, respectively, and corresponding reference compounds. An in vivo PK/PD microdialysis study was also conducted in Sprague-Dawley rats by administering 1, 3, 10, or 30 mg/kg doses of viloxazine, to determine the relationship between viloxazine concentrations in the interstitial fluid (ISF) and changes in concentrations of NE, DA, 5-HT, and their metabolites in the PFC. To answer objective 2, a PET imaging study using a 5-HT2A/2 C radioligand agonist, [11 C]CIMBI-36, was conducted in anesthetized cynomolgus monkeys to evaluate viloxazine's effect on endogenous cortical 5-HT release and direct binding to 5-HT2C receptors in the choroid plexus.

Animal research was performed at Charles River Laboratories (South San Francisco, CA, USA), the Karolinska Institute Centre for Psychiatry Research (Stockholm, Sweden), and approved by local ethics and animal care committees. Animals were cared for according to international standards.

Results: Objective 1: In vitro binding competition assays in cell lines expressing the human isoforms of 5-HT2C, 5-HT7 and 5-HT2A confirmed viloxazine's affinity towards these receptors with Ki values of 1.5, 1.9 and 16.3 μM, respectively. In addition, the microdialysis study in rats was conducted using a 30 mg/kg dose, at which the Cmax of viloxazine in ISF was 3.5 ± 1.6 μM, approximating the unbound viloxazine plasma concentrations (2.1-3.3 μM) observed in pediatric ADHD patients receiving 400 mg/day of viloxazine ER. At this clinically relevant concentration, viloxazine increased NE levels up to 558% over baseline and significantly decreased its DHPG metabolite from baseline, confirming NET inhibition. Serotonin levels were significantly increased up to 213% over baseline; however, no significant changes were observed in its 5-HIAA metabolite, demonstrating that serotonergic effects were not due to 5-HT reuptake inhibition. Objective 2: Administration 3 mg/kg of viloxazine to non-human primates produced an unbound plasma concentration of 3.9 μM (Cmax), aligning with concentrations measured in pediatric ADHD patients. At this clinically relevant viloxazine concentration, changes in binding potential of [11 C]CIMBI-36 in the choroid plexus and cortical regions were 60% and 25-36% respectively, indicating viloxazine may directly bind to 5-HT2C receptors in choroid plexus, and elevate endogenous 5-HT levels in cortical regions.

Conclusions: To date, our experiments to further elucidate the potential serotonergic effects of viloxazine have successfully shown that 1) the previously observed in vitro effects of viloxazine on serotonin receptors and the in vivo augmentation of serotonin in rat PFC are present at clinically relevant concentrations and 2) In vivo, serotonin modulatory effects are also seen in non-human primates, suggesting that they may translate clinically. Overall, it appears viloxazine can increase serotonin concentrations in cortical regions, including the PFC, at concentrations that have been shown to be therapeutic in pediatric ADHD trials. Data from in vitro receptor studies and the PET study are consistent with viloxazine binding to the 5HT2C receptor.

Keywords: Viloxazine, ADHD, PET Imaging, Microdialysis, Serotonin

Disclosure: Supernus Pharmaceuticals: Employee (Self), Johnson and Johnson, Alkermes, Merck: Stock / Equity (Self)

P435. Peripheral Tumor Presence Produces Impairments in Hippocampal-Dependent Cognition and Persistent Elevated Cytokines in Plasma When Treated With Docetaxel

Amber Asher*, David Morilak

The University of Texas Health Science Center, San Antonio, Texas, United States

Background: Prostate cancer (PC) is the second most common cancer in American men such that 1 out of every 8 men will be diagnosed with PC at some point in his life. Despite this high prevalence, treatment of PC is effective, and the 5-year survival rate is over 99%. Unfortunately, however, many survivors of cancer experience persistent cognitive impairments associated with their life-saving treatment, and there are few treatment options available to these patients to prevent or reverse impairments. In particular, patients who receive chemotherapy exhibit impairments in working memory, cognitive flexibility, attention, and visuospatial memory that can last after chemotherapy ends in a phenomenon colloquially referred to as 'chemobrain'. Docetaxel (DTX) is a microtubule-stabilizing agent approved for the treatment of late-stage PC and is associated with cognitive impairments in both human cancer patients and in studies of healthy rodents. Mechanisms behind chemobrain remain to be elucidated. Moreover, in preclinical investigations, the additional factor of cancer pathophysiology is rarely considered, and the contribution of a peripheral tumor to these impairments is also unknown. Here, we use a novel syngeneic rat PC model to characterize cognitive impairments produced by the interaction of a tumor and DTX. Moreover, we investigate changes in two circulating proinflammatory cytokines, tumor necrosis factor-alpha (TNFα) and interleukin-6 (IL-6) as potential physiological changes produced by a tumor that could render the brain susceptible to DTX.

Methods: Male Copenhagen rats, aged 3 months, were used in this study. To generate the PC model, a small fragment of a tumor grown from Dunning R-3327G cells, syngeneic with Copenhagen rats, was implanted subcutaneously along the dorsal midline between the scapula and pelvis. Sham animals received a sham surgery. When the tumor became palpable, animals were treated with DTX (4.5 mg/kg/injection; i.p.) or its vehicle (i.p.) three times over five days followed by a two-week recovery period. Hippocampal-dependent visuospatial memory and working memory were assessed using the novel object location task and spontaneous alternation on a y-maze, respectively. Plasma was collected after behavioral testing, and concentrations of TNFα and IL-6 were quantified using commercially available enzyme-linked immunosorbent assays. Tumors were measured using digital calipers every 7 days and tumor volume was calculated.

Results: Tumor-bearing rats exhibited impairments in visuospatial memory regardless of treatment. Increases of both TNFα and IL-6 were observed in the plasma of tumor-bearing rats compared to sham rats, regardless of treatment. DTX treatment alone in sham-implanted rats also produced an elevation in plasma IL-6 still evident two weeks after cessation of treatment.

Conclusions: The presence of a peripheral tumor produced impairments in hippocampal-mediated visuospatial memory and increases in TNFα and IL-6 in plasma compared to sham animals. DTX administration in sham-implanted rats produced modest impairments in hippocampal-dependent behavior that appeared to be worsened in animals with tumors. In peripheral measures of inflammatory cytokines, DTX treatment in sham animals produced an elevation in IL-6 in plasma still evident 2 weeks after the last injection, indicating lasting inflammation in sham animals. TNFα and IL-6 were elevated in both vehicle-treated tumor-bearing rats

and those treated with DTX, despite the tumor volume being significantly reduced in the latter group. Taken together, the presence of a peripheral tumor produces impairments in hippocampal-dependent cognition and elevates TNFα and IL-6 in plasma even when treated effectively with DTX. Experiments are ongoing to examine more acute changes in cytokine production, as well as changes in blood-brain integrity produced by the combination of tumor and DTX.

Keywords: Cancer, Cognitive Impairment, Hippocampus, Inflammation

Disclosure: Nothing to disclose.

P436. Astrocyte-Neuronal Metabolic Coupling in the Cingulate Cortex Promotes Chronic Pain Development

Giannina Descalzi*, Kaitlin Scherer, Paige Reid, James Tang

University of Guelph, Guelph, Canada

Background: Chronic pain is a major risk factor for anxiety and depression, with over sixty percent of people with chronic pain experiencing severe depression and anxiety; significant reductions in physical and social functioning; and underemployment or job loss due to their pain. Tragically, the risk of death by suicide is doubled in people with chronic pain. Present treatment options are woefully inadequate, with less than half of all patients reporting pain relief with current treatments. Human and rodent neuroimaging studies indicate that chronic pain corresponds with reorganization of an emotion-pain brain circuit, and evidence indicates that neuroplasticity of the anterior cingulate cortex (ACC) is a critical step in this reorganization. We previously showed chronic pain and fear learning enhance neuronal excitability and induce similar plasticity-related gene expression changes in the anterior cingulate cortex of mice. We recently showed that fear learning requires astrocyte-neuronal lactate shuttling (ANLS) in the dorsal hippocampus, and that ANLS is necessary for learning-induced associated molecular changes, including increases in plasticity-related gene expression. Here we present data that indicate that ANLS in the ACC may also be involved in neuroplasticity associated with murine models of chronic pain.

Methods: Male adult mice were exposed to chronic inflammatory pain, modelled by injection of complete Freund's adjuvant (CFA) into the hindpaw. Vehicle injections were used as control. Pain thresholds were measured at 3 hours (h) 24 h, 72 h, and 7 days post-injury, and ACC samples were extracted immediately after threshold testing. Lactate levels were quantified through a colorimetric lactate assay, and western blot analyses determined the expression of proteins involved in astrocyte-neuronal lactate shuttling. Temporary, continuous inflammatory pain was modelled through formalin injections into the hindpaw, with vehicle injections serving as control. We used antisense oligonucleotides (AS-ODN) targeting monocarboxylate MCT4, which is exclusively expressed on astrocytes, to decrease the export of lactate out of astrocytes.

Results: Male ($n = 10$/group) mice showed a rapid increase in lactate levels in the ACC, detectable at three hours post CFA injection compared to vehicle injected controls. These levels returned to baseline by one day post injury, but steadily increased thereafter, resulting in significantly larger levels of lactate seven days post injury, compared to non-pain, vehicle-injected control mice. Mice injected with MCT4 AS-ODN in the ACC showed significant reductions in MCT4 protein expression in the ACC 24 hours (hrs) after injection, compared to injections of a non-coding, control scrambled ODN. We found that male mice that had intra-ACC MCT4 AS-ODN 24 h prior to formalin injection showed significantly less pain-related licking behavior in the formalin test, compared to non-coding, scramble control, which was rescued by intra-ACC injection of lactate 15 min prior to formalin injection ($n = 9$/group).

Conclusions: Our data indicates that astrocyte-neuronal coupling is critically involved during the early stages of chronic pain development. Furthermore, our data indicate that disrupting astrocyte-neuronal lactate shuttling in the ACC blocks the persistence of pain in male mice.

Keywords: Astrocyte, Chronic Pain, Astrocyte-Neuronal Lactate Shuttle

Disclosure: Nothing to disclose.

P437. α5-GABAA Receptor Positive Allosteric Modulation Reverses Cognitive Deficits and Neuronal Atrophy Across Animal Models

Thomas Prevot*, Ashley Bernardo, Michael Marcotte, Kayla Wong, Prithu Mondal, James Cook, Etienne Sibille

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

Background: Multiple psychiatric, stress-related, and neurodegenerative disorders exhibit reduced GABA/somatostatin (SST) signaling. SST + interneurons from cortical layers and the hippocampus exert dendritic inhibition onto excitatory neurons, largely through α5-containing GABAA receptors (α5-GABAAR). Our group showed that α5-positive allosteric modulation contributes to alleviating working memory deficits and reverses neuronal atrophy in old mice, and in stressed mice. Here, we aimed to confirm our prior results in an independent aged cohort and further investigated the behavioral and neurotrophic effects of an α5-positive allosteric modulator (α5-PAM) in animal models of other conditions associated with reduced GABAergic function such as chronic stress and β-amyloid load.

Methods: Three studies are presented, with ~$N = 12$ mice/group, 50% female: 1) Young C57BL6 subjected to unpredictable chronic mild stress (UCMS) to induce cognitive deficits. 2) 20-month-old C57BL6 with an existing cognitive decline. 3) 5xFAD transgenic mice with progressive amyloid-related cognitive decline. In all studies, the efficacy of chronic administration of the α5-PAM (30 mg/kg, p.o, for 4 weeks) at rescuing cognitive deficits across 3 domains was assessed. Working memory was assessed in an alternation task in the Y-maze, spatial learning and memory in the water maze, and cognitive flexibility in a set-shifting assay. Brains were then stained using the Golgi-Cox technique ($n = 4$brain/group; 8cell/brain), sectioned, and mounted for quantification of dendritic length and spine density in the prefrontal cortex and hippocampus (NeuroLucida). All techniques and studies employing laboratory animals were in accordance with Ontario Animals for Research Act (RSO 1990, Chapter A.22), Canadian Council on Animal Care (CCAC), and approved by CAMH Animal Care Committee.

Results: Chronic treatment in stressed, old, or 5xFAD mice reversed cognitive deficits across domains in each model (ps<0.01) with a strong effect on working memory. Chronic treatment also consistently reversed UCMS-, age- or amyloid-induced dendritic shrinkage and spine loss at apical and basal dendrites ($p < 0.001$ in PFC and CA1).

Conclusions: Together, the presented results support that selective targeting of α5-containing GABAA receptors overcomes chronic stress-, aging- or amyloid-related cognitive deficits upon chronic treatment, and reverses detriments in neuronal morphology. Altogether, the data suggest a positive impact on both symptomatic and disease-modifying dimensions, highlighting the high therapeutic potential of α5-PAMs.

Keywords: Cognition, Dendritic Spine, Animal Models, α5 GABAA Positive Allosteric Modulator

Disclosure: Nothing to disclose.

P438. Valbenazine Effects on the Dopamine System in Humans, as Measured by [11 C]-PHNO Positron Emission Tomography (PET)

Ryan Terry-Lorenzo*, Daniel Albrecht, Satjit Brar, Graham Searle, Frans Van Den Berg, Ilan Rabiner, Dietrich Haubenberger

Neurocrine Biosciences, Inc., San Diego, California, United States

Background: Valbenazine, a potent, selective, orally active vesicular monoamine transporter 2 (VMAT2) inhibitor, is approved by the US Food and Drug Administration for the treatment of tardive dyskinesia (TD) at doses of 40 to 80 mg once daily. By inhibiting VMAT2, valbenazine disrupts the packaging of monoamines into synaptic vesicles, subsequently decreasing the release of monoamines including dopamine into the synaptic cleft. The reduction of dopamine is thought to be the foundational basis for the efficacy of valbenazine in treating TD and other hyperkinetic movement disorders. Reduction in synaptic dopamine is also a rationale for the potential utility of valbenazine in the treatment of psychosis. To our knowledge, however, there has not been a demonstration of dopamine reduction by valbenazine, or any other VMAT2 inhibitor, in humans. The aim of this study was to investigate the change in synaptic dopamine following valbenazine administration, using positron emission tomography (PET) imaging in healthy human volunteers.

Methods: Imaging and tolerability data in this adaptive study were collected and analyzed in cohorts of 2-4 healthy volunteers. For each scan, participants received an injection of the D2/D3 dopamine receptor agonist radioligand [11 C]-PHNO, followed by 90 min of data acquisition using Siemens Biograph PET/CT. For post-valbenazine scans, participants received an oral dose of valbenazine 6-8 hours before the administration of [11 C]-PHNO, with PET imaging occurring around the time of maximal valbenazine plasma concentration. The binding potential relative to the non-displaceable binding (BPND) in the putamen, caudate, and ventral striatum, was used as the primary endpoint. The cerebellum was used as the reference region to estimate the regional BPND. Decreases in synaptic dopamine following valbenazine administration corresponded to an increase in [11 C]-PHNO BPND relative to baseline (ΔBPND). Plasma concentrations of valbenazine and (+)-α-dTBZ (dihydrotetrabenazine), the active metabolite of valbenazine, were measured at the start and end of each post-valbenazine PET scan. The mean plasma (+)-α-dTBZ concentration (Cave) was matched to the ΔBPND for each participant to provide an exposure- response curve.

Results: To date, 9 participants (5 male, 4 female) have completed the trial. These participants received between 40-160 mg valbenazine, which resulted in plasma (+)-α-dTBZ Cave between approximately 10-60 ng/mL. Eight participants displayed valbenazine-induced, dose-dependent increases in [11 C]-PHNO ΔBPND (21-44%). Higher exposures to (+)-α-dTBZ from higher doses of valbenazine resulted in greater ΔBPND, revealing a monotonic exposure-response curve. Adverse events in this study were consistent with the known safety and tolerability profile of valbenazine, as reported in TD clinical trials.

Conclusions: Valbenazine appears to decrease synaptic dopamine in a dose- and concentration-dependent manner, as indicated by an increase in [11 C]-PHNO ΔBPND. The approximately 20-40% [11 C] PHNO ΔBPND increase observed in this study is similar to the [11 C]-PHNO ΔBPND seen previously following treatment with a tyrosine hydroxylase inhibitor to deplete dopamine (Caravaggio et al, Neuropsychopharmacology 2014;39:2769). Thus, at pharmacological and therapeutic valbenazine doses, biologically meaningful dopamine decreases were observed in humans. These data will enable future exploration of the relationship between VMAT2 inhibition and the potential treatment of other central nervous system disorders.

Keywords: Positron Emission Tomography (PET), VMAT2, Biomarker

Disclosure: Neurocrine Biosciences, Inc.: Employee (Self)

P439. Dose Dependent Effects of Acute Methamphetamine on EEG Alpha Power, Self-Reported Stimulation and Blood Pressure in Healthy Adults

Connor Haggarty*, Conor Murray, Royce Lee, Ilaria Tare, Harriet de Wit

The University of Chicago, Chicago, Illinois, United States

Background: Methamphetamine (MA) is a stimulant drug characterized by increased feelings of stimulation, cardiovascular effects, and changes in electrophysiological function. In particular, previous studies indicate that stimulants decrease alpha power in EEG studies, consistent with increased wakefulness. In the present study, the subjective, physiological, and cortical effects of acute MA (10 and 20 mg) were measured in healthy volunteers to examine relationships between its effects on alpha power and other measures.

Methods: Healthy volunteers (N = 29), aged 18-35, participated in a within-subject, double-blind procedure in which they received a placebo, 10 mg, and 20 mg MA in 4-hour sessions at least four days apart. During the sessions, they completed subjective effects questionnaires and heart rate and blood pressure measurements were obtained. One hour after ingesting the capsule, resting state EEG measures were obtained to determine power at five frequency bands, with eyes open. Data were analyzed from electrodes representing the default mode network.

Results: MA (10 and 20 mg) dose-dependently increased ratings of drug liking and vigor, and increased mean arterial pressure, compared to placebo. MA (10 and 20 mg) significantly decreased alpha power in a linear fashion, without significantly affecting other frequencies. The decrease in alpha power after MA (20 mg) significantly correlated with increases in feelings of vigor, but it was not correlated with increases in blood pressure.

Conclusions: These data support the association between increased feelings of arousal and decreased alpha frequency power. Single doses of MA increased both subjective stimulation and blood pressure and decreased alpha power, but only the subjective effects were related to the EEG measure. This supports the EEG measure as an index of cortical processing relating to acute drug effects.

Keywords: Methamphetamine, EEG, Subjective Effects

Disclosure: Nothing to disclose.

P440. Clinical Psychopharmacology: Racial and Ethnic Issues

Andrew Olagunju*, Jeffrey Wang

McMaster University, Hamilton, Canada

Background: There are advantages and positive contributions of ethnicity and race to health and wellbeing (e.g., through self/collective identity). However, belonging to a racial or ethnic group has also been linked with health-related disadvantages and negative perceptions —racism, stereotyping, and discrimination. A good knowledge of the ethnic/racial classification and groups in the particular setting of practice is beneficial for a culturally sensitive clinical practice and decisions in psychopharmacology. It

is with this background that we pursue this study to survey and describe current literature on ethnic and racial issues in the practice of clinical psychopharmacology.

Methods: This study is conducted following the Preferred Reporting Items for Systematic reviews and Meta-Analyses extension for Scoping Reviews guideline. We searched major databases, including Pubmed/Medline, PsycINFO, Embase, CINAHL, and Web of Science for eligible studies published till date. The search string combined MeSH terms for clinical psychopharmacology, race and ethnic. This was supplemented by snowball searching of references in relevant studies and authors were contacted to request their work where necessary. All included studies will be rated with the National Institutes of Health Study Quality Assessment Tools based on study designs. Data extraction and quality assessment is conducted on the included studies.

Results: A total of 57 reports are identified for title and abstract screening. This will be followed by data extraction and analysis.

Conclusions: Through this review, we hope to highlight relevant issues to promote racial diversity and equity in the practice and training of psychopharmacology. The integration of ethnic matching and adequate racial/ethnic representation in study participants enrolled into clinical trials on psychotropics and other research activities will be addressed.

Keywords: Racial/Ethnic Differences, Clinical Psychiatry, Psychopharmacology

Disclosure: Nothing to disclose.

P441. Old Rats With Fetal Alcohol Exposure Show Impaired Learning and Memory Functions and Elevated Levels of Various Biochemical Markers of Alzheimer's Disease in the Brain

Shaista Chaudhary, Dipak Sarkar*

Rutgers University, New Brunswick, New Jersey, United States

Background: The influence of alcohol consumption during pregnancy could alter phenotype in the offspring and can have devastating and persistent consequences, such as central nervous system dysfunction. However, it is not known whether fetal alcohol exposure promotes physiological and biochemical characteristics of Alzheimer's disease during aging.

Methods: We employed an established first and second trimester human equivalent rat model of fetal alcohol exposure comprising of feeding liquid diet containing ethanol at a concentration of 6.7% v/v, or pair-feeding an isocaloric liquid diet or ad libitum feeding rat chow from gestational days 7 through 21 in isogenic Fischer 344 (F344) rats. Pups were weaned on postnatal day 21 and housed by sex, and were used for behavioral and biochemical studies at about 12 months of age. In each experimental group, only one male and one female offspring from a rat litter was used. Six rat offspring were assigned in each group. All animal care and procedures were approved by the Rutgers Institutional Animal Care and Facilities Committee and complied with National Institutes of Health policy. We measured learning and memory behavior using Morris water maze test. Since cognitive and memory impairments are often related to cholinergic dysfunction and acetylcholinesterase (AChE) activity, we evaluated the activity of AChE in cerebral cortex and hippocampus regions of the brain. Using Western blot assays we measured the levels of some biochemical markers of Alzheimer's disease, including β-amyloid (Aβ) and Aβ1-42 proteins, total and phosphorylated tau proteins, β-site amyloid precursor protein (APP) cleaving enzyme 1 (BACE1) and netrins receptor UNC5C proteins in cerebral cortex and hippocampus regions of the brain. Data were statistically evaluated using two-way analysis of variance (ANOVA) followed by Bonferroni's multiple comparison test in order to determine the time-dependent changes between groups. Multiple group comparisons at single time point were made using one-way ANOVA followed by Tukey's post-hock test. A p value less than 0.05 was considered significant.

Results: Determination of cognitive and memory functions using Morris water maze test showed enhanced escape latency in both male and female fetal alcohol exposed (alcohol-fed offspring) rats than control (both ad lib-fed and pair-fed offspring) rats at age 12 months. In addition, fetal alcohol exposed rats showed reduced time spent escaping the platform's quadrant, indicating a significant deficiency in memory in these animals. The magnitude of fetal alcohol effects on learning and memory behaviors appears to be similar in both male and female offspring. These animals also had elevated levels of AChE activity, Aβ and Aβ1-42 proteins, hyper-phosphorylated tau, BACE1 and UNC5C proteins in the cerebral cortex and hippocampus regions of the brain during aging. The magnitude of enzyme and protein level responses to FAE was also similar in both male and female offspring.

Conclusions: Overall, these findings provide evidence that fetal alcohol exposure increases the expression of some of the behavioral and biochemical phenotypes of Alzheimer's disease during aging.

Keywords: Fetal Alcohol Spectrum Disorder, Neurodevelopmental Disorders, Alzheimer's

Disclosure: Nothing to disclose.

P442. Elucidating Brain Networks Subserving Working Memory Task Performance Using Interpretable Deep Learning

Mario Serrano-Sosa, Philip Tubiolo, Thomas Hagan, John Williams, Chuan Huang, Jared Van Snellenberg*

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

Background: Working memory (WM) is a key cognitive domain and is disrupted in many psychiatric and neurological disorders. While a bilateral network of brain regions including dorsolateral prefrontal cortex (DLPFC), pre-supplementary motor area (pre-SMA), and intraparietal sulcus (IPS) reliably activate during WM tasks, less is understood about how activation in these regions relates to behavioral measures of WM ability. One major challenge is the high dimensionality of fMRI data, which generally limits analyses to single-voxel mass-univariate linear model approaches, or similar.

However, recent work in computer vision has demonstrated that some neural networks (convolutional neural networks; CNNs) show good performance in classification and prediction from image data. CNNs learn hierarchical features of images across different levels of abstraction in a non-linear and data-driven manner. Critically, while deep learning algorithms are often "black boxes", network back-propagation can be used in a CNN to determine which image locations are most salient for network performance. These "saliency maps" can be generated from a trained network for each input, and reflect the gradient of the output prediction, indicating which image locations most strongly drive network performance.

Here we use data from an n-back WM task from the Human Connectome Project (HCP) and a CNN to identify cortical regions whose activation during WM are predictive of WM performance in a whole-brain, non-linear, data-driven manner. To our knowledge, this work reflects the first use of emerging developments in computer vision to identify brain regions whose activity is predictive of cognitive task performance using fMRI data.

Methods: We used n-back task data from 419 subjects (male and female) of the HCP 1200 Subjects release, using only subjects

with 3 T MR data, 100% WM task completion, no significant quality control issues (issues A, B, or C as per HCP documentation), and MSMAll-registered data. We used only one subject from each kinship set to prevent the CNN from learning features shared by genetically related individuals.

Cortical surface data of the 2-back - 0-back contrast (all stimulus conditions) was extracted from CIFTI images and placed on a flattened surface. Left and right hemispheres were concatenated to form a single 2d image for input to the CNN. The CNN used a U-net architecture with 4 convolutional blocks followed by 3 fully connected layers. The network was trained to predict % correct scores on the 2-back condition for each participant. We employed 5-fold cross-validation to obtain blind predictions and saliency maps for each subject: 5 independent networks were trained, each using 335 subjects for training and 84 subjects for validation. Saliency maps were generated via back-propagation using the SmoothGrad algorithm to reduce spatial noise. To benchmark CNN performance we also trained a kernel ridge regression (KRR) model.

Saliency maps (bounded between 0 and 1) were mean-centered for each subject to permit valid null-hypothesis tests. Maps were then used to identify regions with significantly above-average salience by testing the null hypothesis that salience $<= 0$ using Permutation of Linear Models (PALM) at $P < 0.025$, FWE corrected. Saliency maps were then z-scored and subjected to principal component analysis and correlated with task performance. The above analyses used dummy regressors for the 5 validation sets to remove systematic differences in salience across the 5 independent CNNs.

Results: The CNN outperformed KRR on all metrics evaluated (R2, MAE, MAPE, and RMSE), with R2 of 0.389 (0.373 for KRR). We observed signficantly above-average salience for bilateral ventrolateral PFC, left premotor cortex, and several default-mode network (DMN) regions including bilateral anterior medial PFC (amPFC), precuneus and retrosplenial cingulate cortex, temporal-parietal junction, temporal poles, and left middle-temporal gyrus. The majority of left and all of right DLPFC and bilateral intraparietal sulcus did not exhibit above-average salience.

Four principal components (1, 2, 3, and 9) were significantly associated with task performance, with PC1 showing the strongest relationship ($r = -0.54$). Loadings for PC1 revealed that strong task performance was associated with high salience in amPFC, medial temporal lobe, left middle temporal gyrus, and temporal pole, as well as low salience in bilateral DLPFC, IPS, and pre-SMA.

Conclusions: These results demonstrate that interpretable deep learning using CNNs and saliency maps holds promise as a novel means to gain insight into brain regions whose activation is associated with task performance. Contrary to expectations, our results suggest that the magnitude of deactivation in DMN contains more information about WM task performance than does activation of DLPFC, IPS, or pre-SMA. PCA results further suggest that activation in these latter regions is predictive of performance, but only in low-performing individuals. Because greater activation in these regions was associated with high performance, this suggests that only low levels of activation are predictive of poor performance—once activation in this network reaches some level, the CNN instead attends to deactivation in amPFC and other DMN regions to identify the strongest performers. While in its infancy, this approach has clear potential to produce new insights into how the human brain subserves higher-order cognition in both health and disease.

Keywords: Working Memory fMRI, Convolutional Neural Network (CNN), Human Connectome Project (HCP), Multivariate Approaches, Deep Learning

Disclosure: Nothing to disclose.

P443. A Novel Dissociation Between Two Parallel Thalamo-Striatal Pathways Encoding Distinct Motivational States

Sofia Beas*, Isbah Khan, Claire Gao, Emma McDonald, Alison Bashford, Shakira Rodriguez-Gonzalez, Mario Penzo

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

Background: Previous research has mainly centered on investigating the contribution of the dopaminergic mesolimbic system to these processes. Yet, the role that glutamatergic inputs to the NAc play in the control of motivation is far less clear. The paraventricular nucleus of the thalamus (PVT), a brain region that integrates bottom-up interoceptive signals with top-down cortical information, sends robust glutamatergic projections to the NAc. Recently, we identified two major distinct subpopulations of neurons in the PVT (Type1PVT and Type2PVT) that differ in their genetic identity, connectional features, and functionality. These subpopulations send divergent projections to the NAc, which raises the possibility that Type1PVT and Type2PVT neurons are likely to enable different but complementary aspects of motivated behavior. However, very little is known about the involvement of thalamic inputs to the NAc in the mediation of motivational processes.

Methods: Here, we trained female and male food-restricted mice ($n = 6$ - 8) to perform a linear runway foraging task. In this task, mice are trained to run from a trigger zone to the reward zone in the maze to retrieve a food reward (strawberry Ensure). Using bulk calcium imaging and fiber photometry, we investigated the in vivo dynamics of the Type1PVT (using Drd2-Cre mice and Cre- dependent GCaMP6s) and Type2PVT (using Drd2-Cre mice and a CreOFFGCaMP6f) parallel thalamo-striatal pathways while mice performed the linear runway foraging task.

Results: Our findings showed that the reward approach was associated with a prominent increase in GCaMP fluorescence in the Type1PVT–NAc pathway. This increase in fluorescence was quantified using the area under the curve, which revealed a significant increase in activity of this pathway when compared to baseline activity ($t(18) = 4.6$, $p < 0.01$) and when compared to GFP injected controls ($t(18) = 4.6$, $p < 0.01$). Moreover, we found that the activity of the Type1PVT–NAc pathway varies with aspects of motivation such as behavioral vigor and level of satiety. In contrast, Type2PVT–NAc neurons showed opposing in vivo dynamics and were not sensitive to motivational variables, suggesting that this pathway may participate in regulating other aspects of goal-oriented behavior.

Conclusions: Altogether, the results gathered from this study show a novel dissociation between the Type1PVT–NAc and Type2PVT–NAc pathways and identify a specific neuronal subpopulation of the PVT that signals motivational states.

Keywords: Motivation, Paraventricular Nucleus of the Thalamus, Nucleus Accumbens

Disclosure: Nothing to disclose.

P444. Neuropsychopharmacology (NPP) Special Projects Update: Efforts to Enhance Rigor in Clinical Trials Research and Promote Early Career Investigators

Sofiya Hupalo*, Chloe Jordan, Terri Bowen, Keri Martinowich, William Carlezon, Tony George

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

Background: NPP engages in special projects which provide insights into journal function and support outreach efforts to promote researchers in the fields represented within NPP and ACNP. In this report, we describe recent activities the NPP Special Projects Team has spearheaded that aim toward improving rigor and transparency in clinical trials research and creating opportunities for early career investigators to publish in NPP.

Methods: To examine the rigor and transparency of clinical research published in NPP, we examined the frequency with which authors provide CONSORT documents (Consolidated Standards of Reporting Trials) during the manuscript submission process. CONSORT documents report experimental details such as how the clinical trial was designed, analyzed, interpreted, and describe the flow of participants through the trial. We tracked NPP manuscript submissions to record the number of clinical trial reports being submitted and the proportion of submissions that provide complete CONSORT materials.

To promote early career scientists in the field of neuropsychopharmacology, in October 2021 NPP launched the Early Career Commentary for trainees and early-stage investigators to share their scientific ideas and perspectives. Pre-submission inquiries are required and are reviewed by the NPP Special Projects Team and Editorial Board. Priority is given to inquiries that focus on novel and timely topics (e.g., scientific, social, policy, equity) of broad interest and importance to the NPP and ACNP community. Selected inquiries are invited to proceed with submission and undergo standard NPP peer review. The Special Projects Team has invited authors of each Early Career Commentary to participate in NPP's "Meet the Author" Interview series. These interviews provide the authors an opportunity to promote their commentary and discuss their research and career goals. The recorded interviews are posted on NPP's YouTube channel and shared on Twitter.

Results: Clinical Trials Submissions:

In 2020, NPP received 140 manuscript submissions that were identified as a clinical trial by the submitting author. Only 70 (50%) of these submissions included complete CONSORT documents. When journal staff followed up with corresponding authors of the 70 submissions that did not include complete CONSORT documents, 14 authors requested an exemption and 5 withdrew their submission. Ongoing tracking of clinical trial manuscripts submitted in 2022 reveals a similar rate of non-compliance (~55%) in providing complete CONSORT documentation on the first submission.

Early Career Commentary: Since the launch, NPP received 36 pre-submission inquiries for the Early Career Commentary. Of these submissions, 7 were ineligible due to one or more of the authors not having early-stage investigator status. NPP additionally considered all pre-submission inquiries as topics for invited reviews unless the inquiry was withdrawn by the authors. To date, NPP has invited 4 groups to author an Early Career Commentary and an additional 3 groups to write an invited Perspective or Review article based on the proposed topic. Two Early Career Commentaries and one Perspectives article have been published. Collectively, these manuscripts have been accessed 7,162 times with Altmetric online attention scores ranging from the 85th-97th percentile.

Conclusions: To improve adherence to transparency and accountability in clinical trial reports published in NPP, the journal is implementing changes to the manuscript submission process. Text describing the definition of a clinical trial is now included directly on the submission webpage with links to information on these definitions. Submitting authors are now required to confirm whether their manuscripts meet the definition of a clinical trial and verify that the submission includes CONSORT documents and the clinical trial registry number. These changes will improve the speed and thoroughness of peer review, improve transparency in reporting clinical trial methodology, and increase the impact of work published in NPP.

The Early Career Commentary was created to provide a venue for early career investigators to share ideas and perspectives independently of senior mentors or collaborators. NPP has received many pre-submission inquiries and has selected additional topics for invited Reviews and Perspectives articles. The articles that have resulted from this initiative have been widely read and shared on social media, demonstrating that this is a successful approach for promoting and supporting early career scientists in the NPP and ACNP community.

Keywords: Clinical Trial, Clinical Trial Methodology, Career Development

Disclosure: Nothing to disclose.

P445. Pharmacokinetics of Cannabidiol: A Systematic Meta-Regression Analysis to Guide Clinical Trials

Ehsan Moazen-Zadeh*, Alexandra Chisholm, Keren Bachi, Yasmin Hurd

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

Background: Cannabidiol (CBD) has exponentially gained attention in research and clinical applications as a potential treatment of several neuropsychiatric and general medical conditions. Today, many CBD-based formulations are in development aiming for FDA approval and numerous non-approved CBD preparations are available over the counter. However, many questions raised by clinicians, researchers and consumers of CBD products often relates to dosing and administration. One of the major challenges that has prevented quantitative aggregation of evidence in previous reviews of pharmacokinetic (PK) studies of CBD, has been different units and scales of reporting outcomes. We aimed to provide an updated systematic assessment of available evidence on the pharmacokinetics of CBD; provide comparable values of PK parameters from different studies on the same scale; demonstrate patterns in outcomes based on the CBD dose and route of administration; and explore the simultaneous impact of different factors on PK outcomes using meta-regression models.

Methods: This systematic review and meta-regression analysis was pre-registered (PROSPERO: CRD42021269857). We systematically searched Medline, Embase, PsychInfo, and Web of Science Core Collection on September 19, 2021. Trials of CBD (pure CBD or in combination with THC) in healthy adults were included if they reported at least one of the PK parameters of interest in serum or plasma, including: time from medication administration to the maximum concentration of CBD in plasma/serum (Tmax), maximum concentration of CBD in plasma/serum (Cmax), the area under the curve of serum/plasma concentrations plotted against time from medication administration to a specific time-point, usually the end of the PK session (AUC0-t), the extrapolation of AUC0-t to the infinity time point (AUC0-inf), and the time needed for the concentration of the drug in the plasma to be reduced by 50% (T1/2). Studies of patient populations or CBD co-administration with other medications were excluded. The National Heart, Lung, and Blood Institute's Quality Assessment Tool for Before-After Studies with no Control Group was used. Random-effects multivariable meta-regression analysis was conducted (alpha=0.05) with hierarchical models of different factors as independent variables and PK parameters as the dependent outcomes.

Results: A total of 97 trial arms from 35 studies were included; 26 trial arms had a "Good" quality, 56 "Fair," and 15 "Poor." Six arms used inhalation CBD, 29 oromucosal, 61 oral, and 1 intravenous. CBD formulations could be categorized to nanotech (n = 13), oil-based (n = 19), alcohol-based (n = 10), water-based (n = 4), Sativex (n = 17), and Epidiolex (n = 21). For single-dose

studies, CBD doses ranged between 2-20 mg in inhalation, 5-50 mg in oromucosal, and 0.42-6000 mg in oral administration. Sixty trial arms had only male participants or a higher number of males than females. The duration of the PK session was between 4h-164h. In meta-regression models, a higher CBD dose was consistently associated with a higher Cmax, AUC0-t, and AUC0-inf across all the models (p-value<0.001). Compared to oral administration, oromucosal administration was associated with lower Cmax, AUC0-t, and AUC0-inf (p-value between 0.001 and 0.007). Fed status was associated with higher Cmax and AUC0-t when compared to the fasting status (p-value<0.001). A higher ratio of female participants was associated with lower Tmax in oral administration and higher Cmax and T1/2 in oral and oromucosal administration (p-value between 0.001 and 0.023). Longer study duration was associated with higher AUC0-inf and T1/2 in all models (p-value<0.001). The highest percentage of variability in the data that could be explained by a model (R2) was 84% for Tmax, 53% for Cmax, 55% for AUC0-t, 54% for AUC0-inf, and 92% for T1/2.

Conclusions: In exploring how different factors potentially influenced the PK outcomes of CBD, consuming food while taking CBD, female sex, and oromucosal administration were associated with higher bioavailability. Recommendations for future research would mainly concern conducting original systematic studies to elucidate the impact of biological sex, duration of PK session, and different formulations in single studies with multiple arms. It would also be beneficial for more studies to examine CBD doses in ranges currently being used clinically, given the increasing number of CBD preparations on the market and their potential application in clinical practice. Finally, reporting PK parameters in both arithmetic and geometric scales would help comparisons across studies, knowledge aggregation, and replicability.

Keywords: Cannabidiol, Population Pharmacokinetics, Serum Levels, Therapeutic Drug Monitoring

Disclosure: Nothing to disclose.

P446. Genetic Basis of Paclitaxel-Induced Peripheral Neuropathy Model Traits in a C57BL/6 Reduced Complexity Cross

Jacob Beierle, Bryan McKiver, Arthur Vanvalkenburg, Emily Yao, Jared Mann, Binh-Minh Nguyen, Kayla Richardson, William Johnson, Imad Damaj, Camron Bryant*

Boston University School of Medicine, Boston, Massachusetts, United States

Background: Chemotherapy-induced peripheral neuropathy (CIPN) is a heritable side effect in cancer therapeutics, yet the genetic basis is largely unknown. (PAC) exerts antitumor effects through stabilization of microtubules and interruption of cell division and is accompanied by CIPN in 60-70% of patients, resulting in numbness, cold hypersensitivity, and allodynia. Human genetic variants have been associated with CIPN within/near genes associated with cytoskeletal, neurodevelopmental, and pharmacokinetics. Discovery-based genetics in rodents offers a powerful, complementary approach to uncover new genetic and biological insight into CIPN, with the goal of improving chemotherapeutic profiles. Genetic crosses in closely related rodent substrains (reduced complexity crosses; RCCs) permit rapid identification of causal genes/variants underlying complex traits like CIPN, providing > 2-order magnitude reduction in the number of segregating variants within a locus and providing 2 nearly isogenic backgrounds to edit/validate necessity and sufficiency of causal variants. Our goal was to identify C57BL/6 substrain differences in CIPN model traits, conduct quantitative trait locus (QTL) mapping of behavior and gene expression and conduct transcriptome analysis of PAC Tx. We then identified candidate genes and downstream biological pathways associated with CIPN model traits.

Methods: We assessed CIPN model behaviors over 1 month in C57BL/6 J (J) versus C57BL/6NCrl (NCrl) mouse substrains following a regimen of paclitaxel (PAC: 2 mg/kg, every 48 h, 4X) vs. vehicle (VEH: 1:1:18 – ethanol, kolliphor, water), including mechanical hindpaw hypersensitivity (von Frey), cold hindpaw hypersensitivity (acetone test), dorsal caudal nerve conductance, sucrose preference, and tactile hyposensitivity (plantar hindpaw surface adhesive test). Following identification of substrain differences, we made a reciprocal F2 cross and phenotyped 182-206 F2 mice (½ VEH-Tx, ½ PAC-Tx). Genotyping was conducted using the miniMUGA DNA array, with > 200 genetic markers distinguishing J vs. NCrl. QTL mapping was conducted in R/qtl in 182-206 F2 mice using Hayley-Knott regression (1000 perm) to determine significance thresholds. Bayes credible intervals comprised confidence intervals. Treatment (Tx) was included as an interactive covariate and Sex and Cohort as additive covariates. RNA-seq of J and NCrl was conducted on dorsal root ganglia (DRG), spinal cord (SC), and periaqueductal gray (PAG) using poly-A selected RNA. We used Illumina Nova-Seq, with 100 bp paired-end reads and yielding > 30 million paired-end reads per sample.

Results: We identified robust inbred mouse substrain differences in CIPN model traits between NCrl and J, with J mice showing robust hypersensitivity to mechanical stimulation and cold hypersensitivity following PAC Tx. QTL mapping in an F2 RCC identified a major QTL on chr.1 mediating PAC-induced mechanical hypersensitivity on Day(D)7 ($LOD = 7.7$; $p < 0.001$; chr.1: peak 69 Mb:; Bayes: 47-76 Mb), and a trending QTL on chr.14 on D15 mediating cold hypersensitivity ($LOD = 4.4$; $p = 0.095$; chr.14: peak: 124 Mb; Bayes: 23-125 Mb).

For the chr.1 QTL for mechanical hypersensitivity, there are 2 compelling candidate causal genes with coding variants, including Abca12 (splice site, chr1: 71 Mb; a gene encoding a known efflux transporter for PAC) and Bmpr2 (chr.1: 60 Mb; bone morphogenic protein receptor 2, missense). Differential gene expression analysis of parental substrains via RNA-seq within the chr.1 QTL interval identified a Substrain x Treatment interaction in Abca12 expression in SC ($p = 0.02$; J > NCrl). In contrast, neither RNA-seq nor protein analysis identified any differences in Bmpr2 transcript levels (DRGs, SC, PAG), BMPR2 protein levels (DRGs), or in the downstream signaling target SMAD 1/5/9 (DRG protein expression and phosphorylation). For the chr.14 QTL interval for cold hypersensitivity, we identified two candidate genes with missense mutations, including Abcc4 (chr.14: 119 Mb) and Nalcn (chr.14: 124 Mb). Abcc4 codes for an efflux transporter that transports chemotherapeutics to influence toxicity and Nalcn ($Na+$ leak channel, non-selective) codes for a voltage-gated $Na+$ channel implicated in chronic pain and was mapped in drosophila studies for chemotherapy toxicity.

To identify biological pathways modulated by PAC in the CIPN-sensitive J substrain on D7 (chr.1 QTL), enrichment analysis identified GO enrichment terms ($p < 0.01$; padjusted<0.2) that included insulin-like growth factor receptor signaling, epithelial morphogenesis, centromeric sister chromatid cohesion, and elastic fiber (up in DRG), as well as tyrosine catabolic process (down in DRG). For SC, GO terms included skeletal muscle thin filament assembly, striated muscle hypertrophy, detection of/response to muscle stretch, skeletal myofibril assembly, muscle filament sliding, myotube cell development, and muscle tissue morphogenesis (up in SC).

Conclusions: We identified distinct genetic loci for PAC-induced mechanical hypersensitivity and cold hypersensitivity. These loci contain candidate genes/variants involved in both pharmacokinetics (transport), pain/inflammation signaling pathways, and ion channels related to pain processing. Ongoing studies seek to identify additional functional evidence that would

implicate a particular gene or variant prior to gene editing and causal validation studies.

Keywords: Nociception, Hyperalgesia, Chemotherapy-Induced Peripheral Neuropathy, Pharmacokinetics, Allodynia

Disclosure: Nothing to disclose.

P447. Characterization and Optimization of Rat Models of Osteoarthritis Pain to Profile Novel Assets in Support of the NIH HEAL Initiative Preclinical Screening Platform for Pain (PSPP)

Mark Urban*, Elizabeth Dugan, Katelyn Buban, Jennifer Hagedorn, Sarah A. Woller, Smriti Iyengar, Taleen Hanania

PsychoGenics, Inc., Paramus, New Jersey, United States

Background: The National Institutes of Health Helping to End Addiction Long-termSM Initiative, or NIH HEAL InitiativeSM, Preclinical Screening Platform for Pain (PSPP) program aims to accelerate the discovery and development of novel non-opioid, non-addictive pain therapeutics. Under the HEAL initiative, the NIH is collaborating with PsychoGenics, Inc. to screen and profile novel assets in vitro and in vivo including small molecules, biologics, natural products, and devices. Assets are screened in vivo in rat models of pain to determine efficacy, and in additional tests to examine potential adverse effects and abuse liability. Efforts in the PSPP program are also focused on the characterization and optimization of disease-specific models of pain to evaluate select assets and provide further support for specific pain indications. Here, we describe one such effort to characterize and validate rat models of osteoarthritis pain, including the monoiodoacetate (MIA) and medial meniscal tear (MMT) models.

Methods: Adult male and female Sprague Dawley rats (n = 10/group, each sex) were used in these studies. For the MIA model, MIA (0.3 – 4.5 mg) was injected intraarticularly into the left hindlimb knee joint. For the medial meniscal tear model (MMT) model, rats received a surgical procedure in which the medial collateral ligament was transected to reflect the meniscus towards the femur, and the meniscus was then cut at its narrowest point to simulate a complete tear. Behavioral pain endpoints included hind paw mechanical allodynia, knee joint mechanical allodynia, weight bearing deficits, and changes in gait. Behavioral pain phenotype in these studies was evaluated for 4-6 weeks following the induction of osteoarthritis. Pharmacology was examined by evaluating the effects of the reference analgesics morphine sulfate (3 mg/kg), ketoprofen (6 mg/kg), and duloxetine (60 mg/kg) after single and repeated administration at both early and later times following the induction of osteoarthritis likely representing initial and advanced disease states.

Data were analyzed using two-way repeated measures ANOVA with Bonferroni's or Dunnett's post hoc test when appropriate. Effects $p < 0.05$ were considered to be statistically significant. Power analysis and effect size were determined using SAS/STAT, and appropriate sample size was based on a power value of 0.8 to ensure adequate power for F-tests for two-way interactions.

Results: Intraarticular injection of MIA (0.3- 4.5 mg) into the hindlimb knee joint produced unilateral hind paw mechanical allodynia in male and female rats which was maximal at Week 2. Unilateral knee joint mechanical allodynia to pressure and pinch stimuli was observed in female, but not male, rats at Week 2. Weight bearing deficits associated with the affected hind limb were modest when measuring static weight bearing, but pronounced when measuring dynamic weight bearing with maximal effects observed at Week 1. In male rats, weight bearing deficits were stable for entire testing period through Week 6, whereas weight bearing deficits in female rats were observed during Weeks 1 and 2, and were no longer apparent by Week 4. Changes in gait were also observed in male and female rats following MIA injection at Weeks 1 and 2. In the MMT model, male rats that had received MMT surgery displayed unilateral hind paw mechanical allodynia that was maximal at Week 3, while changes in hind paw tactile sensitivity were not observed in female rats. Knee joint sensitivity to a pressure stimulus was unaffected in male and female MMT rats, and dynamic weight bearing was also unaffected in male and female rats that had received MMT surgery. Reference analgesic compounds were evaluated in the MIA model by examining effects on hind paw mechanical allodynia and weight bearing deficits at both early (Week 1) and later (Week 6) times following induction of osteoarthritis. In Week 1, single administration of morphine (3 mg/kg) significantly reduced mechanical allodynia and weight bearing deficits in male and female rats, whereas single administration of ketoprofen (6 mg/kg) or duloxetine (60 mg/kg) was less effective. In contrast, repeated administration of ketoprofen or duloxetine (4 days, b.i.d.) significantly reduced mechanical allodynia and weight bearing deficits in Week 1. In Week 6, morphine, ketoprofen, and duloxetine were ineffective in reducing weight bearing deficits.

Conclusions: The results from these studies demonstrate that a variety of pain behaviors associated with knee joint osteoarthritis can be measured using the rat MIA model, while pain behaviors in the MMT model were less robust or not observable. Evaluation of novel assets following single and repeated administration in the MIA model using multiple pain endpoints may be a viable strategy to accelerate the development of non-opioid, non-addictive therapeutics for the treatment of osteoarthritis pain. The utility of evaluating novel assets at Week 1 vs Week 6 will continue to be examined.

Keywords: Chronic Pain, Monoiodoacetate Model of Osteoarthritis, Preclinical Pharmacology

Disclosure: Nothing to disclose.

P448. A Novel Brain Permeable Epigenetic Inhibitor Ameliorates Neuropathic Pain in Mouse

Changning Wang*

Harvard Medical School, Massachusetts General Hospital, Charlestown, Massachusetts, United States

Background: Histone lysine residue acetylation/deacetylation represents an important epigenetic modification that affects gene expression without DNA sequence modification. Histone deacetylases (HDACs) are a class of amide hydrolases that catalyze a variety of substrates, including histones and other proteins. Eleven $Zn2+$-dependent HDACs have been found in mammals, including class I (HDAC1, HDAC2, HDAC3, HDAC8), class IIa (HDAC4, HDAC5, HDAC7, HDAC9), class IIb (HDAC6, HDAC10), and class IV (HDAC11), each of these isoforms has distinct functions in epigenetic regulation. Among them, HDAC11 is the most recently identified member of the class IV HDAC with accumulating evidence suggesting its connection with many diseases. A few HDAC11-selective inhibitors have been reported in recent years, yet none of them exhibited brain penetrability, nor have they been investigated in animal models of neurological disorders. Herein, we describe the development and characterization of a selective HDAC11 inhibitor PB94, which was identified by structural optimization of our lead compounds and evaluated its therapeutic efficacy on neuropathic pain.

Methods: HDAC 1-11 Enzyme Inhibition Assays. HDAC inhibition assay of target compounds was carried out at Nanosyn (Santa Clara CA, United States). Test compounds were diluted in 100% DMSO using 3-fold dilution steps. TSA was tested in an identical manner.

Chronic Constriction Injury (CCI). The mice (n = 8 in each group) were anesthetized in an anesthesia induction chamber. The left lower extremity was prepared with an alcohol swab. A 0.5 - 1 cm incision was made using a blade on the left lower extremity. The left side of the sciatic nerve was exposed in the mid-thigh. Four ligatures using 6.0 mm chronic gut sutures were loosely placed around the exposed sciatic nerve with a 1.0 - 1.5 mm interval between each ligature. Skin incision was closed with two 6-0 vicryl sutures.

Mechanical withdrawal threshold. Mice were habituated by placing on a platform with a clear chamber individually 30 minutes daily for 3 consecutive days. Mechanical paw withdrawal thresholds (PWTs)) was carried out using calibrated manual Von Frey filaments. The positive response was recorded when the mice withdrew or shacked its paw during the stimulation. A negative response was followed by testing with the next larger filament. All CCI mice were tested before surgery (baseline) and 3, 5, 7, 10, and 14 days after surgery.

Hindpaw withdrawal latency. Mice were placed on a preheated glass platform (28 - 29°C) and clear Plexiglas cubicles to acclimate to the testing room 30 minutes daily for 3 consecutive days before the testing. Paw withdrawal latency was defined as the time (seconds) from the initiation of heat exposure to the hind paw withdrawal. A cut-off time was set at 20 seconds to avoid tissue damage.

Results: A series of lead compound analogues were synthesized to investigate the structure-activity relationship (SAR). Among them, PB94 had significant HDAC11 potency and the highest HDAC11 selectivity (IC50 = 108 nM and over 39-fold over other HDAC isoforms.

Next, ADME assessments were carried out to evaluate the drug-like profiles of PB94. Our data indicated PB94 has good metabolic stability in human liver microsomal and mouse plasma, with half-lives (t1/2) of 54.6 min and 133.8 min, respectively. In addition, no significant inhibitory effects were observed on cytochrome P450 enzymes (CYPs) 1A2, 2C19, and 2D6 at 10 μM of PB94. PB94 has a reasonably good brain/plasma ratio of 2.3 at 30 min and 2.2 at 4 h post-injection by IP administrating PB94 at 1 mg/kg in C57BL/6 mice. We further tested its off-target binding in a panel of 46 targets (PDSP) and observed no significant off-target binding at 10 μM.

To examine the potential roles of HDAC11 in a pathological status, we chose a mouse neuropathic pain model, chronic constriction injury to the sciatic nerve (CCI). At 14 days after surgery, HDAC11 protein was increased in the cortex compared with sham mice, indicating that HDAC11 is critically implicated in pain and that targeting HDAC11 might provide therapeutic effects. To test these, we administered PB94 in mice that underwent CCI to assess the development of nociceptive behavior. PB94 (10 mg/kg) was able to significantly alleviate mechanical and thermal pain-like behaviors during the entire experimental period of 14 days. Importantly, there were no significant side effects noticed during PB94 treatment. Mouse body weight remained similar between saline or PB94 treated animals.

Conclusions: The promising drug-like properties of PB94 support its therapeutic potential for neurologic disorders. Since HDACs have been implicated in neurologic pain, we further determined whether PB94 treatment could ameliorate neurologic pain using the CCI mice model. The unilateral sciatic nerve chronic constriction injury surgery is associated with postoperative spontaneous pain, mechanical allodynia, and thermal hyperalgesia. In our study, a battery of tests was used to assess the analgesic effect of PB94 on pain-like behaviors, which led to findings supporting its effectiveness at 10 mg/kg. It has been indicated that HDAC11 is widely expressed in the brain in rodents. More interestingly, we found HDAC11upregulation in the somatosensory cortex of CCI mice. It is, therefore, reasonable to postulate that increased HDAC11 expression in the somatosensory cortex may be implicated in persistent nociception after CCI surgery, and inhibition of HDAC11 by PB94 in CCI could attenuate neuropathic pain.

Keywords: Epigenetics, Pain Therapeutics, CNS Drugs

Disclosure: Nothing to disclose.

P449. Time-Dependent Differences on Pain Assessments After Orexin Receptor-1 Antagonism in a Mouse Model of Sickle Cell Disease

Kimberlei Richardson*, Alexa Ryan, Nia Sweatt, Victor Apprey, Robert Taylor, Kalpna Gupta

Howard University, Washington, District of Columbia, United States

Background: The orexins are neuropeptides that are synthesized in the perifornical area (PFA), lateral hypothalamus (LH) and dorsomedial hypothalamus (DMH) and mediate arousal, sleep, energy balance, food and drug reward. Data suggest that Orexin–A neuropeptide has an analgesic effect on inflammatory pain upon binding the orexin receptor-1. It may also affect mechanisms underlying the maintenance of hyperalgesia associated with neuropathic pain. Data from our prior study suggests that subpopulations of orexin neurons are preferentially recruited during behavioral assessments for hyperalgesia. We seek to determine whether there are differences in pain assessments after orexin receptor-1 antagonism (SB-334867) and whether that pain is potentiated in sickle mice that express exclusively (99%) human sickle hemoglobin (HbSS-BERK) and age-/gender-matched controls (HbAA-BERK).

Methods: Female transgenic HbSS-BERK and HbAA-BERK mice (n = 6/group, 20-30 g) were habituated to each test protocol for thermal/heat and mechanical hyperalgesia. Thermal/heat hyperalgesia was monitored using the Plantar Test (Hargreave's Apparatus) for thermal stimulation. Latency was measured as the time (in seconds) for the mice to withdrawal their hindpaws from the heat stimulus. Mechanical hyperalgesia was monitored by the Von Frey filament test. Paw withdraw frequency was measured by the number of times that the mice lifted their hindpaws after tactile stimulation from the filament. The behavioral data were recorded prior to SB-334867 (20 mg/kg, IP), 1 hour, and 24 hours post-injection.

Results: Significant time-dependent differences were observed in heat latency after SB-334867. Mean heat latency for HbAA mice was not significantly different between the various time points(p-value=0.103). Mean heat latency for HbSS-BERK mice was significantly different between time points (p-value=0.004) and post-hoc tests showed significant reductions in heat latency in HbSS versus HbAA-BERK mice before SB-334867 ($p < 0.05$) and 1 hour post-SB-334867 ($p < 0.005$). There was a significant reduction in heat latency exclusively in HbSS-BERK mice when comparing observations pre-SB versus 1 hour post-SB-334867 ($p < 0.05$). Statistical analyses are underway to determine time-dependent differences for mechanical hyperalgesia.

Conclusions: These findings suggest differential orexin receptor responsiveness in HbSS-BERK mice vs. HbAA-BERK mice. These data may provide a mechanism for the increased hyperalgesia in HbSS-BERK mice, which contributes to neuropathic pain observed in sickle cell disease.

Keywords: Hyperalgesia, Orexin Receptor Antagonist, Sickle Cell Disease

Disclosure: Nothing to disclose.

P450. Better Subject Recruitment: Can Aligned Messaging Improve Sample Accession?

Gary Sachs*, Jenicka Engler, D. Thorpe, Miriam Evans, D. Fagundo, S. Starling, S. Nicholas, H. Zandi, M. Moy, A. Vittoria

Signant Health, Lincoln, Massachusetts, United States

Background: Sample accession is key to clinical trial success. Social media advertisement is now a common means of initial recruitment outreach, with considerable success in increasing the number of pre-screening study inquiries, yet slow enrollment remains a chronic problem. Because efforts to combat slow recruitment by increasing compensation for time and travel risks increasing the enrollment of inappropriate participants and so called "professional subjects," the goal of enrolling an appropriate study sample can be compromised by recruitment efforts to attract "clinical trial seeking" volunteers rather than participants representative of "treatment seeking" patients.

Notably, Major Depressive Disorder (MDD) clinical trial recruitment is regarded as particularly challenging because depressive symptoms can be obstacles to attending screening visits.

To begin addressing this complex issue, we ask a simple question: Does the "promise to refer" for post-study treatment impact show rates and participant quality for MDD trials?

Most current quality control methodologies aim to disqualify inappropriate enrollment in clinical trials for MDD after the screening assessment. This pilot study examines the impact of prescreening recruitment messaging more aligned with the interests of "treatment seeking" subjects which clearly states an offer of referral for post-study clinical care as a benefit of clinical trial participation.

This pilot experiment explores differences at various points in the recruitment-enrollment process (recruitment funnel) between participants who were promised referrals (experimental group) versus a control group who received usual study recruitment messaging.

Methods: Starting in mid-May 2022, prospective research participants were recruited and randomized using the Facebook A/B testing algorithm. The algorithm randomly assigned users to a control group shown our usual Facebook advertisement content (information about MDD and clinical trials) or the experimental group shown advertisement text about MDD clinical trials and the promise that all respondents would receive referral to treatment providers and information about clinical resources regardless of trial eligibility. Both groups were offered phone screening interviews with Adams Clinical site staff and subsequently an in person pre-screening appointment. For those in the experimental group, recruiters and clinicians reiterated the promise of resources and referrals via IRB-approved scripts. Those in the experimental group who completed a phone screening but were not study candidates or declined were sent information for their local NAMI. Those who attended their in-person prescreening visit and were ineligible or declined were given a treatment referral letter with their prescreening assessment results and treatment recommendations, NAMI information, plus at least 2 referrals for in-network local clinicians listed as accepting new referrals on psychologytoday.com. The two groups were compared on rate of completing phone screens, completing the prescreening process and study eligibility using Chi Square.

Results: A total of 1,358 prospective research participants clicked on Adams Clinical's Facebook ads and submitted their contact information online as part of this A/B testing experiment ($n = 751$ in the experimental group, and $n = 607$ in the control group). Participants in the experimental group were significantly more likely to complete a phone screening (22%) than the control group (16%), $X2$ (1, $N = 1358$) $= 6.46$, $p = .011$. Participants in the experimental group were significantly more likely to have a prescreening visit scheduled (83%) than the control group (68%), $X2$ (1, $N = 265$) $= 8.03$, $p = .005$. The effect on prescreening visit attendance reached marginal significance, with participants in the experimental group being more likely to attend their prescreening visit (49%) than the control group (35%), $X2$ (1, $N = 190$) $= 3.19$, $p = .074$. The effect on study eligibility also reached marginal significance, with participants in the experimental group being more likely to be eligible (48%) than the control group (27%), $X2$ (1, $N = 85$) $= 2.78$, $p = .096$.

At this time, scheduled prescreens and screening visits from both groups are still upcoming. Data from all participants will be available prior to the meeting, allowing full data on prescreening attendance and consenting to study participation to be presented in this poster.

Conclusions: To our knowledge this pilot study represents the first empirical data on efforts to improve recruitment by offering a value proposition appealing to treatment seeking patients. We found a promise to refer and provide resources for prospective MDD study participants had positive impacts at all points examined in the recruitment funnel.

Our results suggest that articulating and delivering on this value proposition has the potential to speed up trial recruitment by significantly improving potential participants' likelihood of completing a phone screen and in-person prescreening visit. More work is needed to determine if the increase in willingness to engage with the clinical research screening process is accompanied by improved study participation rates, improved sample quality and enhanced potential for signal detection.

Keywords: Clinical Trial Methodology, Recruitment, Social Media Use, Mood Disorders

Disclosure: Signant Health: Employee (Self)

P451. Risk of Major Malformations in Infants After First-Trimester Exposure to Stimulants: Results From the Massachusetts General Hospital National Pregnancy Registry for Psychiatric Medications

Mercedes Szpunar*, Marlene Freeman, Lauren Kobylski, Ella Rossa, Peter Gaccione, David Chitayat, Sonia Hernandez-Diaz, Adele Viguera, Lee Cohen

Massachusetts General Hospital, Harvard Medical School, Boston, Massachusetts, United States

Background: The prevalence of attention-deficit/hyperactivity disorder (ADHD) in adult women is 3-4%. ADHD is highly comorbid with other psychiatric disorders such as mood, anxiety, and substance use disorders. For reproductive-aged women, the treatment of ADHD with stimulant medications may be considered during pregnancy or breastfeeding, although historically, data are lacking to inform these decisions. The aim of this investigation was to determine the risk of major malformations in infants after first-trimester stimulant exposure.

Methods: The Massachusetts General Hospital National Pregnancy Registry for Psychiatric Medications was established in 2008 to increase reproductive safety knowledge of psychiatric medications using rigorous, prospective data collection. The Registry systematically ascertains information from its participants including demographic information, medical and psychiatric history, use of prescription medications, and other information relevant to fetal outcomes. Participants provide verbal informed consent and are interviewed twice during gestation and again at approximately 3 months postpartum. The primary outcome of interest is the presence of a major malformation identified within 6 months after birth. Redacted cases of major malformations are reviewed by a dysmorphologist blinded to medication exposure. Women taking

a stimulant during the first trimester of pregnancy (n = 233) were compared with controls (n = 1755 women) taking other psychiatric medication(s) during pregnancy to treat mental health disorders. The study is registered with ClinicalTrials.gov (NCT01246765).

Results: N = 1988 women who were eligible for this analysis, including n = 173 exposed to mixed amphetamines, n = 40 exposed to lisdexamfetamine, n = 45 exposed to methylphenidate, and n = 3 exposed to dexmethylphenidate; the comparison group included n = 1755 women. Because of twin gestations, a total of 235 infants were exposed to one or two stimulant medications. The odds ratio of a major malformation among infants after first-trimester exposure to any stimulant was 0.57 (95% CI 0.17-1.85) compared to controls. The odds ratio of a major malformation after exposure to mixed amphetamines or lisdexamfetamine was 0.70 (95% CI 0.70-2.28). There were no major malformations observed in the infants exposed to lisdexamfetamine, methylphenidate, or dexmethylphenidate. The prevalence of major malformations after first-trimester exposure to mixed amphetamines was 1.2%.

Conclusions: Although preliminary, this analysis from an ongoing pregnancy registry provides reassurance that these prescription stimulants do not appear to have major teratogenic effects.

Keywords: ADHD, Stimulants, Pregnancy

Disclosure: Intepros: Employee (Spouse)

P452. Psychiatric Disorders and Income: A Population-Based, Longitudinal Study

Mark Weiser*, Ariel Asper, Adam Noy, Michael Davidson, Itzhak Levav, John M Davis

Sheba Medical Center, Ramat Gan, Israel

Background: Data suggest that having a psychiatric diagnosis is negatively associated with life-long income. We examined the effect of having a psychiatric diagnosis between ages 25-34, on income through ages 63-72.

Methods: Baseline assessments were obtained from a population-based, epidemiological study of a random sample of 5000, 25-34 year-olds, conducted in Israel during the 1980s, the subjects are now aged 63-72. Data on mean annual income between 1983 to 2018 were obtained from the Israeli Social Security Administration. For each participant the median of his/her lifetime mean annual income was calculated; for each diagnostic group the mean of the medians of the participant was calculated. The difference (in standard deviations) was calculated between the mean annual income between healthy participant and each diagnostic group.

Results: Healthy participant had the highest mean annual income. Mean income worked varied by diagnosis: Anxiety: -.2 SD, Depression: -.43 SD, Labile Personality Disorder -.49 SD, Bipolar Disorder -.52 SD, Schizotypal Personality Disorder -.58 SD, Antisocial personality disorder -.84 SD, Schizophrenia -.96 SD.

Conclusions: Over their lifetime, young adults diagnosed with psychiatric disorders earn significantly less money, compared with young adults who do not suffer from psychiatric disorders. These longitudinal data indicate that having a psychiatric diagnosis increases risk for low income, but cannot dismiss the possibility that in parallel, low income increases risk for having a psychiatric disorder.

Keywords: Psychiatric Disorders, Income, Population-Based

Disclosure: Jansen: Advisory Board (Self), Teva: Consultant (Self), Dexcel: Consultant (Self), Lundeck: Advisory Board (Self), Minerva: Contracted Research (Self), MSD: Consultant (Self), Pfizer: Honoraria (Self), Orion: Consultant (Self), Clearmind: Consultant (Self)

P453. The Effects of MDMA and Methamphetamine on Feelings of Closeness and Connection During Semi-Structured Conversations With Strangers

Hanna Molla*, Tang Li, Sonja Lyubomirsky, Harriet de Wit

University of Chicago, Chicago, Illinois, United States

Background: MDMA is classified as an "empathogenic" drug, as it increases feelings of empathy and closeness to others. These effects are thought to be partly mediated by increased oxytocin release via enhanced serotonergic transmission. Despite its prosocial effects, few studies in humans have examined MDMA in the context of interpersonal interactions. The objectives of this study were to determine the effects of MDMA on feelings of closeness and connection during a semi-structured dyadic conversation, and to investigate the relations between feelings of closeness and oxytocin levels. To investigate pharmacological specificity, a separate group of subjects was tested with the prototypic stimulant methamphetamine.

Methods: Two studies were conducted in separate sets of healthy volunteers. In within-subject, double-blind, placebo-controlled designs, subjects aged 18-35 received a single dose of MDMA (100 mg) or placebo (Study 1; n = 18), or a single dose of methamphetamine (20 mg) or placebo (Study 2; n = 19), during two in-lab sessions separated by 72 hours. During peak drug effect, they engaged in a 45 min semi-structured conversation with a novel same-sex partner. Subjective mood, physiological, and oxytocin levels were obtained at regular intervals, and subjects rated feelings of closeness and connection with their partners at the end of each session and one week later.

Results: Both MDMA and methamphetamine increased ratings of feeling connected to the partner, and increased ratings of enjoyment and meaningfulness of the conversation, relative to placebo (Study 1 MDMA; p = 0.009 and Study 2 methamphetamine p = 0.01). Both MDMA and methamphetamine increased subjective ratings of "sociable," "loving," and "friendly." Only MDMA increased ratings of "trusting," "grateful," and "appreciated," and only methamphetamine increased ratings of "understood" MDMA (p = 0.001), and to a lesser extent methamphetamine (p = 0.02) increased salivary oxytocin levels. Notably, oxytocin levels were positively correlated with feelings of closeness after MDMA but not after methamphetamine (Study 1; r = 0.52, p = 0.03).

Conclusions: Both MDMA and methamphetamine increased feelings of closeness and connection to strangers during controlled interpersonal interactions, and both drugs increased subjective ratings of sociability. However, these feelings were related to oxytocin only after MDMA, suggesting that the mechanisms mediating prosocial effects may differ across drugs. This study provides a model for future studies assessing effects of drugs on interpersonal interactions.

Keywords: MDMA, Social Interaction, Oxytocin, Closeness, Stimulant

Disclosure: Nothing to disclose.

P454. Inflexible Thinking Style Predicts COVID-19 Vaccine Hesitancy

Naomi Fineberg*, Pellegrini Luca, Aaron Clarke, Jemma Reid, Keith Laws

University of Hertfordshire, Welwyn Garden City, United Kingdom

Background: Mass vaccination against COVID-19 heralded the return to a state of normality following the recent pandemic. The success of this initiative needs to be set against the backdrop of a

substantial minority who decline vaccination. Understanding the factors influencing vaccine attitudes and intentions represents an important plank in the armory against this and any future pandemics (1). Objective cognitive tasks hold certain advantages over clinical measurements as they are potentially less subject to rater-related error and may provide a window on the underpinning neuropsychological mechanisms. Few studies have investigated cognitive function in relation to vaccine hesitancy. One such study identified a relationship with executive function, broadly defined, using a Stroop task and the Montreal Imaging Stress Task (2). Another (3) identified a relationship with self-control on a Stroop-task. A third (4) found evidence suggesting a relationship with flexible adaptation to new concepts. In a fourth (5), parents' psychological flexibility and positive coping-style were linked with reduced COVID-19 vaccine-hesitancy among a pediatric sample. This is the first study to our knowledge to investigate the relationship between cognitive inflexibility and COVID-19 hesitancy in a large adult sample using a specific digital neurocognitive task (Wisconsin Card Sort Task; WCST).

Methods: This study is part of a larger research programme investigating post-pandemic adjustment. The study was approved by the University-of-Hertfordshire-Health-and-Human-Science-Ethics-Committee-With-Delegated-Authority-(ECDA)-(LMS/PGR/UH/04554) and a protocol was preregistered on Open-Science-Framework-(OSF)-https://doi.org/10.17605/OSF.IO/XD5WZ. We surveyed adults UK-adult-population (>18 years) between June 2021 – July 2022 (using Gorilla-software) during a period when lockdown restrictions had eased. In order to recruit a sample that included traditionally underserved groups such as those with mental disorders, diverse groups were targeted. Sociodemographic information (age, gender, education and ethnicity) was collected. Cognitive inflexibility was measured using a digital version of the WCST. For this analysis, we focused on "WCST perseverative errors" as the most specific item for capturing attentional set-shifting problems. Vaccine hesitancy was measured with the Oxford Covid Vaccine Hesitancy Scale (OCVHS). We ran correlational and regression analyses to detect potential associations between cognitive inflexibility and vaccine hesitancy.

Results: A sample of 207 individuals completed the survey and the WCST. The mean age of the sample was 37.3 years, 68% were females, 61% defined themselves as white ethnicity with 28% Asian, 5.8% South American and 6.3% of African background. The mean Total OCVHS score was 12.8 (SD:6.9), which is in line with the mean score found in a larger population-based sample (mean 13.6 (SD: 7.3))(6). A statistically significant correlation was found between WCST perseverative errors and Total OCVHS (Pearson's r, $p = 0.003$). We ran a multivariate regression analysis in which we included OCVHS as the outcome, perseverative errors as the predictor and sociodemographic factors thought to have an impact on vaccine hesitancy (age, gender, education, ethnicity) as covariates. Perseverative errors significantly predicted increased vaccine hesitancy even after controlling for these covariates ($p = 0.01$).

Conclusions: Cognitive inflexibility, measured with an objective computerized neurocognitive task in a large adult population sample, was a significant predictor of vaccine hesitancy, which applied irrespective of age, gender, level of education and ethnicity. Cognitive inflexibility is usually an enduring trait (7) and might represent a "red flag" for vaccine hesitancy, for use to identify those most at risk of refusing vaccination in the current and future pandemics. Public health education strategies should take into account the impact of an inflexible thinking style on the decision-making of those most at risk of vaccine hesitancy and adapt interventions accordingly.

References:

1. Wang Y, Zhang X. Frontiers in Psychology 2021;12

2. Acar-Burkay S, Cristian D-C. Soc Sci Med 2022;301:114911. https://doi.org/10.1016/j.socscimed.2022.114911

3. Cao and Li 2022, Pers Indiv Diff 188:11147

4. Mazzuca C, et al. 2021. https://doi.org/10.31234/osf.io/4ndb8.

5. Wang Y, Zhang X. Frontiers in Psychology 2021;12.

6.Freeman D, Loe BS, Chadwick A, Vaccari C, Waite F, Rosebrock L, et al. Psychological Medicine 2020:1–15. https://doi.org/10.1017/S0033291720005188

7. Chamberlain SR, Fineberg NA, Menzies LA, Blackwell AD, Bullmore ET, Robbins TW, et al. Am J Psychiatry 2007;164:335–8. https://doi.org/10.1176/appi.ajp.164.2.335.

Keywords: Cognitive Inflexibility, Vaccine Hesitancy, Public Health, COVID-19

Disclosure: Nothing to disclose.

P455. The National Pregnancy Registry for Psychiatric Medications: Effects of Fetal Exposure to Second-Generation Antipsychotics on Risk for Major Malformations

Lee Cohen*, Marlene Freeman, Lauren Kobylski, Ella Rossa, Charlotte Clifford, Sonia Hernández-Díaz, David Chitayat, Adele Viguera

Mass General Hospital, Harvard Medical School, Boston, Massachusetts, United States

Background: Second-generation antipsychotics (SGAs), or atypical antipsychotics, are widely used by women of reproductive age to treat a number of psychiatric disorders. The National Pregnancy Registry for Psychiatric Medications (NPRPM) is a systematic prospective pharmacovigilance program used to collect reproductive safety data on second-generation antipsychotics, as well as other classes of psychotropic medications, in order to inform the care of reproductive aged women with psychiatric disorders.

Methods: Data are prospectively collected from women, aged 18-45 years, with histories of psychiatric disorders. Two phone interviews are conducted during pregnancy and a third interview is completed at 3 months postpartum. Enrollment and longitudinal follow-up of participants is ongoing. In this analysis, the exposed group is comprised of women who took an SGA during the first trimester of pregnancy. The comparison group is composed of women who did not take this class of medication during pregnancy but who were prenatally exposed to other psychotropics. Information regarding the presence of major malformations is abstracted from medical records and identified cases of potential major malformations are adjudicated by a dysmorphologist blinded to drug exposures and psychiatric diagnoses.

Results: As of July 25th, 2022, 2676 women were enrolled in the NPRPM, including 1031 in the exposure group and 1551 in the comparison group. Medical records were obtained for 78% of participants. A total of 1997 participants (787 exposed to an SGA in the first trimester, 1210 unexposed to an SGA during pregnancy) were eligible for analysis, having completed the postpartum interview. Of 810 infants in the exposure group, 22 confirmed major malformations were identified. 19 malformations were identified in the control group, which consisted of 1234 infants. No consistent pattern of malformation was seen in either group. The absolute risk of major malformations was 2.72% in the exposure group and 1.54% in the comparison group. The odds ratio was OR = 1.79 (95% CI: 0.96 – 3.32).

Conclusions: The NPRPM offers a systematic way to collect prospective reproductive safety information which informs the care of women who may use second-generation antipsychotics to sustain psychiatric well-being. Current data suggest that it is unlikely that SGAs as a class have a major teratogenic effect. Future analyses will aim to better estimate risk of major malformations with even larger sample sizes and more generalizable cohort characteristics. We also continue to assess the reproductive safety of individual medications in this class. Additionally, we are studying other classes of

medications, such as newer antidepressants and medications to treat ADHD and insomnia.

Keywords: Pregnancy, Bipolar Disorder, Atypical Antipsychotics, Depression, Reproductive Psychiatry

Disclosures: Alkermes Inc., Johnson and Johnson/Janssen Pharmaceuticals Inc., Otsuka America Pharmaceutical, Inc., Sage Therapeutics, Sunovion Pharmaceuticals Inc., Supernus Pharmaceuticals, Teva Pharmaceutical Industries Ltd., Forest/Actavis/Allergan:, AstraZeneca Pharmaceuticals, AuroMedics Pharma LLC, Aurobindo Pharma, Ortho-McNeil-Janssen Pharmaceuticals Inc., Pfizer Inc., MGH CTNI, Brain and Behavior Research Foundation, National Institute on Aging, National Institutes of Health, SAGE Therapeutics: Other Financial or Material Support (Self), JDS Therapeutics LLC: Consultant (Self)

P456. Effects of Sex on the Relationships Between Visual Food Cue Ratings and Eating Behaviors, Weight, and Mood

Kristina Legget*, Marc-Andre Cornier, Lauren Sarabia, Eve Delao, Susan Mikulich, Christina Erpelding, Tessa Mitchell, Jason Tregellas

University of Colorado School of Medicine, Aurora, Colorado, United States

Background: Obesity rates are rapidly rising, giving rise to myriad health and quality-of-life concerns. Responsivity to food cues is a key factor underlying eating behaviors that may contribute to obesity. A critical factor in understanding these relationships, however, is how response to food cues may differ between men and women, including sex-based differences in factors that may relate to food cue responsivity, such as weight, eating behaviors, and mood. As such, the goal of the current study was to investigate sex-based differences in both appeal and "desire to eat" high-and low-calorie foods, including how sex may influence relationships between food cue ratings and BMI, eating behaviors, and mood measures.

Methods: 334 adults (164 men, 169 women) completed this study, for which they completed an online survey in a neutral state of hunger. Survey measures included weight and height (from which BMI was calculated); the Three Factor Eating Questionnaire (TFEQ), which assesses trait-based eating behaviors relating to restraint, disinhibition, and hunger; the Eating Attitudes Test 26 (EAT-26), which assesses eating disorder-related behaviors; the Center for Epidemiologic Studies-Depression Scale (CESD-R), which assesses symptoms associated with depression over the past week; the Perceived Stress Scale (PSS), which assesses feelings of stress over the past week; and visual analog scale (VAS) measures assessing current hunger ("how hungry are you?" from "not at all hungry" to "extremely hungry") and satiety ("how full do you feel right now?" from "not at all full" to "extremely full"), on a 0-100 scale. After these questionnaires were completed, participants began the Food Pictures Task, for which they viewed 96 different food images (48 high-calorie and 48 low-calorie) and were asked to rate how appealing each image was and how much they desired to eat each of the foods, using a visual analog scale (0-100). Independent samples t-tests were used to assess group differences (men vs. women) in questionnaire scores (TFEQ: restraint, disinhibition, and hunger scores; EAT-26 total eating disorder-related behavior score; CESD-R depression score; PSS perceived stress score; VAS hunger and satiety scores) and ratings of appeal and "desire to eat" on the Food Pictures Task, separately for high-calorie (HC) and low-calorie (LC) foods. Regression models investigated how questionnaire scores were associated with HC and LC ratings of appeal and desire to eat from the Food Pictures Task. Sex-based interactions were included in analyses; models with non-significant sex-based interactions were re-evaluated without the interaction component.

Results: Two women were excluded from further analyses as outliers for BMI (BMI > 60 kg/m2), with 27 (12 men, 15 women) excluded due to being outliers on time to complete the survey (> 75 min). As such, 306 participants were included in the final sample (151 men, 155 women). Compared to men, women in the sample reported significantly greater dietary restraint ($p = 0.004$), disinhibition ($p < 0.001$), and trait-based hunger ($p = .044$), as measured by the TFEQ. Significantly higher scores on the EAT-26 were also observed in women compared to men ($p < 0.001$), as were significantly higher levels of perceived stress ($p < 0.001$) and depression ($p < 0.001$). We did not observe sex-based differences in "desire to eat" HC or LC foods or HC food appeal ($p > 0.05$ for all), but women did report greater appeal of LC foods compared to men ($p = 0.002$). A significant interaction with sex was observed in the relationship between perceived stress and HC food appeal ($p = 0.006$), such that higher stress was associated with increased HC appeal in men, but reduced HC appeal in women. For LC food appeal, we observed a significant sex-based interaction with disinhibition ($p = 0.024$), such that greater disinhibition was associated with lower LC appeal in women, but higher LC appeal in men. In both groups, increased satiety was associated with a decreased desire to eat LC foods, but a sex-based interaction was observed, such that stronger associations were observed in women compared to men ($p = 0.049$). Across sex, increased BMI was associated with increased HC food appeal ($p = 0.042$), with increased depression scores associated with increased desire to eat HC foods ($p = 0.008$). No association between BMI and LC foods was observed, but increased depression scores were associated with increased desire to eat LC foods ($p = 0.008$) and increased perceived stress was associated with increased LC food appeal ($p = 0.009$).

Conclusions: We observed greater dietary restraint, disinhibition, and trait-based hunger in women compared to men, in addition to greater appeal of LC foods. Higher scores on eating disorder-related behaviors, depression, and perceived stress were also observed in women compared to men. Interestingly, we observed a sex-based interaction between perceived stress and HC appeal, with increased stress associated with reduced HC appeal in women, but increased HC appeal in men. This may indicate differential impacts of stress on eating behaviors in women compared to men, which may suggest stress as an important factor to consider in individualized weight management approaches. Across groups, BMI was associated with greater HC appeal, stress was associated with increased LC appeal, and depression with both HC and LC desire, further supporting the importance of considering the role of mood in eating behaviors and weight maintenance approaches.

Keywords: Obesity, Sex-specific Effects, Food Cues, Eating Behavior, Mood

Disclosure: Nothing to disclose.

P457. Adolescent Stress Impairs Postpartum Social Behavior via Glucocorticoid Receptor-Mediated Anterior Insula-Prelimbic Pathway

Kongpyung Kim, Shin-ichi Kano, Minae Niwa*

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

Background: Postpartum social behavior is important not only for the mother's health, but also for the child's development. Unfortunately, postpartum social behavior is sensitive to stress that can occur at different points in the mother's lifespan. Adolescent stress can be associated with changes in later

postpartum behavior, including social behavior. Nonetheless, the neural circuit mechanisms by which adolescent stress leads to postpartum changes in social behavior are unclear. We recently found that mice exposed to social isolation in late adolescence (SILA), which alone causes no endocrine or behavioral changes, show long-lasting behavioral deficits only when accompanied by pregnancy and delivery. The behavioral deficits in dams exposed to SILA (stressed dams) were caused by an aberrantly sustained elevation of corticosterone levels during the postpartum period. More importantly, we have shown that short-term post-delivery treatment with a glucocorticoid receptor (GR) antagonist is sufficient to ameliorate the behavioral deficits observed in stressed dams (Niwa et al BioRxiv 2021). Using this mouse model, we also found that SILA, in conjunction with pregnancy and delivery, leads to hypofunction of anterior insula-prelimbic cortex (AI-PrL) glutamatergic pathway, which in turn alters activity patterns in PrL, resulting in behavioral changes in social novelty recognition (Kim and Niwa, ACNP 2021). Based on these findings, the present study further examined whether enhanced corticosterone signaling in AI-PrL pathway leads to social behavioral changes in stressed dams in the postpartum period.

Methods: AAVretro-CaMKIIa-EGFP and a mix of CRE-DOG viruses (AAV1-EF1a-N-Cretrcint G and AAV-EF1a-C-Creint G) were bilaterally injected to PrL and AI of mice floxed with the GR gene (GRfl/fl mice), B6J, or Ai14 mice at eight weeks old. AAV1-Flex-tdTomato was also injected into AI for a validation study with B6J mice. For the Cre recombinase dependent on GFP (CRE-DOG) validation studies, mice were perfused at one, two, or three weeks after virus injections. EGFP +, tdTomato+EGFP +, EGFP + GR +, mCherry +, or mCherry+GR + cells were counted. For the behavioral cohort, animals were exposed to SILA (from 5 to 8 weeks of age) after surgery. Each mouse was then mated with a healthy C57BL/6J male and gave birth to pups. Three-chamber social interaction tests (SIT) were performed one week postpartum. Two hours after the last behavioral test, mice were perfused for c-Fos staining.

Results: By combining the GFP-dependent Cre recombinase system and AAV-mediated retrograde tracing in GRfl/fl mice, pathway-specific GR knock-out (GR-KO) mice were successfully generated. The CRE-DOG method, in which GFP-binding proteins were used for the molecular assembly of Cre recombinase on a GFP scaffold, allowed us to express Cre recombinase in a GFP-dependent manner. We validated that Cre recombinase was successfully expressed in AI-PrL pathway when AAV-retro-CaMKIIα-EGFP and a mix of CRE-DOG viruses (AAV-EF1a-N-Cretrcint G and AAV-EF1a-C-Creint G) were injected into PrL and AI, respectively. As time passes after the injections, a gradual increase in the expression level of EGFP was observed in the AI, accompanied by a gradual decrease in the expression level of GR in AI neurons expressing EGFP. Such expression changes were not observed when AAV-retro-CaMKIIα-mCherry was injected instead of AAV2-retro-CaMKIIα-EGFP as a control virus. Deletion of GR in AI-PrL pathway ameliorated the behavioral changes in the social novelty-trial, but not the sociability-trial, of SIT in stressed dams. AI-PrL-specific GR-KO also normalized the reduced c-Fos immunoreactivity after social novelty-trials in stressed dams. These findings suggested that activation of GR signaling in AI-PrL pathway, but not in PrL itself, might play a causal role in PrL dysfunction and subsequent behavioral changes in social novelty recognition in the postpartum period.

Conclusions: We successfully generated pathway-specific GR-KO mice using the CRE-DOC method. Using these GR-KO mice, we elucidated the causal role of GR-mediated AI-PrL pathway in postpartum behavioral changes during social novelty recognition. Further research on how GR signaling specifically regulates AI-PrL function and recognition of social cues would facilitate our mechanistic understanding of the pathological trajectory of adolescent stress leading to abnormal social behavior in the postpartum period.

Keywords: Adolescent Stress, Social Behaviors, Glucocorticoid Receptor, Anterior Insula, Prelimbic Cortex

Disclosure: Nothing to disclose.

P458. The Role of Hypocretin/Orexin Neurons in Social Behavior

Derya Sargin*, Matthew Dawson, Dylan J. Terstege, Naila Jamani, Dmitrii Pavlov, Mio Tsutsui, Jonathan R. Epp, Gina M. Leinninger

Hotchkiss Brain Institute, Alberta Children's Hospital Research Institute, University of Calgary, Calgary, Canada

Background: Intraspecific social interactions are integral for survival and maintenance of society among all mammalian species. Yet, our understanding of the neural systems and mechanisms involved in the establishment of social connectedness and the consequences of the detrimental effects of social isolation are limited. Since their initial discovery as regulators of sleep/wakefulness and appetite in the brain, the hypocretin/orexin neurons have also been shown to play an essential role in modulating energy homeostasis, motivated and emotional behavior. These neurons are located exclusively in the hypothalamus that regulates complex and goal-directed behaviours. The hypothalamus has previously been shown to play an important role in the modulation of social behavior by encoding internal states. However, the information for the role of hypocretin neurons in social behavior and deficits is limited.

Methods: We infused AAV encoding GCaMP6s into the LH of female and male Hcrt-cre mice and performed fiber photometry to record the activity of hypocretin neuron population during social interaction. We next inhibited the activity of hypocretin neurons using optogenetics and determined the necessity of these neurons for social behavior in female and male mice. Using a mouse model of chronic social isolation, we additionally determined how hypocretin neuron activity is affected in mice with social deficits.

Results: We identified hypocretin/orexin neurons to exhibit a robust increase in activity in response to social interaction in female and male mice. We demonstrate that the activity of hypocretin neuron population is differentially encoded during interaction between familiar and stranger conspecifics. Moreover, the optogenetic inhibition of hypocretin neuron activity during social behaviour leads to a reduction in the amount of time mice are engaged in social interaction in a sex specific manner. We also show that isolated mice have deficits in social behavior and disruption in hcrt neuron activity.

Conclusions: Together, these data situate the hypocretin/orexin system as the part of a larger network that plays an integral role in the modulation of social behaviour. Here, we will additionally discuss the implications of these findings in an animal model of chronic social isolation that develops long-term social impairments.

Keywords: Orexin/Hypocretin, Social Isolation, Social Deficits, Fiber Photometry, Optogenetics

Disclosure: Nothing to disclose.

P459. The Epigenetic Reader Plant Homeodomain Finger Protein 21B (PHF21B) Regulates Social Memory, Associated Behaviors, Glutamatergic Neurotransmission, and Synaptic Plasticity–Related Genes in the Hippocampus

Julio Licinio*, Eunice Chin, Qin Ma, Hongyu Ruan, Camile Chin, Aditya Somasundaram, Ma-Li Wong

State University of New York, Syracuse, New York, United States

Background: We have reported that the PHF21B gene is located in the genomic region of human chromosome 22q13.31. The 22q13.3 deletion syndrome, also known as Phelan-McDermid syndrome, is characterized by developmental delays, moderate to profound intellectual disability, decreased muscle tone (hypotonia), absent or delayed speech as well as autism or autistic-like behavior that affects communication and social interaction. We have hypothesized that while the full 22q13.3 deletion causes a severe syndrome, a specific decrease of PHF21B function might cause a more subtle and specific phenotype, possibly related to social interaction. In a model of stress, we reported significantly decreased hippocampal Phf21b gene expression in rats resilient to chronic restraint stress compared to non-chronically stressed rats. The PHF21B (aka PHF4) gene, a member of PHD finger proteins encodes a histone reader and is expressed in several brain regions, including the frontal cortex and the hippocampus; however, its functions in the brain have remained unclear. We generated a Phf21b knockdown mouse model to mechanistically understand how this gene regulates behavior, including social interaction, and hippocampal functions.

Methods: All animal experiments were performed according to approved protocols by the State University of New York Upstate Medical University, South Australian Health and Medical Research Institute, Flinders University, and University of Adelaide. Male and female mice were studied. The following approaches were used – 1) CRISPR/Cas9 technology (SA Genome Editing Facility University of Adelaide, South Australia (SA), Australia) to generate a Phf21b mutant mice in which the exon 4 of the mouse Phf21b gene was mostly deleted and a frame shift mutation generated a premature stop codon. 2) A battery of cognitive and emotional behavioral tests was used to determine the effects of Phf21b deficiency (Phf21bΔ4/Δ4) compared to littermate wildtype (WT, Phf21b + /+) mice. 3) Standard whole-cell patch clamp was used to study long-term potentiation in hippocampal CA1 pyramidal neurons. 4) Transcriptome profile was performed using hippocampal tissues, followed by differential gene expression analysis and qRT-PCR confirmation studies. 5) Immunoblotting, immunohistochemistry, and Golgi staining. 6) Histone peptide array.

Results: Phf21b WT and Phf21bΔ4/Δ4 mice were in generally good health. Phf21bΔ4/Δ4 expressed 60% less PHF21B than WT mice. Male and female mice had similar changes in behaviors, and their combined data were used for data analyses. Phf21b deficiency resulted in very specific social memory deficits (increased social novelty in the 3-chamber social test ($P < 0.01$); no decrease interaction time during the habituation trials in the 5-trial social memory test and the score difference was significantly lower ($P < 0.05$). Importantly, these animals did not display other memory, cognitive, or emotional differences. PHF21B deficit led to thinner cortices ($P < 0.001$) with lower astrocyte number ($P < 0.0001$), and reduced neurogenesis (DCX + cells, $P < 0.001$). Phf21bΔ4/Δ4 hippocampi had decreased synaptic protein expression (fewer PSD95 + clusters/unit area, $P < 0.001$; smaller PSD95 + puncta size, $P < 0.01$), diminished glutamatergic neurotransmission (reduced I/O relationship, $P < 0.001$; impaired LTP), and GluN2B ($P < 0.05$) and Grin2b ($P < 0.01$) levels. RNAseq showed that PHF21B modulates the expression of neurotransmission genes. Histone peptide studies showed that PHF21B regulates transcription through H3K9ac, H3K9Me2, and CREB (cAMP response element-binding protein), and that it interacts with H3K36me3.

Conclusions: Our results demonstrate that PHF21B regulates social memory and that reduced PHF21B function brings about impaired hippocampal long-term potentiation. There are fewer AMPAR subunit GLUR1-expressing clusters in hippocampal tissues of Phf21bΔ4/Δ4 mice, resulting in decreased glutamatergic neurotransmission. We show that PHF21B is a critical upstream regulator of synaptic plasticity-related genes, functioning as an epigenetic reader. Furthermore, we characterized a potentially novel interaction of PHF21B with histone H3 trimethylated lysine 36 (H3K36me3), a histone modification associated with transcriptional activation and also with the transcriptional factor CREB. After identifying a novel function for PHF21B as a regulator of social memory, and dissecting the underlying neurobiology at the structural and functional levels, we now suggest that PHF21B may be a novel candidate translational therapeutic target for neurobehavioral disorders.

Keywords: Social and Behavioral Deficits, Epigenetics, Neuroepigenetic Editing, Glutamatergic Transmission, AMPA Glutamate Receptors

Disclosure: Nothing to disclose.

P460. The Gene Expression Landscape of the Human Locus Coeruleus Revealed by Single-Nucleus and Spatially-Resolved Transcriptomics

Lukas Weber*, Heena R. Divecha, Matthew N. Tran, Sang Ho Kwon, Abby Spangler, Kelsey D. Montgomery, Madhavi Tippani, Rahul Bharadwaj, Joel E. Kleinman, Stephanie C. Page, Thomas M. Hyde, Leonardo Collado-Torres, Kristen R. Maynard, Keri Martinowich, Stephanie C. Hicks

Johns Hopkins Bloomberg School of Public Health, Baltimore, Maryland, United States

Background: The locus coeruleus (LC) is a small bilateral nucleus located in the dorsal pons of the brainstem, which serves as the brain's primary site for production of the neuromodulator norepinephrine (NE). NE-producing neurons in the LC project widely throughout the central nervous system, playing critical roles in arousal and mood, as well as various components of cognition including attention, learning, and memory. In line with its prominent role in a wide variety of core behavioral and physiological functions, the LC-NE system is highly implicated in multiple neurological and neuropsychiatric disorders. Importantly, LC-NE neurons are highly sensitive to degeneration in both Alzheimer's and Parkinson's disease. However, despite its prominent involvement in a number of critical brain functions, the small size and positioning of the nucleus deep within the brainstem has rendered it relatively intractable to a full cellular, molecular, and physiological characterization.

Methods: Here, we used the 10x Genomics Visium Spatial Gene Expression platform to capture spatially-resolved transcriptomics (SRT) along with histology images to characterize the spatial gene expression landscape of the LC and surrounding region ($N = 5$ neurotypical adult male donors, 23,387 spatial locations, 8 capture areas), and the Chromium Single Cell Gene Expression single-nucleus RNA sequencing (snRNA-seq) platform to capture the transcriptomic profile of individual nuclei from LC-NE neurons and other neuronal and non-neuronal cell populations ($N = 3$ neurotypical adult male donors, 19,927 nuclei). Spatial regions in the SRT data containing LC neurons were annotated based on the histology images and validated by confirming expression of NE neuron marker genes (TH, SLC6A2, DBH), and used for further downstream analyses including differential expression analyses to identify LC-associated genes and for comparison with LC-associated genes previously identified in rodents. LC-NE neurons and other cell populations in the snRNA-seq data were identified by unsupervised clustering and validated by confirming expression of known marker genes, and used for analyses including integration with the SRT measurements and testing for genetic risk associations.

Results: We identified sets of highly associated (false discovery rate, FDR < 10^-3, absolute fold-change, FC > 3) and statistically

significant (FDR < 0.05, absolute FC > 2) genes characterizing the transcriptomic profiles of the LC regions in the human SRT samples, which included both known NE neuron marker genes (TH, SLC6A2, DBH) and other genes. A subset of LC-associated genes previously identified in rodents using alternative technological platforms were found to be highly expressed within the human samples. We identified a population of LC-NE neurons at single-nucleus resolution, and in addition, identified a population of 5-hydroxytryptamine (5-HT, serotonin) neurons that have not previously been characterized at the molecular level in human brain samples. We mapped the spatial distribution of the single-nucleus populations by integrating the SRT and snRNA-seq data, and investigated association of genomic risk for Alzheimer's and Parkinson's disease and attention-deficit hyperactivity disorder across the single-nucleus populations.

Conclusions: We have generated a spatially-resolved and single-nucleus gene expression atlas of the LC and surrounding region in the neurotypical adult human brain, and provide a freely accessible data resource containing spatially-resolved and single-nucleus data in web-accessible and R/Bioconductor formats. This resource containing the full transcriptomic profile of LC-NE neurons and other cell populations within the LC region will contribute to understanding of neurodegenerative and other disorders, which is particularly relevant given that maintaining the neural density of LC-NE neurons prevents cognitive decline during aging.

Keywords: Locus Coeruleus, Norepinephrine, Spatial Transcriptomics, Single-Nucleus RNA Sequencing, Postmortem Human Brain Tissue

Disclosure: Nothing to disclose.

P461. Noradrenergic Modulation of the Medial Prefrontal Cortex

Alexandria Athanason, Marcis Scroger, Noah Paperny, Andrea Liss, Florence Varodayan*

Binghamton University, Binghamton, New York, United States

Background: Norepinephrine (or noradrenaline; NE) is a major brain stress system, that tightly controls cognitive and affective behavior. During periods of arousal, stress and alcohol exposure and withdrawal, NE is released from the locus coeruleus (LC) into the medial prefrontal cortex (mPFC) where it binds to α1, α2 and β adrenergic receptors to modulate cognitive function. In particular, the prelimbic (PL) subregion of the mPFC plays a critical role in sustained attention, decision making, working memory and episodic memory, as well as coping with perceived stressful events. Unfortunately, most of these studies have focused on male subjects. This is particularly surprising as the LC is sexually dimorphic; females have a larger LC than males and their LC does not sensitize to repeated stress or alcohol exposure. Therefore, here we investigated potential sex differences in basal mPFC noradrenergic signaling in male and female mice.

Methods: All experiments were performed male and female C57BL/6J mice (n = 10-18 per sex for each experiment). We used high-performance liquid chromatography to measure basal mPFC NE levels. Gene expression of α1 and β adrenergic receptors were also characterized using real-time polymerase chain reaction. To assess noradrenergic function, we conducted ex vivo patch-clamp electrophysiology recordings in PL layers 2/3 and 5 neurons. Finally, behavioral pharmacology experiments are currently probing the influence of noradrenergic signaling on cognitive and anxiety-like behavior (Barnes maze, elevated plus maze, etc.). Final values were analyzed for independent significance using one-sample t-tests and compared using unpaired t-tests or one-way ANOVA with post hoc analyses as appropriate with Prism software (GraphPad, San Diego, CA).

Results: There were no significant sex differences in basal noradrenergic signaling in the mPFC. However, there were key sex differences in its modulation of PL glutamatergic synapses, with NE increasing glutamate release in layers 2/3 and 5 in males, but only layer 5 in females (all $p < 0.05$). Behavioral studies are ongoing.

Conclusions: Thus, here we have identified key sex differences in noradrenergic influence over mPFC function that may need be taken into consideration during ongoing efforts to develop noradrenergic therapeutics to treat neuropsychiatric diseases, such as alcohol use disorder and posttraumatic stress disorder. Future studies should examine whether a role for NE emerges under conditions of increased cognitive load (e.g., stress or alcohol withdrawal).

Keywords: Norepinephrine, Sex Differences, mPFC

Disclosure: Nothing to disclose.

P462. Selective Effects of Acute and Chronic Stress on Cortical Functional Connectivity

Alexander McGirr*, Donovan Ashby

Hotchkiss Brain Institute, University of Calgary, Calgary, Canada

Background: Depressive, traumatic and other stress-related disorders are associated with large scale brain network functional connectivity changes. Because it is unclear what functional connectivity changes occur during acute stress, it is unclear how these relate to the emergence of persistent large scale network organization.

Methods: Using Thy 1-jRGECO1a transgenic mice, we repeatedly sampled mesoscale cortical calcium activity across dorsal neocortex during quiet wakefulness in several experimental conditions. First, animals were imaged in a homecage condition, after which animals underwent an acute foot-shock stress, a chronic variable stress protocol, an acute on chronic foot-shock stress, and treatment with the prototype rapid anting antidepressant ketamine. We focused on slow cortical activity (0.3-4 Hz) and theta-alpha cortical activity (4-15 Hz) to characterize functional connectivity.

Results: Compared to homecage, acute foot-shock stress induced widespread increases in cortical functional connectivity in the 4-15 Hz temporal band both within functional modules ($t(31)=-4.66$, $p < .001$) and between functional modules ($t(31)=-4.51$, $p < .001$) before returning to control after 24 hours. Chronic stress produced a selective increase in between-module functional connectivity in the 0.3-4 Hz band ($t(14)=-2.81$, $p < .05$), which was reversed after treatment with the rapid antidepressant ketamine. Functional connectivity changes induced by acute stress in the 4-15 Hz band were strongly related to functional connectivity changes after chronic stress and to the pattern of reversal of functional connectivity by subanesthetic ketamine.

Conclusions: Stress induces changes in functional connectivity, and these have specific effects that transcend spatiotemporal aspects of connectivity to link acute, chronic, and treatment effects. The mechanisms by which acute stress connectivity changes become embedded in a lower temporal band over time remain to be determined.

Keywords: Functional Connectivity, Acute Stress, Chronic Stress

Disclosure: Provisional Patent: Patent (Self)

P463. Mesoaccumbal Core Circuit Dynamics During Aversive Processing

Gabriela Lopez*, Louis Van Camp, Talia Lerner

Feinberg School of Medicine, Northwestern University, Chicago, Chicago, Illinois, United States

Background: Behavioral adaptations to aversive stimuli are crucial for survival. The mesoaccumbal core circuit, which includes dopamine neurons in the ventral tegmental area (VTA) projecting to the nucleus accumbens core (cNAc) and their projection targets within cNAc, has been heavily studied for its role in motivation, cue associations, and goal-directed behaviors. Maladaptive changes in this circuit are associated with psychiatric disorders such as chronic pain, addiction, and depression. While the mesoaccumbal core circuit has most often been associated with reward processing, its role in aversive processing and different aversion scenarios remains unclear. Previous studies have largely investigated dopamine release in response to unavoidable aversive stimuli, observing inconsistent results. While understanding direct responses to aversive stimuli is important, it may be more ethologically relevant to study how an animal behaves when they have the chance to avoid an aversive stimulus, which can be accomplished with instrumental tasks. Additionally, many previous studies examined dopamine release and cNAc neuron activity separately, making it difficult to determine how cNAc dopamine influences local cNAc circuit dynamics. I used Pavlovian and instrumental behavioral paradigms in conjunction with multicolor fiber photometry to identify how variations in mesoaccumbal core circuit dynamics may predict differences in aversive processing.

Methods: To investigate aversive processing in the mesoaccumbal core circuit, I used two shock-paired behavioral tasks – active avoidance, an instrumental behavior allowing mice to avoid shocks, and unavoidable shocks. I used fiber photometry to record dopamine release in cNAc by expressing a fluorescent dopamine sensor (pAAV9-CAV-dLight1.3b). To record the activity of dopamine D1 receptor (D1R)-expressing or dopamine D2 receptor (D2R)-expressing striatal projection neurons (SPNs), I also injected a virus carrying a Cre-dependent, fluorescent calcium sensor (pAAV1-CAG-FLEX-NES-jRCaMP1b) into the cNAc of D1-Cre and A2A-Cre mice, respectively. Fiber optic implants were then placed above the injection site and neural activity was recorded as mice freely behaved. During the active avoidance task, which used a 2-compartment chamber, mice were given light and sound cues that preceded administration of a footshock in whatever chamber they resided, and the mouse had 5 s to run to the opposite compartment to avoid the footshock. After 5 s, a 0.4 mA shock began and continued for 25 s or until the mouse moved to the opposite chamber. There were two behavioral scenarios in this task: 1) Avoided Shocks – in which the mouse moved to the opposite chamber before the shock began (<5 s), and 2) Escaped Shock – in which the mouse moved to the opposite chamber after the shock had begun (>5 s). Mice were tested on this task for 7 days, 30 trials per day. Finally, mice were subjected to an unavoidable shock task where a 5 s light and sound cue predicted 5 s of 0.4 mA shock that they could not avoid or escape. Mice were tested on this task for 1 session only consisting of 10 trials.

Results: After 7 days of active avoidance testing, mice learned to avoid ≥80% of shocks. cNAc dopamine release increased during avoidance and escape events, as well as the time of shock cessation. The magnitude and duration of these increases changed as the animals learned the avoidance task. Dopamine release, in contrast, decreased in response to shocks and shock-predicting cues. These dynamics differed in magnitude depending on whether the mouse avoided or escaped the shock. In D1- and D2-SPNs, I observed ramping activity followed by prolonged decreases in response to avoidance and escape events. D1- and D2-SPNs additionally showed increased activity in response to shocks and shock-predicting cues. D1- and D2-SPNs often showed similar activity patterns rather than opposing activity that would be predicted by their differing responses to dopamine. During the unavoidable shock session, dopamine release dynamics and SPN activity differed in magnitude and duration in response to shocks and shock predicting cues compared to active avoidance trials and within unavoidable trials in a given session.

Conclusions: My data illustrate how cNAc dopamine release dynamics as well as D1- and D2-SPN activity differ in response to varying aspects of Pavlovian and instrumental aversive learning. These data also highlight the importance of utilizing more complex and ethologically relevant behaviors to further understand how the mesoaccumbal core circuit is involved in neural and behavioral adaptations towards aversive stimuli. I am currently investigating how this circuit activity is impaired following spared nerve injury as a model of chronic pain, to understand how pain may modulate aversive processing, potentially contributing to opioid addiction and depression. Understanding fundamental processes important for survival, such as aversive learning, will allow us to better understand what may go awry in conditions where aversive processing is dysregulated such as in chronic pain, addiction, and depression.

Keywords: Dopamine, Fiber Photometry, Mesolimbic Circuitry, Active Avoidance, Nucleus Accumbens Core

Disclosure: Nothing to disclose.

P464. Understanding Common Mechanisms in Suicide Across Different Psychiatric Disorders: The Role of CRH in the Central Amygdala

Jonathan Vogelgsang*, Kiely French, Anna Lally, Kerry Ressler, Sabina Berretta

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

Background: Suicide is one of the most serious concerns, but also described as extremely burdensome for patients in psychiatry across all different psychiatric disorders. Emotional dysregulation, such as depression, anxiety, and impulsivity are one of the most important clinical features related to suicidal behavior. However, no biomarker that are clearly associated with suicide attempts have been identified so far and no common brain alterations have been described. As stress and its related factors and hormones are one of the most important risk factors in mental health, we have been focusing on corticotropin releasing hormone (CRH), its corresponding receptor (CRH1R), and the pituitary adenylate cyclase-activating peptide receptor (PAC1R) to identify common molecular patterns in suicide across different psychiatric disorders. Since all these markers have been described to be associated with emotional dysregulation, we postulate that changes in CRH, CRH1R, or PAC1R expression in the amygdala are involved in suicidal behavior.

Methods: Protein lysates form human post-mortem brain tissue from the central amygdala nucleus of 150 brain donors, including controls, patients with major depression (MDD), and post-traumatic stress disorder (PTSD), were analyzed. Out of these 150 donors, 19 donors died by suicide, while 99 died by natural death or accident, and 32 were classified as "undetermined". 52% of the donors were women. Four different races, Caucasian, Asian American, Hispanic, and African American donors were included. Western blotting analysis was carried out using antibodies raised

against CRH, CRH1R, and PAC1R. Demographical information, such as age, sex, post-mortem interval (PMI) as well as clinical information on medication, medical history, or substance abuse were included in the analysis, using multiple linear regression models as well as elastic net analyses.

Results: Using a regression model including diagnosis, suicide, and sex, we saw that donors diagnosed with MDD had lower CRH levels ($p = 0.014$) in the central amygdala. Donors died by suicide had significantly higher CRH levels in comparison to donors died by a natural death or accident ($p = 0.024$). Sex had no effect on CRH levels in the central amygdala. No associations between primary diagnosis, suicide, or sex and PAC1R or CRH1R levels were observed. We also performed elastic net regression analyses to include and control for all available covariates ($n = 58$) and using imputations ($m = 1000$) to take missing values into consideration. We calculated a pseudo-p-value as a surrogate for significant associations between a covariate and protein measurements. Death by suicide ($p < 0.000$) and childhood trauma ($p = 0.019$) were associated with increased CRH levels, while a primary diagnosis of MDD ($p < 0.000$), asthma ($p = 0.019$) and history of military service ($p = 0.069$) lead to decreased CRH levels in the central amygdala. Smoking, BMI, hypothyroidism (all $p < 0.000$), and a primary diagnosis of MDD ($p = 0.008$) were highly associated with decreased PAC1R levels in the central amygdala, while PMI ($p < 0.000$) and antipsychotic medication intake ($p = 0.063$) lead to increased PAC1R levels.

Conclusions: These finding show that CRH might be a key component in suicidal behavior. This study suggests to further examination the role of CRH and its related pathways to be used as biomarkers but also trying to modify these circuits to treat patients with suicidal ideations or intents.

Keywords: Suicide Risk Factors, CRH, MDD, PTSD

Disclosure: Nothing to disclose.

P465. Suicide Risk in a Transdiagnostic Outpatient Population: Interacting Long-Term and Immediate Action Regulation

Nicholas Murphy*, Marijn Lijffijt, Sidra Iqbal, Dania Armaneh, Nithya Ramakrishnan, Brittany O' Brien, Amanda Tamman, Lynnette Averill, Sanjay Mathew, Alan Swann

Baylor College of Medicine, Houston, Texas, United States

Background: Suicide is the leading cause of trauma-related mortality in the US, exceeding vehicular accidents and homicides. While often associated with depression there is mounting evidence that suicide attempts (SA) result from transdiagnostic mechanisms. Delineating potential mechanisms critical challenge because many suicides are first attempts, and suicidal ideation and impulsivity can fluctuate unpredictably before an attempt. Addressing these challenges requires 1) defining a transdiagnostic behavioral model of suicidal behavior; 2) identifying indicators of potential suicidal behavior that do not require a previous attempt. In this investigation we used patients at high risk for a recurrent attempt based on their proximity to a previous medically severe SA (MSSA). Compared to people without history of SA, those with MSSA have elevated risk for all-cause mortality up to ten years post SA. This appears related to increased arousal and behavioral sensitization (BS) predisposing to impulsive behavior and reduced tendency for self-preservation. Accordingly, we investigated self-report behavioral measures of arousal, impulsivity, and potential sensitization, in people who had stabilized from MSSA compared with diagnostic controls. We focused on relationships between long-term (sensitization) and short-term (impulsivity, symptoms, arousal) regulation of action.

Methods: We recruited 28 trans-diagnostic outpatients at high risk for SA, defined as MSSA in the past year (high-risk, HR), and 23 age and symptom-matched outpatients without a history of MSSA (low-risk, LR). Psychiatric diagnoses were based on the MINI; severity of affective, anxious, and psychotic symptoms was measured using the Schedule for Affective Disorders and Schizophrenia – Change (SADS-C) questionnaire, designed to measure these symptoms concurrently. Fifty patients reported symptoms consistent with a major depressive episode, anxiety symptoms were present in 45%, bipolar 35%, PTSD 41%, psychotic disorder 13%, BPD 4%, OCD 3.9%. All patients completed clinical and behavioral evaluations; 21 HR and 11 LR patients completed combined clinical, behavioral, and neurophysiological tasks. Clinical and behavioral analysis focused on 1) clinical symptoms, 2) aggressive behavior, 3) Impulsivity, 4) sensitization behaviors. Risk group comparisons used independent samples t-tests and Cohen's D effect sizes. Correlations with Beck's SSI-W were determined across all subjects to assess relationships to risk factors and suicidal ideation. Only HR subjects had suicide attempt histories, but we addressed the frequency and specificity of SI by allowing it in either group.

Results: Groups were matched on symptom severity (SADS-C) and subjective self-rate symptoms (Internal State Scale (ISS)). Behavioral analysis revealed increased lifetime history of aggressive behaviors ($p = 0.004$,$d = 0.87$) and provoked aggressive behavior (PSAP, $p = 0.02$,$d = 1.29$) but not current aggressive behavior (MOAS). Trait (Barratt Impulsivity Scale, UPPS) and state (Immediate Memory Task (IMT)) did not differ significantly between groups across sub-domains. However, Cohen's d effect sizes indicated trends for IMT correct detections ($p = 0.059$,$d = 0.5$; state impulsivity). History of abuse was significantly greater for physical abuse in HR patients ($p = 0.04$,$d = 0.6$) with a trend for sexual abuse ($p = 0.055$,$d = 0.53$). CAPS-5 scores did not differ between groups across domains. Lifetime cumulative adversity was greater in HR ($p = 0.04$, $d = 0.67$). HR patients also demonstrated stronger history of alcohol use disorder (Addiction Severity Index) than LR patients ($p = 0.01$, $d = 0.69$).

Beck's worst suicidal ideation (SSI-W) was significantly correlated with a broad pattern of clinical and behavioral scores. SSI-W differed significantly between HR and LR but ranged widely with substantial overlap between groups. Symptoms (SADS-C total $r = 0.56$, Depression $r = 0.54$, Anxiety $r = 0.48$), ISS (Depression $r = 0.45$, Perceived Threat $r = 0.43$), Aggression (PSAP $r = 0.48$), BIS (Attentional $r = 0.55$, Total $r = 0.47$), UPPS (Lack of Perseverance $r = 0.43$), IMT (Commission Error Reaction Time $r = 0.48$), CTQ (Emotional Abuse $r = 0.46$), and CAPS-5 (Arousal $r = 0.41$) correlated with SSI-W.

Conclusions: We investigated behavioral markers of arousal and BS as indices of suicide risk using MSSA patients at HR for recurrent SA. Our findings suggest that lifetime history of aggression, PSAP provoked aggressive responses, childhood trauma, and alcohol use are potential behavioral markers of high suicide risk. In general, measures related to sensitization were more likely to be related to HR, while impulsivity related more to SSI-W scores. This suggests that a sensitization-like process may predispose to HR; overt suicidal behavior may require combined long-term (HR; sensitization) and immediate (SI, impulsivity) action dysregulation.

Keywords: Suicide Risk Factors, Suicide Attempt, Suicide Mechanisms

Disclosure: Nothing to disclose.

P466. Meaningfully Addressing Suicide Requires a Sustained and Integrated Effort Comparable to the War on Cancer

Sheldon Preskorn*

University of Kansas, Wichita, Kansas, United States

Background: Over 20 years, a year over year increase in the number of Americans who die from suicide (currently that is one out of 62) has occurred despite multiple US Surgeon Generals highlighting this problem and multiple attempts to address using public health approaches. However, these have been piecemeal rather that a sustained, integrated approach.

The problems with such approaches is a biopsychosocial phenomenon with different components including different drivers and protective factors in different individuals who die (or do not) by suicide. This topic is of direct relevance to the many, if not most, members of American College of Neuropsychopharmacology (ACNP) and the ACNP which can be a vital voice in advocating for a comprehensive approach to this problem.

Methods: The methods included: personal knowledge, focused literature reviews and conversation with individuals at organization such as the NIMH section on suicide. The material on War on Cancer came from personal experience augmented by focused literature review on the history of its establishment and evolution into a cohesive organization with central tissue and treatment databases, Cancer Centers at academic medical centers in most states, and community based cancer sites with bi-directional and collaborative interactions including collection of tissue for biological examination, data collection from multiple clinical protocols run, analyzed and then refined by new protocol in an iterative process.

Additional literature reviews were focused on risk factors for suicide and the medicolegal death investigation system in the United States.

Results: Given space constraints, an exhaustive review of the results is not possible but can be more fully presented in the poster. The following is a top-level summary:

The War on Cancer has provided a blueprint for the development of a similar initiative to meaningfully address suicide and has established the amazing ability of such a structure to make amazing advances in the understanding and effective treatment of multiple different cancers. A criticism could be that cancer permits collection and analyze of pathological tissue but that same is true for suicide using blood and even brain tissue, if necessary.

Of importance, there are two distinct populations who complete suicide or make multiple suicide attempts. Most individuals who die by suicide do so on their 1st attempt and approximately 95% of those who die by suicide do so by their 3rd attempt. There are three psychiatric diagnoses that account for the majority of suicide completers. Three different psychiatric diagnoses that are heavily represented in the population who is at high risk for multiple suicide attempts. In the interest of space, this data will be present in the poster rathe than here.

Suffice to say: the genes in these two populations may be quite different because of the different underlying diagnoses but the bottom-line is that piecemeal studies which use multiple attempters for suicide completers can cause confusion rather than clarity. That can be addressed in the database that will be established under this initiative and will be great importance to dissecting the genetic based risk of suicide.

Briefly, the biopsychosocial understanding of suicide includes the following by way of example but not limited to:

Biological Data: 1) The individual's DNA and epigenetic changes; 2) Messenger RNA reflecting the state of the individual at the time of death by suicide; 4) Markers of inflammatory processes and stress response systemically and in the brain particularly the hypothalamic-pituitary axis; 6) Analysis of whole brain samples (when possible) and of brain sections in high value regions when examination of whole brain samples is not possible. These samples will be analyzed to determine whether the brain circuits in these regions have been damaged by concussive damage versus chemical damage on a smaller scale caused by sound waves or chemical exposure); 7) Measures of cognitive and emotional processing when available; 8) Surrogate measures of impulsivity (e.g., CSF levels of serotonin and/or monoamine oxidase-A (MAO-A) activity; 9) Age and gender; 11) Co-morbid medical illnesses and substance abuse and/or dependence.

Psychological Data: 1) The individual's religious and/or philosophical beliefs when they can be ascertained with sufficient certainty. For example, some religions forbid suicide, whereas other religions may praise martyrdom suicide); 2) The individual's sense of locus of control (i.e., primarily internally or externally driven); 3) Adverse childhood experiences;4) Psychiatric diagnosis(es); 5) Number of previous suicide attempts.

Social Data:

How the individual functioned within the nuclear family and subsequent social networks including how the individual functioned within society as assessed by educational attainment, marriage, and ability to sustain functioning and advance within her/his chosen profession.

All the above information and more as results indicate will be included in the database and statistical models constructed to determine the highest risk combination(s) realizing there may be multiple pathways to suicide completion requiring different predictive profiles and treatment interventions.

Conclusions: This abstract lays out a meaningful pathway forward in understanding and ability to meaningful treat individuals at high risk of death due to suicide. The ACNP organization and its members have a vital role in making this idea reality.

Keywords: Suicide, Suicide Risk Factors, Database

Disclosure: Nothing to disclose.

P467. Protective and Risk Factors Among American Indian Peoples Modulate the Relationship Between Mental Health Problems and Multi-Modal Neural Markers During an Inhibitory Control Task

Evan White*, Ricardo Wilhelm, Nicole Baughman, Breanna McNaughton, Rayus Kuplicki, Jennifer L. Stewart, Martin Paulus, Robin Aupperle

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

Background: American Indian (AI) communities suffer from increased burden of mental health risk factors as a result of historical and ongoing sociopolitical and environmental challenges (e.g., historical loss, discrimination, disenfranchisement). Although this has resulted in disproportionate rates of mental health conditions in AI populations, there is also evidence of high levels of positive mental health in AIs. A growing body of literature shows traditional cultural factors (e.g., spirituality) can be protective against poor mental or physical health outcomes. This is consistent with research conducted in the broader population that shows religiosity is associated with decreased prevalence of deaths of despair. Moreover, our recent work has demonstrated that when accounting for disproportionate risk factors (i.e., sociodemographic factors, trauma exposure), a heterogenous AI group had equivalent rates of substance use disorder (SUD) and lower rates of suicidal thoughts and behaviors (STB) relative to matched non-Hispanic white peers. Furthermore, our work demonstrated that among the AI sample, brain activation during an inhibitory control task differentiated both individuals with and without STBs from those without STB and individuals with SUD and those without SUD. This investigation used a systems neuroscience framework to examine the possible cognitive and neural processes that mediate the protective effects of cultural factors among AI peoples. Specifically, the current study investigated the potential modulation of the relationship between STB and multi-modal neural markers of inhibitory control by culturally relevant protective (i.e., spirituality) and risk (i.e.,

historical loss thinking) factors in a sample of AI individuals. It was hypothesized that 1) AI spirituality would diminish the influence of STB on inhibitory control and 2) that historical loss thinking would exacerbate the same relationship.

Methods: Participants were drawn from an initial pool of 60 individuals who self-identified as AI to any degree in the Tulsa-1000 (T1000) study and agreed to participate in a follow-up session to assess culturally-relevant protective and risk factors. The T1000 study was a naturalistic longitudinal study that included assessment of mental health and substance use conditions using the Mini International Neuropsychiatric Interview as well as multimodal neural markers of inhibitory control using concurrent functional Magnetic Resonance Imaging (fMRI) and electroencephalography (EEG) recording during a stop signal task (SST). Participants (n = 37 total; no STB = 17) were included in analysis if they had completed the MINI interview and had quality fMRI and EEG data for the SST. Neural correlates of inhibitory control included the contrast of percent signal change during hard versus easy stop signal trials in the dorsolateral prefrontal cortex (dlPFC) and the P300 event-related potential. The Historical Loss Scale (HLS) was used to measure self-reported historical loss thinking and the Native American Spirituality Scale (NASS) was used to quantify self-reported spirituality.

Results: Linear mixed effect (LME) models were used to examine the interaction of STB and NASS on dlPFC PSC and P300 amplitudes separately. LMEs were also used to examine the interaction of STB and HLS on both neural outcomes. For spirituality the hypothesis was supported for the dlPFC, wherein results indicated significant main effects of both NASS ($F = 5.144$, $p = .026$) and STB ($F = 12.76$, $p < .001$) on dlPFC; this was qualified by a significant STB*NASS interaction on dlPFC ($F = 13.54$, $p < .001$). Results also supported the hypothesis for the P300 demonstrating a significant STB*NASS interaction ($F = 4.41$, $p = .03$). For historical loss thinking results also supported the hypotheses. dlPFC results indicated a significant main effect of STB ($F = 10.65$, $p = .002$) qualified by a significant STB*HLS interaction ($F = 9.47$ $p = .003$). Finally, P300 results demonstrated a marginal main effect of HLS ($F = 3.23$ $p = .07$) qualified by a STB*HLS interaction ($F = 5.56$, $p = .02$).

Conclusions: These preliminary results indicate that in a heterogenous sample of AI adults (1) spirituality may attenuate the relationship between STB and inhibitory control disruption and (2) this relationship may be exacerbated by historical loss thinking. These findings extend our previous work demonstrating that inhibitory control may be an important mechanism for understanding mental health risk and protective factors for AI populations. Furthermore, results bolster a growing body of research indicating that traditional cultural engagement may serve as a protective factor among AI communities. The current analysis is limited to a cross-sectional empirical design, a relatively small sample, and does not consider comorbidities (physical or mental health) or other mental health or substance use concerns. This analysis does suggest that inhibitory control may be an important neural mechanism of risk and protective factors specific to AI mental health. Future work is needed to delineate the prospective and potential causal directionality of these effects.

Keywords: Suicide, Native Americans, Protective Factor, fMRI, Event Related Potentials

Disclosure: Nothing to disclose.

P468. Plasticity in the Gustatory Insular Cortex Underlying Perceptual Learning in Behaving Mice

Joshua Kogan*, Ayesha Vermani, Memming Park, Alfredo Fontanini

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

Background: To adapt and survive, animals must learn to discriminate overlapping stimuli predicting different outcomes. This phenomenon, known as perceptual learning (PL), has been well described in the visual, auditory, somatosensory, and olfactory systems. Specifically, improved performance on PL tasks is associated with plasticity of both sensory representations and decision related neural signals. However, few studies have addressed PL in the gustatory system. The primary sensory region for taste, known as the gustatory insular cortex (GC), encodes both sensory and decision-related information, but how these representations might change with PL remains unknown. Additionally, as part of the larger insular cortex, GC has been implicated in many processes including homeostatic regulation of food consumption and eating disorders. Thus, the present study addresses important gaps in the literature by improving our understanding of PL in the gustatory system and enhancing knowledge of neural coding in a region involved in many important gustatory-related functions and pathologies.

Methods: The experiments are based on a novel gustatory PL paradigm, which relies on a two alternative forced choice task. Mice first learn a sucrose (100 mM) vs NaCl (100 mM) discrimination, in which sucrose presentation at a central spout is associated with reward at one lateral spout and NaCl with reward at the other. They are then trained to discriminate between increasingly similar pairs of mixtures: 75/25 vs 25/75, 65/35 vs 35/65 and 60/40 vs 40/60 (%sucrose/%NaCl). When animals make an error, they receive a timeout and no reward. Before and after learning, mice are tested on a battery of mixtures to establish psychometric curves. To assess GC plasticity, ensemble activity is monitored with two photon calcium imaging. Single neuron and population-level analysis are used to identify changes in both sensory and decision-related neural responses that occur with PL.

Results: After learning, performance increased for all mixture pairs, indicating an improvement in discrimination (n = 6 mice, two-way ANOVA, effect of learning $p < 0.001$). Previous work from our lab using a similar behavioral task has shown that GC neurons encode sensory and decision-related information in separate temporal epochs. Specifically, sensory information is encoded in the epoch directly after taste sampling, and decision-related information is encoded in the delay epoch directly before the choice. Analysis of all recorded neurons (Pre-learning: 2337 neurons; Post-learning: 2658 neurons) showed an increase in the proportion of delay responsive neurons (Pre - 19%, Post – 24% X2 test, $p < 0.001$), but no significant change in the sampling responsive population. In the delay but not sampling period, responsive neurons became more selective to the upcoming choice after learning, and the magnitude of the activity difference between trial types in the delay was larger. These findings suggest that gustatory PL may be mostly associated with improvements in decision-coding. To test is this is true at the population level, we employed logistic regression to decode the choice from the population of recorded neurons, and similarly found an improvement in decoding performance after learning.

Conclusions: These results demonstrate that improved behavioral performance on a gustatory PL task is associated with enhanced decision-related coding in GC. We confirm this result using single neuron and population level analyses of neural activity. Overall, these experiments further elucidate the GC plasticity associated with PL and provide novel information about neural processing in gustatory insular cortical circuits.

Keywords: Insular Cortex, Taste, Two-Photon Calcium Imaging, Mice, Perceptual Decision-Making

Disclosure: Nothing to disclose.

P469. NMRA-511, a Novel Vasopressin 1a (V1a) Receptor Antagonist Reduces Threat Behaviors in Marmosets and Alters EEG Power Spectra Similarly in Marmosets and Humans

Tanya Wallace*, Filomene Morrison, Philip Gerrard, Larry Ereshefsky, Michael Ballard, Joel Posener, Katherine Tracy, Jane Tiller

Neumora Therapeutics, Brisbane, California, United States

Background: The arginine-vasopressin (AVP) neuropeptide system is involved in regulating complex social behaviors and emotional states across species. Within the brain, the vasopressin 1a (V1a) receptor is the predominant receptor subtype and has been implicated in mediating anxiety and threat processing. Given such emotional processing impairments are present in many brain disorders, modulation of the AVP system through the V1a receptor provides a viable approach for the treatment of these disorders. NMRA-511 has been extensively characterized in in vitro and in vivo systems and is a potent small molecule antagonist at the V1a receptor with selectivity over closely related V1b, V2 and oxytocin receptors (OTR) as well as to broad panels of off-target receptors, ion channels, and transporters, and is brain penetrant and orally bioavailable across species. The current studies were designed to evaluate the potential of NMRA-511 to modulate anxiety-related behaviors in marmoset monkeys (Callithrix jacchus) to a human intruder using the Human Threat Test (HTT). In the HTT, marmosets recognize the presence of a human observer as a threat and will present characteristic behavioral postures accompanied biochemically with elevated plasma cortisol concentrations. Additionally, using a translational pharmacoelectroencephalography (phEEG) approach, the effects of NMRA-511 on relative power across spectral frequency bands were evaluated in both marmosets under physiological and threat conditions and extended into a human healthy control population under physiological (resting state) conditions as an exploratory measure in a phase 1 single/multiple ascending dose trial.

Methods: Adult marmosets ($n = 8$) treated with NMRA-511 (0-30 mg/kg, PO) or the positive control chlordiazepoxide (CDP; 2 mg/kg, SC) were used for the HTT. A separate cohort of marmosets ($n = 6$) was implanted with EEG electrodes above the frontal and parietal cortices and electromyography electrodes were fixed to the neck muscles. A single dose of NMRA-511 (10 mg/kg, po) or vehicle was evaluated in the marmoset phEEG study. Animals were treated according to a random blind crossover design, with a washout period of at least three days between treatments. Data were analyzed using RM-ANOVA ($p < 0.05$). In the healthy control study, NMRA-511 (5, 10, 15 mg dose) or placebo was administered once orally ($n = 6$:2; NMRA-511:placebo) and EEG from a 19-channel, 10/20 system was recorded for 6.5 hours. Tmax-centered mixed models for repeated measures served as the primary analysis ($p < 0.1$).

Results: In the marmoset, orally administered NMRA-511 (10 and 30 mg/kg) significantly reduced anxiety-related behaviors as measured by a decrease in the number of threat-elicited postures observed in the HTT without affecting locomotor activity or causing sedation. Results from the phEEG study in the marmoset revealed significant changes specifically in the theta (4-8 Hz) and alpha (8-12 Hz) bands under physiological conditions that were further exacerbated under the threat condition. Non-significant trends were also observed in the beta (12-30 Hz) band. The phEEG studies were extended to a human healthy control population and in line with hypotheses, Tmax-centered analyses indicated significant increases from baseline in theta (eyes open [EO] and eyes closed [EC] conditions) and alpha (EO) in the midline frontal region of the 15 mg cohort ($n = 6$) on Day 1 compared to placebo ($n = 11$). Non-significant trends of moderate to large effect sizes suggesting an increase in beta were also noted at the 15 mg dose. At the 10 mg dose ($n = 24$), a significant increase in beta was observed (EO and EC conditions) with a moderate-sized non-significant increase in theta (EO) proximal to Tmax. The ascending single and multiple doses of NMRA-511 were safe and well tolerated by the healthy participants in this study.

Conclusions: Oral administration of the V1a antagonist NMRA-511 produced anxiolytic-like activity without sedation in the marmoset threat test. The same dose of NMRA-511 (10 mg/kg) that reduced anxiety in the marmoset demonstrated increased relative power in the alpha and theta spectral bands with trends in the beta band in this species. Furthermore, using phEEG as a translatable biomarker, NMRA-511 was evaluated in a healthy control population and demonstrated similar changes in alpha and theta power and trends in the beta band as were observed in the marmoset. These studies support the utility of phEEG as a translational tool bridging preclinical and human populations; and the continued investigation of NMRA-511 in the treatment of brain disorders.

Keywords: Vasopressin 1a Receptor Antagonist, EEG Biomarkers, Anxiety, Translational Neuroscience

Disclosure: Neumora Therapeutics: Employee (Self), Neumora Therapeutics: Stock / Equity (Self)

P470. Compassionate Imagery Practices Priming for Psilocybin Ceremonies: A Large Scale Field Study

Carla Pallavicini*, Lorena Llobenes, Federico Cavanna, Natali Gumiy, Maria Costa, Nicolas Bruno, Stephanie Muller, Laura Alethia De La Fuente, Paul Gilbert, Enzo Tagliazuchhi

Computational Cognitive Neuroscience Lab, Universidad de Buenos Aires, Caba, Argentina

Background: Classical psychedelics like psilocybin, N-N dimethyltryptamine (DMT), lysergic acid diethylamide (LSD) and mescaline are known to induce profound, transient alterations in consciousness. Psychedelic research has shed light on the neurophysiology of these altered states of consciousness, as well as on their potential for the treatment of several mental health disorders, and for the promotion of a sustained state of well-being. On the other hand, the increased focus on meditation in cognitive neuroscience resulted in a cross-cultural classification of standard meditation styles, which was validated by functional and structural neuroanatomical data. Compassionate imagery is a form of guided meditation focusing on building a compassionate image of oneself and others to work with and develop. This practice has been linked to psychological benefits and general increases in wellbeing markers; one of our main objectives was to understand whether these benefits can be facilitated by the sense of contentedness catalyzed by psilocybin mushrooms.

Methods: After thorough screening, specially focused on participant safety, 105 healthy volunteers were measured in this experiment. We designed an ambitious setup in order to cover as many dimensions of the experience and its effects as possible. Participants were divided into 4 groups: low vs. high dose of psilocybin and compassion vs. control meditation. Physiological and activity data was acquired 1 week before and after the day of the ceremony using Fitbits. Also, fMRI scans, blood and saliva extractions were performed 1-3 of days before and after as well. Participants responded an extensive selection of scales and questionnaires, respecting personality traits, well-being, anxiety, among others one week before the ceremony. These measures are repeated a week, 1, 3 and 6 months after the ceremony. Starting the day of the ceremony we perform individual interviews, psychometric scales responses and EEG registers. After this,

volunteers received the meditative practice corresponding to their group, followed by an individual interview and proceed to their ceremony with a facilitator. We did not interact with participants during their experience. After the ceremony, participants responded a set of questionnaires designed to assess their experience, and we finished by taking EEG registers, saliva samples, individual interviews and group sharing sessions. Follow-up measures consisted of the scales and questionnaires mentioned above and a blood and saliva extraction performed 2 months after the ceremony.

Results: Preliminary results, show that participants in the compassion groups scored significantly higher on interoceptive constructs such as listening to body and confidence as reported by the MAIA scale ($p < 0,05$), and also reported to have a subjective experience with a larger influence of elementary imagery ($p < 0.05$). Only for the compassion group, there was a difference in the subjective construct of changed meaning between high and low dose. Reported incidents of anxiety and difficult experiences only occurred in the high dose groups and were of low incidence (<5% of participants). Interviews presented differences in verbosity amongst high and low dose groups and EEG data also tend towards differences amongst these groups.

Follow-up measures are still in progress, but we expect the differences to accentuate over time and to find new indications on how this combination of practices reflects on subjective well-being markers.

Conclusions: We were able to perform a very large amount of measures on a large amount of participants in a combined psychedelic and meditation study. This experiment is ground breaking for this kind of research in Latin America. We found that these ceremonies were safe for participants and the combination with meditative practices contributed to the low amount of difficult experiences observed. The ceremonial use of Psilocybin can have a positive effect in self-reported measures of psychological well-being, mood, prosocial behavior, emotional regulation and cognitive capacities. Psychological priming with a compassionate imagery practices could be a useful tool to increment the positive impact of the subjective experience, our results show a trend in that direction.

Keywords: Psilocybin, Meditation, Psychological Wellbeing, EEG, Functional MRI (fMRI)

Disclosure: Nothing to disclose.

P471. Kappa Opioid Receptor Availability in Borderline Personality Disorder and Relationship to Suicidal Behavior: Preliminary Results From a [11 C]EKAP PET Study

Margaret Davis*, Olivia Wilson, Ashley Wagner, Nabeel Nabulsi, David Matuskey

Yale University School of Medicine, Waterbury, Connecticut, United States

Background: Borderline personality disorder (BPD) is a serious psychiatric condition associated with high morbidity and mortality and elevated risk for suicide. Despite this, treatments capable of affecting overall symptom severity and reducing suicide risk in BPD are limited. Promisingly, recent evidence has implicated the kappa opioid receptor (KOR) in both BPD and suicidal behavior. Specifically, postmortem studies have shown an association between KOR and death by suicide, and studies in both animals and humans have shown that KOR antagonists can produce antidepressant, anxiolytic, and even anti-suicidal effects. To date, however, the relationships between KOR, BPD, and suicidal behavior have not been directly investigated. The aim of this study was to examine the relationship between KOR availability, BPD, and suicidal behavior in vivo using [11 C]EKAP PET.

Methods: Individuals with BPD ($N = 8$, 4 with a history of suicide attempt (SA), 4 without (NSA)), and healthy adults (HA; $N = 8$) matched for age, sex (6 women and 2 men in each group), and smoking history were recruited from the community. All participants completed 1 MRI scan and 1 PET scan with [11 C] EKAP, as well as a battery of psychiatric and cognitive assessments. The radiotracer was injected as bolus and subjects were scanned for 120 minutes. Volume of distribution (VT: the ratio of activity in tissue relative to that in blood, corrected for metabolites) was computed using arterial input function. Primary analyses focused on a circuit of 6 frontal and limbic brain regions relevant to the pathophysiology of BPD and suicidal behavior: dorsolateral prefrontal cortex (dlPFC); orbitofrontal cortex (OFC); ventromedial prefrontal cortex (vmPFC); anterior cingulate cortex (ACC); amygdala; hippocampus.

Results: ANOVA analyses revealed significant group differences in KOR availability (VT; $p = .009$, $d = 1.85$, 27% lower). Notably, differences were most pronounced among BPD individuals reporting a history of SA. Specifically, BPD individuals reporting a history of SA had significantly lower KOR availability relative to both BPD-NSA ($p < .001$; $d = 1.93$; 24% difference), and HA ($p < .001$; $d = 2.24$; 35% difference). Further, frontolimbic KOR availability in BPD was negatively correlated with endophenotypic correlates of suicide risk in BPD, including difficulties in emotion regulation ($r = -.83$), impulsivity ($r = -.80$), and depressed mood ($r = -.82$).

Conclusions: Preliminary results support a relationship between KOR, BPD, and suicidal behavior in BPD. Individuals with BPD had lower frontolimbic KOR availability in relative matched HA. Additionally, individuals with BPD who had a history of SA had lower frontolimbic KOR availability relative to both BPD-NSA and HA, suggesting a potential role for KOR as a trait marker of suicide risk in BPD. Of clinical significance, KOR availability was negatively correlated with endophenotypic correlates of suicidal behavior in BPD including difficulties in emotion regulation, impulsivity, and depressed mood, variables which have been shown to predict both future SA and suicide mortality. Taken together, these results provide the first direct evidence for a possible role for KOR in the pathophysiology of BPD and suicidal behavior. Further evaluation of KOR targets for the treatment of suicidal behavior in BPD is warranted.

Keywords: Borderline Personality Disorder, Suicide Attempt, Kappa Opioid Receptor, PET Imaging Study

Disclosure: Nothing to disclose.

P472. MicroRNA-124 and Suicide Ideation in Borderline Personality Disorder: Possible Protective Effect of the Globus Pallidus

Macarena Aloi, Guillermo Poblete, John Oldham, Michelle Patriquin, David Nielsen, Thomas Kosten, Ramiro Salas*

Baylor College of Medicine, Houston, Texas, United States

Background: Borderline personality disorder (BPD) patients suffer from unstable affect and interpersonal relationships, and strong suicide risk. MicroRNA-124-3p (miR-124) was identified in a Genome Wide Association Study as likely associated with BPD.

Methods: Using TargetScan and the Allen Atlas datasets we identified the brain regions where co-expression of genes likely modulated by miR-124 are highest and compared morphometry in those brain regions in BPD inpatients ($N = 111$) vs. controls matched for psychiatric comorbidities ($N = 111$) in the Menninger Clinic in Houston, Texas. Next, we correlated personality measures, suicidal ideation intensity, and recovery from suicidal ideation with volumetrics.

Results: Gene targets of miR-124 were significantly co-expressed in the left Globus Pallidus (GP), which was smaller

in BPD than psychiatric controls. In BPD but not controls, smaller GP volume was negatively correlated with agreeableness and with recovery from suicidal ideation post-treatment. In BPD but not controls, the GP was smaller in older than younger patients.

Conclusions: In BPD, GP volume may be reduced through miR-124 regulation and suppression of its target genes. Importantly, we identified that reduction of the GP could serve as a potential biomarker for recovery from suicidal ideation in BPD.

Keywords: Borderline Personality Disorder, Human Epigenetics/microRNA, Globus Pallidus, Morphometry, Human Brain Imaging

Disclosure: Nothing to disclose.

P473. Insular-Prefrontal Circuit Driving Compassionate Social Behavior

Songjun Li*, Pauline Gabrieli, Moeko Suzuki, Omer Zeliger, Jack Demaree, Renee Cauchon, Nicole Occidental, Ziv Williams

Massachusetts General Hospital, Boston, Massachusetts, United States

Background: Compassionate behavior, or the ability to help others in need, is a cornerstone of prosocial interaction in mammals and humans. To benefit others, it is necessary for individuals not only to perceive the internal states or emotions of others but also to take appropriate actions. For example, one must identify the distress of another and perform specific actions that will relieve their discomfort. Yet, how mammalian neurons precisely link social-specific information with such complex adaptive behavior has been a major challenge to understand.

Methods: We recorded single-neuronal activity in the dorsal anterior cingulate cortex (dACC) and the anterior insula (AI), two brain regions involved in prosocial behaviors, while mice directly controlled the experience of a nearby conspecific partner in real-time. This novel task allowed the animal's own actions to be dissociated from the other's identity and experiences. Additionally, we utilized chemogenetic tools to either inhibit or stimulate the dACC-AI circuit to modulate these behaviors.

Results: We found that wild-type male mice consistently chose to reduce aversive experiences of familiar but not unfamiliar partners, actions that were not observed when visual and olfactory cues were blocked. AI cells preferentially encoded task-relevant information including the social identity of partners and their specific experience, while dACC cells preferentially encoded information about the act of helping their partners. Whereas information about the experience of others could be predominantly decoded from AI activity, information about the animal's prosocial actions could be predominantly decoded from dACC activity. Chemogenetic excitation of AI-to-dACC projectors but not dACC-to-AI projections increased compassionate behavior, while inhibition of both dACC to AI as well as AI to dACC projectors decreased compassionate behavior with familiar partners.

Conclusions: Our data suggest that the mice displayed consistent compassionate behavior towards familiar partners, where there exists a partitioning of information within the insular-prefrontal circuit. Specifically, the AI appears to transmit social-specific information to the dACC while the dACC controlled compassionate decisions based on information from the AI. Taken together, these findings identify a putative insular-prefrontal circuit for driving compassionate behavior and a mechanism that could allow insular neurons to instruct social-specific actions through prefrontal control.

Keywords: Social and Behavioral Deficits, Empathy, Decision Making, Mice, Single-Unit Electrophysiology In Vivo

Disclosure: Nothing to disclose.

P474. Activity-Based Mapping of Altered Neural Network in a Mouse Model of Social Deficits

Ruixiang Wang*, Zeru Peterson, Nagalakshmi Balasubramanian, Kanza Khan, Gabrielle Bierlein-De La Rosa, Thomas Nickl-Jockschat, Catherine Marcinkiewcz

The University of Iowa, Iowa City, Iowa, United States

Background: Deficits in social functioning are a common feature of various psychological disorders, such as schizophrenia and autism spectrum disorders. However, the neural mechanisms underlying such deficits are still unclear due to the dynamic, complex nature of social behavior. Combining the methods of immunolabeling-enabled 3D imaging (iDISCO), chemogenetics, and functional MRI (fMRI), the present study aimed to map the altered neural network associated with social deficits in a mouse model.

Methods: Male C57BL/6 J mice were treated with ketamine (3 mg/kg, i.p.) or vehicle (0.9% saline) for 10 days. 24 hours after the last treatment, they underwent a three-chamber sociability test or an equivalent test with two empty enclosures. Their whole brains were then immunostained with a c-fos antibody, cleared with dibenzyl ether, and imaged under a light sheet microscope. Because a marked reduction in c-fos activation was observed in the lateral septum (LS) after chronic ketamine exposure, we tested whether reduced activity in the LS was necessary and sufficient for social deficits by chemogenetically manipulating LS neurons. Furthermore, to examine the functional connectivity originating from the LS that regulates social behavior, we performed chemogenetic fMRI mapping of the LS neural network.

Results: Chronic ketamine exposure led to a significant reduction in social interaction ($p < 0.001$), which was associated with blunted c-fos induction in the LS ($p < 0.05$), shown by iDISCO imaging. As such, we transduced naïve C57BL/6 J mice with a Gq-coupled DREADD (hM3Dq-mCherry) in the LS and then treated them with chronic ketamine. 24 hours after the last ketamine administration, it was shown that activation of the Gq-DREADD in the LS mediated by clozapine-N-oxide (CNO; 3 mg/kg, i.p.) rescued the social withdrawal induced by chronic ketamine exposure ($p < 0.001$).

To examine if inhibition of previously activated LS neurons during social interaction was sufficient to induce social deficits, we microinjected a Cre-inducible Gi-coupled DREADD (DIO-hM4Di-mCherry) in the LS of fos-iCreER mice. After undergoing a social-interaction session, these mice were administered with 4-OH-tamoxifen (10 mg/kg, i.p.) to induce Cre-mediated recombination and DREADD expression in active neuronal populations. Four weeks later, we conducted the sociability test 45 minutes after a CNO injection. It was shown that inhibition of LS neurons by the Gi-DREADD led to reduced social interaction ($p < 0.001$).

In eGFP-L10A mice microinjected with an hSyn-Cre virus in the LS, we observed strong eGFP expression in the hippocampus, which, accordingly, constitutes a major projection area of the LS neurons. Therefore, we studied this circuitry with fMRI. Fos-iCreER mice were transduced with a Cre-inducible Gq-coupled DREADD (DIO-hM3Dq-mCherry) in the LS. They were then exposed to a social-interaction session and injected with 4-OH-tamoxifen. After four weeks, an fMRI scan was performed following CNO administration. A trend of decreased functional connectivity between the LS and the hippocampal formation was observed ($p < 0.09$).

Conclusions: As a valid mouse model for studying social deficits, chronic ketamine exposure results in hypoactivation of the LS neurons, which is necessary for inducing social withdrawal. Moreover, inhibiting activated LS neurons in social interaction is sufficient for the expression of social deficits. In

addition, previously, it has been determined that the majority of LS neurons are GABAergic and thus inhibitory in nature. As such, activating LS neurons leads to a trend reduction in connectivity with the hippocampus, a major projection area of LS neurons.

Keywords: Social Deficits, Subchronic Ketamine Model, Lateral Septum, iDISCO Mapping, fMRI

Disclosure: Nothing to disclose.

P475. Investigating Protein Glycosylation in Schizophrenia: From Genetics to Treatment Targets

Robert Mealer*, Richard Cummings, Jordan Smoller

Harvard Medical School, Massachusetts General Hospital, Boston, Massachusetts, United States

Background: Schizophrenia is a severe and highly heritable neuropsychiatric disorder. Recent studies have shed light on the complex genetic architecture of the condition, identifying over 270 common variants and 10 rare variants which confer significant changes in risk. However, treatment development remains a considerable challenge given the polygenic pattern of inheritance and clinical heterogeneity of schizophrenia. One pathway with convergent evidence for a role in schizophrenia pathogenesis is altered glycosylation, the enzymatic attachment of carbohydrates to proteins and lipids to regulate their function.

Methods: To generate a broad framework for understanding the relationship between schizophrenia and glycosylation, we will review results from several recent, well powered, and peer reviewed studies. The primary objectives are to: 1) Explore publicly available schizophrenia GWAS and exome sequencing data for enzymes of glycosylation and proteins regulated by glycosylation (glycoproteins); 2) Describe post-mortem studies of schizophrenia investigating of brain glycoproteins; 3) Summarize animal models of schizophrenia risk genes involved in glycosylation.

Results: Among GWAS results, the list of glycosylation enzymes unambiguously associated with schizophrenia continues to grow, with nearly a dozen prioritized genes implicated across several domains of protein glycosylation. Of the rare coding variants, protein products of the 3 genes directly involved in neurotransmission (GRIN2A, CACNA1G, and GRIA3) are all well known to be regulated through glycosylation. Post-mortem studies have identified changes in levels of glycoproteins and their transcripts within several glycosylation pathways, though these findings may result from the environment encountered while living with schizophrenia. Finally, mouse models of genetic risk for schizophrenia, including our work involving the SLC39A8-A391T common variant, display brain glycosylation changes in glycoproteins both genetically and functionally implicated in schizophrenia pathogenesis, including cell adhesion molecules and neurotransmitter receptors.

Conclusions: Several lines of evidence support a role for altered protein glycosylation in the pathogenesis of schizophrenia. As congenital disorders caused by severe mutations in many of the same genes can be effectively treated through targeted supplementation of enzymatic precursors and cofactors, the glycosylation pathway may represent a novel opportunity for therapeutic development in schizophrenia. Further, we hope this information will help researchers better understand and approach protein glycosylation in their studies of schizophrenia pathogenesis.

Keywords: Schizophrenia (SCZ), Genetics, Glycosylation

Disclosure: Nothing to disclose.

P476. OMgp Signaling as a Mechanism of Cortical Dendritic Regression

Melanie Grubisha*, Nicholas Lehman, Ryan Salisbury, Susan Erickson, Gregg Homanics, Matthew MacDonald, Robert Sweet

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

Background: Onset of gray matter reductions during adolescence, coupled with postmortem findings of smaller dendrites, have consistently been shown in schizophrenia (Sz). Once formed, dendrites are largely stable structures due to competing growth and regression signals. Understanding the pathways involved in this dynamic homeostasis is crucial to developing interventions targeting dendritic regression in Sz. We have recently shown that OMGp, a ligand for Nogo receptor 1 (NGR1), activates a signaling pathway that shifts this balance leading to dendritic regression. This pathway requires the active domain of KAL9. Knock-in of a KAL9 gain-of-function mutation (KAL9-PT) resulted in adolescent-onset dendritic regression, coincident with both the timing of normal developmental increase in OMGp expression and clinical symptom onset in SZ. We hypothesized that activation of OMGp/NGR1/KAL9 leads to alterations in the phosphorylation of downstream mediators of dendritic structure, some of which are Sz-risk genes.

Methods: We used OMGp stimulation in dissociated cortical culture followed by phosphoenrichment and mass spectrometry to identify the course of downstream signaling events using phosphoproteomics. Crispr/Cas9 was used to create a floxed OMGp mouse for developmental studies of OMGp KO on cortical dendritic architecture.

Results: Phosphorylation is a post-translational modification capable of regulating a protein's function, either activating or inactivating it depending on the site of phosphorylation. Among the proteins with significantly increased phosphorylation downstream of OMGp treatment were Trio and Cacna1g, both of which have an excess of loss of function mutations in SZ that are genome-wide significant. Loxp sites were successfully inserted flanking the OMGp gene without disrupting OMGp expression levels prior to recombination by Cre. Studies of dendritic architecture in OMGp KO mice are ongoing.

Conclusions: We identified an overlap between the adolescent-onset OMGp signaling pathway and genetic risk for Sz (ie Trio and Cacna1g). Using mouse models, we will next determine whether targeting these mediators in vivo during the normative adolescent increase in OMGp activity can be used to prevent or reverse dendritic regression. These findings provide a new opportunity for developing novel targeted therapies.

Keywords: Dendrites, Schizophrenia (SCZ), Adolescence

Disclosure: Nothing to disclose.

P477. 12 H Rhythm Abnormalities in the Human Dorsolateral Prefrontal Cortex of Subjects With Schizophrenia

Madeline Scott*, Wei Zong, Kyle Ketchesin, Marianne Seney, Bokai Zhu, George Tseng, Colleen McClung

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

Background: Schizophrenia (SZ) is a chronic neuropsychiatric illness associated with cognitive dysfunction and disrupted circadian behaviors. Consistent with circadian rhythm dysfunction, studies have demonstrated abnormal peripheral gene expression of circadian clock genes and changes in rhythmic

expression of hormones in individuals with SZ. Molecular rhythm patterns, however, have only just begun to be characterized in the human brain. Our lab has recently observed a distinct set of diurnally rhythmic transcripts in SZ subjects relative to a cohort with no history of psychiatric illness (non-psychiatric; NP) using rhythmic analysis of RNA-sequencing (RNA-seq) data from the dorsolateral prefrontal cortex (DLPFC), a region associated with executive dysfunction and heavily implicated in SZ. Pathway analysis demonstrated that genes that gained 24 h rhythmicity in SZ were associated with mitochondria dysfunction and GABA-ergic signaling, which is consistent with previously observed abnormal expression of transcripts associated with mitochondria and GABA-ergic signaling. A growing body of literature, however, has demonstrated that gene expression can also be regulated by ultradian rhythms (rhythms with a period < 24 h). 12 h rhythms are observed in various aspects of human behavior (sleep patterns, cognitive performance) and biology (body temperature, blood pressure, migraine onset, circulating hormone levels). 12 h transcript expression rhythms are enriched for mitochondria-associated proteins across species and tissues, suggesting 12 h rhythmicity is a conserved component of mitochondria gene expression and may have a potential role in mitochondria rhythmic disruptions in SZ DLPFC.

Methods: In the current study, we performed rhythmicity analyses on RNA-seq data obtained by the CommonMind Consortium through the NIMH Repository and Genomics Resource, a centralized national biorepository for genetic studies of psychiatric disorders, in both NP ($n = 104$) subjects and subjects with SZ ($n = 46$). Both sexes were included in all analyses. Prior to either analysis, time of death (TOD) was determined for each subject and normalized to a zeitgeiber time (ZT) scale. We then applied multiple types of rhythmicity analyses to each cohort of subjects, including cosinor nonlinear regression, the eigenvalue-pencil method, and a Lomb-Scargle analysis, followed by pathway (Ingenuity Pathway Analysis, Metascape) and motif enrichment (LISA Cistrome Combined) analyses.

Results: Multiple types of rhythmicity analyses identified 12 h rhythms in transcript expression in the human DLPFC. These transcripts were enriched for mitochondria-associated processes and motif enrichment analyses implicated ETS domain containing transcription factors. In subjects with SZ, fewer transcripts with 12 h rhythms were observed (800 transcripts, $p < 0.05$), and this loss of rhythmicity was associated with the unfolded protein response. In both cohorts, two timing patterns emerged, one in which transcripts peaked in expression in the morning/evening (ZT 2-3; ~9 AM/PM) and another in which transcripts peak during the afternoon/night (ZT 9; ~3 PM/AM). Transcripts associated with neuronal structural maintenance peaked during the afternoon/night in NP subjects but were not rhythmic in SZ subjects. Mitochondria-associated transcripts peaked in expression during the morning/evening in NP subjects, but during the afternoon/night in SZ subjects.

Conclusions: Overall, this study identified 12 h rhythms in transcript expression in human DLPFC. These rhythms are associated with fundamental cellular processes and had distinct timing patterns. However, in SZ, there is a strong reduction in the number of transcripts with 12 h rhythms. Additionally, in the NP cohort, transcripts associated with mitochondria peak in expression during transition periods (~9 AM/PM), in which people are likely switching from states of wake/sleep and/or fasting/eating and may require increased energy. However, in the SZ cohort these genes have an anti-phasic shift and peak during static periods (~3 PM/AM), implicating a deficit in mitochondria expression during transition periods in SZ. These data suggest alterations at multiple levels in the rhythmic regulation of mitochondria-associated genes in schizophrenia. Future work will be necessary to determine whether these changes underlie disruptions in energetic homeostasis and neuronal dysfunction in the DLPFC that could potentially contribute to symptoms of cognitive dysfunction in SZ.

Keywords: DLPFC, Circadian Rhythms, Postmortem Human Brain Tissue, Mitochondria

Disclosure: Nothing to disclose.

P478. Alterations in RBFOX1 and its Target Transcript in Prefrontal Cortical Dysfunction in Schizophrenia

Youjin Chung*, Daniel Chung, Samuel Dienel, Kenneth Fish, David Lewis

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: Current findings suggest that prefrontal cortical dysfunction in schizophrenia involves alterations in parvalbumin interneurons, but not calretinin interneurons. For example, molecular markers of inhibitory drive from parvalbumin interneurons are lower in the prefrontal cortex of schizophrenia subjects. In rodents, the strength of inhibitory drive from parvalbumin interneurons is partly regulated by RNA binding protein Rbfox1. Rbfox1 is alternatively spliced into isoforms that localize to either the nucleus (nuclear Rbfox1) or cytoplasm (cytoplasmic Rbfox1) of neurons. The binding of cytoplasmic Rbfox1 to its target transcripts increases the stability of, and therefore the cellular levels of these transcripts. The targets of cytoplasmic Rbfox1 are enriched for transcripts that regulate neurotransmitter release. Thus, we hypothesize that cytoplasmic Rbfox1 levels are altered in schizophrenia and could contribute to lower inhibitory drive from parvalbumin interneurons by disrupting the expression of its target transcripts.

Methods: Multi-label fluorescent immunohistochemistry was performed to quantify the protein levels of Rbfox1 in parvalbumin interneurons in the prefrontal cortex of schizophrenia and unaffected comparison subjects. Next, a prior microarray study that assessed the transcriptome of parvalbumin interneurons in schizophrenia and unaffected comparison subjects was explored to identify targets of cytoplasmic Rbfox1 that might be altered in schizophrenia. Finally, multi-label in situ hybridization combined with immunohistochemistry was used to quantify the levels of cytoplasmic Rbfox1 protein and its target mRNA in parvalbumin interneurons in schizophrenia and unaffected control subjects.

Results: In the prefrontal cortex of unaffected comparison subjects, levels of total Rbfox1 were significantly 4.0-fold greater in parvalbumin interneurons relative to calretinin interneurons. The levels of cytoplasmic or nuclear Rbfox1 were 9.6-fold or 3.0-fold greater in parvalbumin interneurons relative to calretinin interneurons, respectively. Levels of total Rbfox1 were significantly 27% lower in parvalbumin interneurons in schizophrenia relative to unaffected comparison subjects, with both cytoplasmic and nuclear isoforms of Rbfox1 showing similarly lower levels.

Given that levels of cytoplasmic Rbfox1 are markedly enriched in parvalbumin interneurons and show a reduction in schizophrenia, we explored whether the targets of cytoplasmic Rbfox1 are altered in schizophrenia. Of the 109 transcripts that have been identified as direct targets of cytoplasmic Rbfox1, levels of 7 transcripts were positively correlated with cytoplasmic Rbfox1 levels in parvalbumin interneurons across schizophrenia subjects. Of these 7 transcripts, Vamp1 was chosen for further study given its enrichment in parvalbumin interneurons and its role in regulating the strength of inhibitory drive from these neurons. The highest densities of Vamp1-immunoreactive cells were found in layer 4 of the prefrontal cortex where parvalbumin interneurons are enriched and show lower parvalbumin levels in

schizophrenia. Vamp1 mRNA levels were significantly 20% lower in parvalbumin interneurons in schizophrenia compared to unaffected comparison subjects. Finally, Vamp1 mRNA levels were positively correlated with cytoplasmic Rbfox1 levels in parvalbumin interneurons across both schizophrenia and comparison subjects.

Conclusions: Our findings demonstrate a cell type-specific enrichment of Rbfox1 in parvalbumin interneurons in the human prefrontal cortex. Additionally, we show that the levels of Rbfox1 and its two isoforms are lower in parvalbumin interneurons in schizophrenia. Finally, lower levels of cytoplasmic Rbfox1 in schizophrenia predicted lower levels of its downstream presynaptic target transcript, Vamp1, in parvalbumin interneurons. Together, these findings suggest that the lower level of cytoplasmic Rbfox1 in parvalbumin interneurons in schizophrenia may contribute to reduced inhibitory synaptic drive from these neurons by disrupting the stability of, resulting in lower levels of, its downstream transcript Vamp1. Given that Rbfox1 has been implicated in the genetic risk for schizophrenia, our findings suggest that lower inhibitory drive from parvalbumin interneurons in schizophrenia might arise, at least in part, from an intrinsic deficit in these neurons via altered regulation of the Rbfox1 pathway.

Keywords: Rbfox1, Parvalbumin Interneurons, Schizophrenia (SCZ), Prefrontal Cortex, Inhibitory Synaptic Transmission

Disclosure: Nothing to disclose.

P479. Effects of Psychiatric Disease and Aging on FKBP5/1 Expression are Specific to Cortical Supragranular Neurons

Natalie Matosin*, Darina Czamara, Janine Knauer-Arloth, Naguib Mechawar, Brian Dean, Gustavo Turecki, Thomas Hyde, Elisabeth Binder

Illawarra Health and Medical Research Institute, Wollongong, Australia

Background: Deducing genes capable of classifying biologically-distinct psychiatric subtypes, and their targets for treatment, is a priority approach for the field of psychiatry. FKBP5 is a gene with decades of evidence indicating its pathogenic role in a subset of psychiatric patients, with high potential to be leveraged as a therapeutic target for these individuals. While it is widely reported that FKBP5/FKBP51 protein (FKBP5/1) expression is impacted by psychiatric disease state, risk genotype and age, in which cell-types and sub-anatomical brain areas FKBP5/1 is specifically affected is not known. This knowledge is critical to propel FKBP5/1-targeted treatment development.

Methods: We performed an extensive, large-scale postmortem study (n = 1024, 6 cohorts) of FKBP5/1 examining dorsolateral prefrontal/orbitofrontal cortex (BA9, BA11, BA24) samples derived from subjects that lived with schizophrenia, major depression or bipolar disorder. With an extensive battery of RNA (bulk RNA sequencing, single-nucleus RNA sequencing, microarray, qPCR, RNAscope) and protein (immunoblot, immunohistochemistry) analysis approaches, we thoroughly investigated the effects of disease-state, aging and genotype on cortical FKBP5/1 expression.

Results: Our results demonstrate that FKBP5/1 cortical expression was strikingly increased (+17-40%) in individuals with schizophrenia or depression (+24%) vs controls. We also observed a strong effect of age with heightened FKBP5/1 expression in older psychiatric subjects versus older controls (e.g., mRNA: R = 0.664, P(FDR) = 5.6E-06). Further examination of the cell-type specificity of these findings with single nucleus RNA sequencing (snRNAseq) and targeted RNAscope/immunohistochemistry demonstrated that the disease- and aging-effects on FKBP5/1 expression are specific to supragranular neurons in cortical layer 3, with +25-32% increase specifically in supragranular layer 2-3 excitatory neurons in both schizophrenia and depression.

Conclusions: Our results provide a new contribution to the field, indicating that effects of psychiatric disease-state and age converge on supragranular neurons of the superficial cortical layers, thus pinpointing a clear cellular target for future drug development.

Keywords: Early Life Stress (ELS), FKBP5, Schizophrenia Novel Treatment, Major Depression, Schizophrenia (SCZ)

Disclosure: Nothing to disclose.

P480. The Clinical Candidate Xanomeline Displays a Dual Orthosteric and Allosteric Binding Profile at the M4 mAChR

Wessel A.C. Burger, Vi Pham, Alexander Powers, Ziva Vuckovic, Jesse I. Mobbs, Alisa Glukhova, Denise Wootten, Andrew B. Tobin, Patrick M. Sexton, Steven M. Paul, Christian Felder*, Radostin Danev, Arthur Christopoulos, Ron O. Dror, Celine Valant, David M. Thal

Karuna Therapeutics, Indianapolis, Indiana, United States

Background: The M4 muscarinic acetylcholine receptor (M4 mAChR) has emerged as a drug target of high therapeutic interest due to its expression in regions of the brain involved in the regulation of psychosis, cognition, and addiction. The M1 and M4 mAChR preferring investigational drug candidate, xanomeline, has demonstrated clinical efficacy in the Positive and Negative Symptom Scale (PANSS) in a Phase IIb clinical trial for the treatment of schizophrenia. Initially, xanomeline had been considered to bind only to the orthosteric acetylcholine binding site of the mAChRs. However, recent studies have shown much greater complexity as to the precise nature of ligand-receptor binding interactions including efficacy-driven selectivity, subtype-dependent wash-resistant binding, and an atypical interaction with positive allosteric modulators. Understanding how xanomeline binds to the mAChR family and the molecular mechanism behind its unique pharmacological profile are key for the design of novel antipsychotics that target the M4 mAChR.

Methods: We determined a cryogenic electron microscopy (cryo-EM) structure of xanomeline bound to the human M4 mAChR in complex with the heterotrimeric Gi1 transducer protein. Molecular dynamics (MD) simulations were used to corroborate the additional allosteric binding mode that was identified in the cryo-EM structure. The allosteric binding mode of xanomeline was further validated using site-directed mutagenesis.

Results: Unexpectedly, two molecules of xanomeline were found to simultaneously bind to the M4 mAChR. One molecule was bound to the orthosteric acetylcholine binding site and a second molecule was bound to the canonical mAChR allosteric binding site. MD simulations support the structural findings by showing that xanomeline was able to stably bind to the allosteric binding site in a manner similar to the positive allosteric modulator LY2033298. In contrast, the agonist iperoxo does not stably bind to the allosteric site, suggesting the dual orthosteric and allosteric binding profile is specific to xanomeline. Pharmacological validation using kinetic dissociation assays confirmed xanomeline can act as an allosteric modulator and mutation of well-known key allosteric site residues to alanine abolished the allosteric effect of xanomeline.

Conclusions: The clinical candidate xanomeline is capable of binding to both the orthosteric and allosteric sites of the M4 mAChR. Historically, xanomeline has shown a complex pharmacological profile, and the findings of this study provide a potential explanation and serve as the basis for future studies that will include the other mAChR subtypes. Collectively, these findings will inform future rational drug design at the M4 mAChR.

Keywords: M1 and M4 Muscarinic Receptors, Schizophrenia (SCZ), Xanomeline

Disclosure: Karuna Therapeutics: Employee (Self), Karuna Therapeutics: Stock / Equity (Self)

P481. Higher Levels of AKT-Interacting Protein in the Frontal Pole From a Sub-Group of Schizophrenia Patients With Markedly Lower Levels of Muscarinic M1 Receptors

Megan Snelleksz*, Brian Dean

The Florey Institute of Neuroscience and Mental Health, Melbourne, Australia

Background: We have shown there are lower levels (-23%) of AKT-interacting protein (AKTIP) RNA in the frontal pole from people with schizophrenia, a region which has a significant role in its pathophysiology. It is now argued that studying subgroups within schizophrenia is fundamental to understanding its molecular pathology. Our laboratory has defined a subgroup, termed Muscarinic Receptor Deficit Schizophrenia (MRDS), that is defined by markedly lower levels of the muscarinic M1 receptor (CHRM1) in a quarter of people with schizophrenia. We have also shown that AKTIP RNA is higher (+57%) in the cortex of CHRM1 knockout mice. Hence, we now seek to determine if levels of AKTIP protein are altered only in the cortex from patients with MRDS as a result of altered CHRM1 signaling.

Methods: This study uses autoradiography to measure CHRM1 binding in the frontal pole from people with schizophrenia and controls to first establish a cohort of 19 MRDS (15 male, 4 female), 19 non-MRDS (15 male, 4 female) and 19 controls (12 male, 7 female). Based on our gene expression studies in human frontal pole and mouse cortex, this study uses Western Blotting to determine if the lower levels of AKTIP RNA translates to lower levels of protein in the frontal pole from people with schizophrenia and if such changes were limited to those with MRDS due to altered CHRM1 signalling.

Results: This study found that CHRM1 binding is 90% lower in the frontal pole from MRDS, with no differences between non-MRDS and controls. This study also found higher levels (+43%, $p = .02$) of AKTIP protein in the frontal pole from people with schizophrenia. However, when separating schizophrenia into 2 subgroups (MRDS and non-MRDS), this increase in AKTIP protein was specific to the MRDS group (+47%, $p = .05$), with no differences between non-MRDS and controls.

Conclusions: Our findings suggest that, as predicted, AKTIP is altered in the cortex of patients with MRDS possibly due to altered CHRM1 signalling. Significantly, CHRM1 binding in the frontal pole from MRDS was profoundly lower than any other region previously studied. The cohort used in our previous transcriptomic study was predominantly patients who were non-MRDS and therefore the lower levels of AKTIP RNA in those subjects may be an attempt to normalize levels of that protein. As AKTIP is implicated in cell signaling and vesicle trafficking, our data suggests these important functions may be particularly affected in the MRDS subtype of schizophrenia.

Keywords: Schizophrenia Subtypes, M1 Muscarinic Receptors, Postmortem Human Brain Tissue, Frontal Pole (Brodmann's Area 10), Schizophrenia

Disclosure: Nothing to disclose.

P482. Altered Expression of Excitatory and Inhibitory Ionotropic Receptor Subunit across the Cortical Visuospatial Working Memory Network in Schizophrenia

Kirsten Schoonover*, Samuel Dienel, Holly Bazmi, Nora Miller, David Lewis

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: Working memory dysfunction in individuals with schizophrenia is thought to reflect altered excitatory and inhibitory neurotransmission across multiple regions of the cortical visuospatial working memory (vsWM) network. However, key ionotropic glutamatergic and GABAergic receptor subunits have only been studied in the dorsolateral prefrontal cortex (BA46).

Methods: Using qPCR on total gray matter homogenate samples from BA46, posterior parietal cortex (BA7), and primary (BA17) and association (BA18) visual cortices, we quantified transcript levels of critical subunits for excitatory N-methyl-D-aspartate receptors (NMDARs), excitatory alpha-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid receptors (AMPARs), and inhibitory GABAA receptors (GABRAs) in 20 matched pairs of schizophrenia (SZ) and unaffected comparison (UC) subjects.

Results: In UC subjects, AMPAR and NMDAR levels generally exhibited opposite rostral-to-caudal gradients, with AMPAR GRIA1 and GRIA2 expression highest in BA46 and NMDAR GRIN1 and GRIN2A expression highest in BA17; however, the regional pattern of NMDAR GRIN2B expression was like that of AMPARs. GABRA5 and GABRA1 levels were highest in BA46 and BA17, respectively. In SZ subjects, levels of all transcripts (except GRIN2B and GABRA5) were lower in caudal regions, with no differences in BA46.

Conclusions: Our analyses of transcript levels across regions of the cortical vsWM network revealed distinct regional patterns of ionotropic glutamatergic and GABAergic receptor subunits in UC subjects, suggesting that balances between excitation and inhibition are achieved in a region-specific manner. In SZ subjects, the distinct alterations in excitatory and inhibitory receptor transcripts across regions suggests differential contributions of each region to impaired WM performance in the illness.

Keywords: Postmortem Brain Tissue Gene Expression, AMPA Glutamate Receptors, NMDA Glutamate Receptors, GABA-A Receptors, Schizophrenia (SCZ)

Disclosure: Nothing to disclose.

P483. Prefrontal D2 Dopamine Receptor Regulates Psychomotor Symptoms via Controlling Striatal Cholinergic Modulation, an Intersectional CRISPR/Cas9 Driven Circuit Mapping

Clementine Quintana, Jivan Khlghatyan, Lakshmi Rajakrishna, Jean-Martin Beaulieu*

University of Toronto, Toronto, Canada

Background: Psychomotor abnormalities have been abundantly reported in psychiatric disorders like bipolar disorder (BD), schizophrenia and major depressive disorder. Although neglected in contemporary psychiatry, studies have focalized to psychomotor dimension, which has been recently proposed and officially included as an additional domain in the RDoC. The dopamine D2 receptor (DRD2) remains the principal target of antipsychotic drugs used for the management of psychomotor agitation. Besides the high expression of DRD2 in striatum, DRD2 is expressed into the prefrontal cortex where it has been shown its expression upregulated in BD dorsolateral prefrontal cortex (PFC) in post-mortem tissues patients. However, the function of DRD2 in PFC in psychomotor symptoms in bipolar mania remains unknown.

Methods: Amphetamine-induced psychomotor excitability allowed a predictive validity model that mimicking many aspects of bipolar mania in humans. We employed intersectional CRISPR/Cas9 gene editing to investigate the role of DRD2 in PFC in adult

mice and characterize the circuits that are regulated by this receptor.

Results: The deletion of DRD2 in PFC induced an abolition of amphetamine-induced psychomotor behavior without inducing extrapyramidal side effects and cognitive deficit. An investigation of prefronto-striatal circuit revealed the role of PFC DRD2 projecting neurons into the regulation of the cholinergic interneuron activity. Injection of an antagonist of muscarinic receptors rescued the psychomotor behavior induced by amphetamine in mice lacking DRD2 in PFC.

Conclusions: This study highlights the role of PFC DRD2 in regulation of psychomotor symptoms via the modulation of striatal cholinergic interneuron activity. This comprehensive analysis paves the way for re-examination of cortical DRD2 functions and treatments in psychomotor disorders.

Keywords: Hypofrontality, D2 Dopamine Receptor, Acetylcholine, CRISPR/Cas9, RNAseq

Disclosure: Nothing to disclose.

P484. The Relationship Between Synaptic and Cognitive Markers in Schizophrenia: A Positron Emission Tomography Study Using [11 C]UCB-J

Ellis Onwordi*, Thomas Whitehurst, Ekaterina Shatalina, Ayla Mansur, Atheeshaan Arumuham, Martin Osugo, Ben Statton, Alaine Berry, Marina Quinlan, Declan O'Regan, Maria Rogdaki, Tiago Reis Marques, Eugenii Rabiner, Roger Gunn, Sridhar Natesan, Oliver Howes

Bart's and The London, Queen Mary's School of Medicine, London, United Kingdom

Background: Cognitive impairment remains an unmet clinical need in the treatment of schizophrenia, and its pathoaetiology is poorly understood. Converging lines of evidence implicate synaptic loss in schizophrenia pathogenesis. Recent studies have used positron emission tomography with novel radioligand [11 C]UCB-J to index levels of synaptic vesicle glycoprotein 2 A (SV2A) in vivo as a proxy for synaptic density, finding lower levels of [11 C]UCB-J binding in the frontal cortex and hippocampus in patients with schizophrenia compared to healthy volunteers. Using [11 C]UCB-J PET, we tested directly whether SV2A levels and markers of cognitive function are linked in schizophrenia.

Methods: Thirty-two volunteers with schizophrenia and 24 healthy volunteers underwent [11 C]UCB-J PET and T1-weighted structural MRI. Volumes of distribution (VT) were estimated for two regions of interest (ROIs) (frontal cortex and hippocampus). Cognitive performance was tested using the National Adult Reading Test (NART), which evaluates premorbid intelligence, and Rey's Auditory Verbal Learning Test (AVLT). We explored relationships between NART and AVLT scores and VT using Pearson's correlation coefficient.

Results: There was a positive relationship between NART score and [11 C]UCB-J VT in the frontal cortex (FC, $r = 0.46$, $p = 0.01$, 95% CI = 0.11 to 0.71) and hippocampus ($r = 0.72$, $p = 0.046$, 95% CI = 0.02 to 0.45) in the schizophrenia group ($n = 29$). There were no significant relationships between NART score and [11 C]UCB-J VT in any region of interest in the healthy volunteer group ($n = 21$).

There was a positive relationship between Trial I AVLT score and [11 C]UCB-J VT in the frontal cortex (FC, $r = 0.42$, $p = 0.02$, 95% CI = 0.08 to 0.67) but not the hippocampus ($r = 0.32$ $p = 0.07$, 95% CI = -0.03 to 0.60) in the schizophrenia group ($n = 32$). There were no significant relationships between Trial I AVLT score and [11 C] UCB-J VT in either region of interest in the healthy volunteer group ($n = 24$).

There was a positive relationship between AVLT recognition score and [11 C]UCB-J VT in the frontal cortex (FC, $r = 0.46$, $p = 0.02$, 95% CI = 0.07 to 0.73) and the hippocampus ($r = 0.54$ $p = 0.007$, 95% CI = 0.17 to 0.77) in the healthy volunteer group ($n = 24$). There were no significant relationships between AVLT recognition score and [11 C]UCB-J VT in either region of interest in the schizophrenia group ($n = 27$).

There were no significant relationships between total words learned and [11 C]UCB-J VT in either region of interest in the schizophrenia ($n = 32$) or healthy volunteer group ($n = 24$).

Conclusions: SV2A levels are linked to measures of premorbid intelligence and verbal learning but not recognition in schizophrenia, supporting the hypothesis that synaptic dysfunction in vivo is linked to cognitive performance in schizophrenia.

Keywords: SV2A PET imaging, [11 C]UCB-J, Cognition, Schizophrenia, Synaptic Aberrations, PET Imaging

Disclosure: Nothing to disclose.

P485. Plasma Free Fatty Acids After Six Weeks of Antipsychotics Treatment of Schizophrenia

Gregory Oxenkrug*, Hans-Gert Bernstein, Henrik Dobrowolny, Paul Summergrad, Johann Steiner

Tufts University School of Medicine, Boston, Massachusetts, United States

Background: Free (non-esterified) fatty acids (FFA) are partially derived from myelin, a phospholipid sheath that surrounds axons. Elevated blood FFA levels were observed in schizophrenia patients and may reflect an increased degradation of myelin [1]. The data on the effect of antipsychotics on FFA are sparse. In in vitro study, clozapine (but not haloperidol) decreased FFA content in cultured oligodendrocytes and increased the expression of the rate limiting enzyme of FFA synthesis [2]. In healthy volunteers, plasma FFA decline was observed after 3 days of administration of olanzapine [3] but not of haloperidol [4]. In schizophrenia patients, plasma FFA decline was observed after treatment with olanzapine ($n = 20$) and risperidone ($n = 14$) but not abilify ($n = 16$) [5].

Methods: Plasma FFA levels were evaluated in acutely ill and either antipsychotic-naive or antipsychotic-free for at least six weeks patients, before and after 6 weeks of inpatient treatment with olanzapine ($n = 16$), risperidone ($n = 22$) or quetiapine ($n = 12$). Positive and Negative Syndrome Scale (PANSS) total scores were previously evaluated in these patients [6]. Statistical difference between pre- and post-drug treatment were assessed by paired T-test

Results: Plasma FFA were higher in patients ($0.779 + 0.469$ nM, $n = 50$) that in controls ($0.427 + 0.427$, $n = 50$). Plasma FFA levels decreased after treatments with olanzapine (by 55% to 0.345 nM, $p < 0.0004$), and risperidone (by 60% to 0.320, $p < 0.004$), but not quetiapine (0.519 nM, $p < 0.418$). PANSS total corrected scores before treatment were 56.895. Olanzapine treatment resulted in PANSS decrease to 25.500 (by 55%, $p < 0.0001$), risperidone - to 20.217 (by 65%, $p < 0.001$) and quetiapine - to 39.5000 (by 40%, $p < 0.002$).

Conclusions: Plasma FFA levels decreased after olanzapine and risperidone treatment in agreement with previous reports [2,3]. Quetiapine did not change FFA levels similar to haloperidol and abilify [4,5]. Notably, the mean decrease of PANSS total scores was lower in patients treated by quetiapine than in patients treated by olanzapine and risperidone, although we did not observe correlations between FFA and PANSS scores [5]. Early (e.g., 3 days) [4] response of plasma FFA to antipsychotics may help to select an optimal medication for the treatment of a schizophrenia endophenotype characterized by cell membrane phospholipid impairments [7].

References:

1. Kaiya H, Horrobin DF, Manku MS, Fisher NM (1991) Biol Psychiatry; 30: 357–362

2. Steiner J, Martins-de-Souza D, Schiltz K, Sarnyai Z, et al (2014) Front Cell Neurosci 2014) Nov 18;8:384

3. Albaugh VL, Singareddy R, Mauger D, et al (2011) PLoS One. 6(8): e22662

4. Vidarsdottir S, Weenen JE, Frolich M, Roelfsema F, et al. (2010). J Clin Endocrinol Metab 95: 118–125.

5. Kaddurah-Daouk R, J McEvoy1, RA Baillie, D Lee, et al (2007) Molecular Psychiatry 12, 934–945

6. Martins-de-Souza D, Solari FA, Guest PC, Zahedi RP and J Steiner (2015) Schizophrenia Article number: 15050; doi:10.1038/npjschz.2015.50

7. Messamore, E. (2018) Prostaglandins Leukot. Essent. Fat. Acids 136, 95–97.

Keywords: Free Fatty Acids, Antipsychotics, Schizophrenia Subtype

Disclosure: Nothing to disclose.

P486. A Subset of Olfactory Neurons Derived From Schizophrenia Patients Exhibit Increased Markers for Protein Aggregation And Reduced Cognitive Performance

Leslie Nucifora, Koko Ishizuka Ishizuka, Gayane Yenokyan, Nicola Cascella, David Schretlen, Philip Harvey, Christopher Ross, Russell Margolis, Akira Sawa, Frederick Nucifora*

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

Background: Schizophrenia is a debilitating disorder that affects 1% of the population. Symptoms associated with schizophrenia include positive and negative symptoms as well as cognitive deficits. Significant efforts have been made towards understanding the genetic factors underlying schizophrenia, however, the mechanisms implicated in schizophrenia remain unclear. The heterogeneity in clinical symptoms and course of illness, suggest the possibility of different mechanist subtypes of the illness. Reducing the heterogeneity by subtyping patients could help identify the underlying mechanisms. One approach is to subtype patients by clinical presentation or course and determine the mechanism underlying the clinical presentation. Alternatively, pathogenic processes could be identified in a subset of patients, and then correlated with clinical phenotypes. While this biological approach has helped other medical fields advance, it has had less application to research in mental illness. Previously, we reported increased protein insolubility and ubiquitination, two markers for protein aggregation, as a pathological process in human postmortem brain from a subset of patients with schizophrenia. This is an example of biological subtype classification for schizophrenia. In the present study, we investigate whether the process of protein aggregation is observed in an independent model system using olfactory neurons derived from living patients with schizophrenia. Utilizing a complementary system to autopsy brains such as olfactory neuronal cells provides the advantages that they can be acquired from living subjects, with clinical and cognitive data obtained at the time of assessment. In the present study, we build upon our previous work from human autopsy brains and now show that a subset of living patients with schizophrenia exhibit increased markers of protein aggregation as a mechanism utilizing olfactory neurons. We further identify potential clinical and cognitive impairments that relate to this subtype of the illness.

Methods: Olfactory neurons were obtained from the Johns Hopkins Schizophrenia Center. Twenty-three schizophrenia patients and nineteen controls with both sexes were included. Harvested olfactory neurons were processed utilizing a sarkosyl cellular fractionation protocol designed to isolate the insoluble protein fraction. All purified insoluble protein samples were processed by western blot and Coomassie gel analysis and insoluble protein levels and ubiquitin reactivity were quantified using ImageJ software and normalized to total homogenate protein concentrations. Clinical and cognitive testing were performed at the time of olfactory neuron obtainment.

Results: A subset of living patients with schizophrenia exhibit increased protein insolubility and ubiquitination, two markers for protein aggregation, utilizing olfactory neurons. Significant reduction in cognitive performance was observed across every cognitive measure except for processing speed, in the subset of schizophrenia participants with increased protein aggregation compared to schizophrenia participants without aggregation and controls.

Conclusions: These studies further help to establish protein aggregation as a novel mechanism involved in the pathogenesis of a subset of patients with schizophrenia that may relate to specific symptomatology. Further exploration of the mechanisms leading to protein insolubility could lead to novel therapeutic targets and to a reconceptualization of diagnostic categories.

Keywords: Schizophrenia, Protein Aggregation, Ubiquitination, Cognitive Impairment

Disclosure: Teva: Consultant (Self), Galyan Bio: Advisory Board (Self)

P487. Alterations of SGK1 Expression and Activity in Schizophrenia

Abdul Hammoud, Ali Imami, Consuelo Walss-Bass, Robert McCullumsmith*

University of Toledo, Toledo, Ohio, United States

Background: We conducted RNA-sequencing of hiPSC-derived cell lines from schizophrenia (SCZ) subjects, from the Central Valley of Costa Rica. hiPSCs, neural precursor cells, and cortical neurons derived from six healthy controls and seven SCZ subjects were generated using standard methodology. We identified 454 differentially expressed genes in hiPSC-derived neurons, enriched in pathways including phosphoinositide 3-kinase/glycogen synthase kinase 3 (PI3K/GSK3) signaling, with serum-glucocorticoid kinase 1 (SGK1), an inhibitor of glycogen synthase kinase 3β, as part of this pathway. We further found that pharmacological inhibition of downstream effectors of the PI3K/GSK3 pathway, SGK1 and GSK3, induced alterations in levels of neurite markers βIII tubulin and fibroblast growth factor 12, with differential effects in patients compared to controls. These studies support a role for disruption of PI3K/GSK3 and SGK1 signaling as a risk factor for SCZ. To further investigate the role of these kinases in SCZ we developed an in-vitro kinome array assay, to permit assessment of putative pharmacological modulators.

Methods: hiPSC-derived cell lines were generated from schizophrenia (SCZ) subjects from the Central Valley of Costa Rica. RNA was isolated and RNAseq analyses performed to assess differential gene expression. We used the serine threonine PamGene kinomearray chip to develop a specific assay for SGK1 kinase activity. The kinome array assay was run in the presence of recombinant SGK1 protein (2.5 ng, 25 ng, and 250 ng). For the negative control we heat inactivated 250 ng. All assays were run in triplicate. We used our standard kinomic analyses, including KRSA and Kinopedia developed in the McCullumsmith laboratory to analyze our data. To determine protein kinase (hits) we used a Monte Carlo simulation that generates 2000 simulations of protein kinase assignments to peptides. We used this permutation analysis to determine which protein kinase hits are over or

underrepresented in our dataset. A 95% confidence interval is used to determine the range of expected protein kinase hits. When an observed protein kinase falls outside the 95% confidence interval that is expected it is moved forward as a candidate for further confirmation study. All kinome array assays are run in triplicate, and the recombinant protein kinase studies are run with a heat inactivated sample as our negative control.

Results: We identified 11 peptides on the array that have high affinity for SGK1 phosphorylation activity. Pathway analysis of these peptides using EnrichR confirmed several well characterized pathways associated with SGK1 signaling, as well as several novel pathways not previously attributed to this protein kinase. Based on our transcriptional profiling results from hiPSCs derived from SCZ subjects we reexamined our published data for the serine threonine kinome array in postmortem brain in SCZ with a focus on SGK1. Using our KRSA we found increased activity of SGK1 kinase in SCZ in the frontal cortex compared to control ($P > 0.05$).

Conclusions: Converging evidence suggest the perturbations of SGK1 protein kinase activity may contribute to the pathophysiology of severe mental illness; in particular, molecular correlates of learning and memory including long-term potentiation and long-term depression may be impacted by subtle changes in protein signaling networks. In this study we have developed an assay that will allow us to interrogate drugs that modulate protein kinase implicated in SCZ including SGK1. We are currently assessing novel compounds as well as repurposed FDA approved drugs as candidates that may modulate SGK1 in a manner that could lead to improvement in neuroplastic endophenotypes.

Keywords: Schizophrenia, Kinome, Protein Kinase, Bioinformatics

Disclosure: Nothing to disclose.

P488. Extracellular Matrix and Vasculature Dysregulation May Impair Neurogenesis in Schizophrenia Cases With Elevated Inflammation

Hayley North*, Christin Weissleder, Maina Bitar, Theresa Salthouse, Guy Barry, Maree Webster, Jan Fullerton, Cynthia Shannon Weickert

Neuroscience Research Australia, Randwick, Australia

Background: Schizophrenia is a clinically and neuropathologically heterogeneous disorder. We find a subgroup of ~40% of people with schizophrenia have elevated inflammation in various brain regions including the brain's largest niche for neurogenesis, the subependymal zone (SEZ). This subgroup of schizophrenia cases with elevated cytokines have exaggerated changes of transcripts indicating increased stem cell quiescence/dormancy and reduced neuronal differentiation. In addition, the high inflammation schizophrenia group have suppressed microglia marker expression and increased density of macrophages. While studies show that macrophages can secrete factors that may impair neurogenesis, we do not know if the broad molecular changes are consistent with this possibility, or if other factors are key to inflammatory-related suppression of neuronal differentiation in schizophrenia. This research aimed to discover broad transcriptomic differences relating to SEZ inflammatory status within schizophrenia to elucidate disease heterogeneity.

Methods: Deep total-RNA sequencing was performed on RNA extracted from the post-mortem SEZ tissue of 27 schizophrenia cases previously designated into low inflammation ($n = 13$) and high inflammation ($n = 14$) subgroups based on cluster analysis of inflammation marker gene expression (starting with IL6, IL6R, IL1β, IL1R1, CXCR8, SERPINA3 and IL6ST). Differentially expressed (DE) genes were determined by EdgeR software with a false discovery rate adjusted (FDR) p value < 0.05. The DE gene list was subsequently analysed using Ingenuity Pathway Analysis. Immunohistochemistry was conducted on 14μm thick SEZ sections from the same cohort.

Results: 718 genes were DE in high compared to low inflammation schizophrenia (FDR $p < 0.05$). The DE genes were most significantly over-represented in the pathway 'Hepatic Fibrosis/Hepatic Stellate-Cell Activation'. Genes in this pathway, which predominantly had increased expression in high inflammation schizophrenia, encoded proteins involved in extracellular matrix (ECM) rigidity (including ten collagens) and remodelling of the vasculature (including angiogenesis genes VEGFA, VEGFR1, EDN1). Collagen-IV was primarily localised around blood vessels and in the SEZ hypocellular gap. The results suggest novel changes to the SEZ vasculature, which may facilitate immune cell transmigration as indicated by positive correlations between markers of angiogenesis, macrophages and recruitment molecule ICAM1.

Conclusions: This is the first discovery-driven comparison of the transcriptome between inflammatory subgroups in schizophrenia brain tissue. The findings of inflammation-dependent changes in the SEZ suggesting angiogenesis and ECM alterations have important implications for how inflammation contributes to heterogeneity in schizophrenia neuropathology; especially regarding reduced neurogenesis. Considering the SEZ microenvironment and vasculature intricately regulate neurogenesis, alterations to the ECM and collagens surrounding blood vessels would likely dysregulate the development and migration of newborn neurons. This exploration of disease heterogeneity may be a step towards developing more personalised treatment options for those with elevated inflammation.

Keywords: Schizophrenia (SCZ), Inflammation, Subgroups, Neurogenesis

Disclosure: Nothing to disclose.

P489. Impaired Microtubule-Based Structural Dynamic Changes in Monocyte-Derived-Neuronal-Like Cells (MDNCs) From Patients With Schizophrenia

Alonso Cortez-Resendiz, Lauren Forrest, Anjali Iyer, Janani Iyer, Alfredo Bellon*

Penn State Milton S. Hershey Medical Center, Hershey, Pennsylvania, United States

Background: Ample evidence indicates the pathophysiology of schizophrenia (SCZ) begins in early neurodevelopment (1) but the cellular process at fault is yet to be identified. Decades of postmortem research have shown subtle abnormalities in the neuronal structure across the brain that often involve reductions in size and number of neuronal extensions (2-5). But increased number of extensions (6,7) and malformed neurites have also been found (8,9). These findings could suggest that the capacity to modify the neuronal structure is impaired in SCZ. During early human development, neurons undergo dynamic structural changes consisting of continuous formation and retraction of neurites (10). Data from cellular models of neurodevelopment support the possibility of impaired neurostructural dynamics in SCZ. Maturing neurons generated from patients' pluripotent cells evidenced longer neuronal extensions early in development (10, 11) and shorter neuronal outgrowths in more advanced stages of differentiation (12, 13). Microtubules are an indispensable component of the cytoskeleton that play a significant role in sculpting the neuronal shape (14). In patients with SCZ, dynamic changes in microtubules are diminished in olfactory neuroepithelial cells (15), while neuronal precursors generated from the olfactory epithelium showed altered microtubule organization

(16). Whether deficits in microtubule polymerization also affect neurostructural dynamic changes in SCZ remains unknown.

Methods: Human monocytes were transdifferentiated following our protocol (17). After transdifferentiation, light microscopy pictures of the same group of Monocyte-Derived-Neuronal-like cells (MDNCs) were taken at time zero (T0) and then after an hour of incubation (T1hr) either under control conditions or after treatment with colchicine 0.4 μM, a compound that arrests microtubule polymerization. Each single MDNC received a numeric score based on a scoring rubric we developed (18). The sum of all structural changes present in all MDNCs in one hour is the Structural Dynamic Index (SDI). Experiments were conducted with blood samples from 12 controls and 13 patients with SCZ frequency-matched by age and gender as reported previously (19). This study was approved by IRBs from Penn State Hershey and INSERM, France.

Results: We have recently developed a protocol to transdifferentiate blood circulating monocytes into neuronal-like cells in 20 days and without reprograming (17). MDNCs express several neuronal markers and conduct spontaneous action potentials as well as postsynaptic inhibitory and excitatory currents (17). Moreover, MDNCs deliver reproducible results in sequential samples from the same donors (17,18). We have also shown that these cells retract neuronal extensions when treated with colchicine similarly to human neurons (18). In addition, MDNCs from a subgroup of patients with SCZ prune more primary neurites in the presence of dopamine than cells from controls (18). Here we show, based on a mixed model analysis, that the number of differentiated cells between cohorts in comparable (CTL, 6.7 ± 0.95; SCZ, 8.0 ± 0.9; $P = 0.31$; $N = 600$ MDNCs from 12 CTL and 802 MDNCs from 13 patients). At baseline, SDI is similar among groups (CTL, 13.9 ± 1.2; SCZ, 12.0 ± 1.0; $P = 0.25$; $N = 465$ MDNCs from 8 CTL and 879 from 10 patients). But after treatment with colchicine 0.4 μM SDI in patients is higher (CTL, 7.4 ± 1.9; SCZ, 12.7 ± 1.4, $P = 0.06$; effect size, 1.16; $N = 251$ MDNCs from 4 CTL and 393 from 7 patients).

Conclusions: MDNCs from patients with SCZ remain structurally dynamic after treatment with colchicine 0.4 μM but show no differences at baseline when compared with CTL cells. These results suggest that there is an impaired microtubular response in SCZ that interferes with neurostructural dynamic changes. While we have previously reported that haloperidol does not affect SDI (20), the potential influence of other antipsychotics needs to be studied.

Keywords: Adult Stem Cells, Neurodevelopment, Monocytes, Schizophrenia (SCZ), Cytoskeleton

Disclosure: Nothing to disclose.

P490. Cognition as a Moderator of Relapse Risk Among Patients With Early Phase Schizophrenia Treated With Long-Acting Injectable Antipsychotics: Data From the Prelapse Trial

Delbert Robinson*, Nina Schooler, Majnu John, Patricia Marcy, John Kane

Hofstra/Northwell, Glen Oaks, New York, United States

Background: Cognitive deficits are a well-established component of early phase psychosis. Cognition has been found to moderate initial response to antipsychotic treatment with first episode patients. A next step question is whether cognition is also associated with relapse. Studies examining relapse following response among recent onset patients have found variable results. Relapse studies require long follow-up periods and long-term adherence to antipsychotic treatment is challenging for many recent onset patients. Variable long-term antipsychotic adherence may be one factor that has limited the ability to detect possible relationships between cognition and relapse among recent onset patients. Studies with long-acting injectable antipsychotic (LAI) formulations limit nonadherence effects and therefore may be particularly useful for detecting cognition moderator effects on relapse risks.

Methods: The PRELAPSE trial was a large simple trial with cluster randomization of 39 US clinics. Nineteen clinics provided participants the LAI aripiprazole monohydrate (AM) and 20 provided treatment as usual (TAU). The primary outcome was time to first hospitalization; the observation period was 2 years. Inclusion criteria were: DSM-5 schizophrenia diagnosis; fewer than 5 years of lifetime antipsychotic treatment; age 18–35 years. Cognition was assessed with the Repeatable Battery for the Assessment of Neuropsychological Status (RBANS) at baseline and then yearly. The RBANS had the advantage of easy administration for a large simple trial. The RBANS consists of ten subtests that give five domain scores and a total score. For analysis of cognition as a moderator of hospitalization outcomes, the sample was characterized as having poorer or better cognition based upon a median split of the baseline RBANS total score. Time to first hospitalization was analyzed using a Cox Proportional Hazard model with shared random effects for sites (also known as a Shared Frailty model) accounting for clustering effects. A lognormal distribution was assumed for the frailties.

Results: The examination of baseline RBANS as a moderator of time to first hospitalization included 457 of the 489 participants in the PRELAPSE trial. Mean age of the cognition sample (342 men and 115 women) was 25.2 years. Mean duration of prior antipsychotic treatment was 627 days. Poor and better cognition groups did not differ on age, duration of prior antipsychotic treatment or on being at an AOM or TAU site. Women were more likely to be in the better cognition group than men (71 of 115 women versus 159 of 342 men). The Cox Proportional Hazard analyses revealed a treatment condition by baseline cognition interaction (Wald Chi-Square = 3.3540, df =1, $p = 0.0506$). Subsequent analyses of participants in the poorer cognition group ($N = 227$) found no effect of treatment condition (Wald Chi Square = 0.6792; df=1; $p = 0.2367$); the hazard ratio for AM versus TAU was 0.776 (95% CI = 0.424, 1.419). In contrast, among participants with better cognition, there was a significant treatment effect (Wald Chi-Square=10.6210; df=1, $p = 0.0006$) favoring the AM versus the TAU condition (hazard ratio for AM versus TAU was 0.304 (95% CI = 0.149, 0.622)).

Conclusions: Our data suggest that patients with early phase schizophrenia being treated with LAI antipsychotics who have better baseline cognition may have more reduction in relapse risk from treatment than those with poorer baseline cognition. These results extend the findings of cognition moderating acute response to antipsychotic treatment to suggest that cognition may also moderate antipsychotic treatment effects on relapse risk.

Keywords: Cognition, Long-Acting Injectable Antipsychotics, Recent Onset Psychosis, Relapse and Hospitalization

Disclosure: Teva Amylyx, C4 Innovations, Health Choice, Levo, Interactive Forums, PH Associates Limited: Consultant (Self)

P491. Sustained Treatment Response With Long-Term Valbenazine in Patients With Tardive Dyskinesia

Christoph Correll, Jean-Pierre Lindenmayer, Khodayar Farahmand, Scott Siegert, Eduardo Dunayevich*

Neurocrine Biosciences, Inc., San Diego, California, United States

Background: Valbenazine is a once-daily VMAT2 inhibitor approved for the treatment of tardive dyskinesia (TD), a persistent and potentially disabling movement disorder associated with prolonged exposure to antipsychotics, antiemetics, and other

dopamine receptor blocking agents. The efficacy, safety, and tolerability of valbenazine has been established in several phase 3 trials, including a long-term study (KINECT 4 [NCT02405091]) in which participants received open-label valbenazine (40 or 80 mg) for 48 weeks. Post hoc analyses of KINECT 4 data were conducted to assess patterns of treatment response.

Methods: Data from KINECT 4 treatment completers (participants who reached the Week 48 visit and had the longest duration of treatment) were analyzed post hoc. TD was assessed using the Abnormal Involuntary Movement Scale (AIMS) total score (sum of items 1-7, as rated by the study investigator), the Clinical Global Impression of Change-Tardive Dyskinesia (CGI-TD), and the Patient Global Impression of Change (PGIC). Analyses were conducted at Week 8 (first study visit after the valbenazine dose-optimization period) and Week 48 using the following definitions of response: ≥50% and ≥70% improvement from baseline in AIMS total score; rating of "much improved" or "very much improved" (score ≤2) on the CGI-TD and PGIC.

Results: Of the 167 participants who entered KINECT 4, 103 (62%) were treatment completers and included for analysis. Of these 103 participants, 39% and 86% met the ≥50% AIMS response threshold at Weeks 8 and 48, respectively. The percentages of participants who met the highly rigorous AIMS ≥ 70% response threshold at Weeks 8 and 48 were 17% and 52%, respectively. Of the 40 participants with AIMS ≥ 50% total score improvement at Week 8, 95% also met this threshold at Week 48 ("sustained response"). Of the 63 participants with <50% AIMS improvement at Week 8, 81% achieved the ≥50% response threshold by end of treatment at Week 48. The proportion of participants meeting the threshold for CGI-TD response also increased over time, from 50% at Week 8 to 92% at Week 48. PGIC results were similar, with response rates of 53% and 88% at Weeks 8 and 48, respectively.

Conclusions: Post hoc analyses of data from a 48-week, open-label study of once-daily valbenazine showed that the proportion of participants meeting rigorous treatment response thresholds increased over time. By the end of treatment at Week 48, >80% of participants demonstrated robust improvements in TD, as assessed using the AIMS (≥ 50% improvement), CGI-TD (score ≤2), and PGIC (score ≤2).

Keywords: Tardive Dyskinesia, Valbenazine, Long-Term Treatment

Disclosure: Neurocrine Biosciences: Employee (Self)

P492. The Pharmacodynamic Effects of TAAR1 Agonist Ulotaront on Metabolic Biomarkers of Glucose, C-Peptide, and Insulin Following a Meal in Patients With Schizophrenia

Snezana Milanovic, Nina Dedic, Yu-Luan Chen, Kuangnan Xiong, Roberto Gomeni, Robert Lew, Gerald R. Galluppi, Seth Hopkins*, Kenneth S. Koblan

Sunovion Pharmaceuticals, Inc., Marlborough, Massachusetts, United States

Background: Obesity, dyslipidemia, hypertension, hyperglycemia, and non-alcoholic fatty liver disease (NFLD) are highly prevalent in schizophrenia due in large part to the propensity of the current class of treatments to cause these metabolic changes. Hyperglycemia, diabetes mellitus, dyslipidemia and weight gain are highlighted in the Warnings and Precautions sections of the FDA approved labels for the current class of drugs approved for the treatment of schizophrenia. Although each antipsychotic may have its own risk profile, possibly related to pharmacological effects at histamine, serotonin and dopamine receptor subtypes, the metabolic changes associated with all drugs in the current class inform treatment decisions, benefit/risk, and result in increased public health costs. Ulotaront (SEP-363856) is a trace amine-associated receptor 1 (TAAR1) and 5-HT1A agonist currently in Phase 3 clinical trials for the treatment of schizophrenia. Recent preclinical evidence has identified TAAR1 as a novel regulator of metabolic control and a promising target for obesity and type 2 diabetes. Here we evaluated the effects of ulotaront on liquid metabolic biomarkers which were collected in Phase 1 clinical pharmacology studies.

Methods: Metabolic effects of ulotaront were examined in response to a meal following a 8-12 h fast. In a study to determine the effect of ulotaront on QTc interval (NCT04369391), schizophrenia subjects (N = 60) were randomized in a 3-way crossover design to receive single doses of ulotaront, placebo and moxifloxacin. Separately, in a study to evaluate ulotaront for a drug-drug interaction (NCT04865835), schizophrenia subjects (N = 25) were randomized in a 2-way crossover design to receive single doses of ulotaront or placebo. Plasma samples were analyzed for C-peptide, insulin, and glucose.

Results: Following administration of a meal, ulotaront lowered insulin and C-peptide levels compared to placebo, indicating an effect of ulotaront on glycemic control in response to feeding, with large (0.8 – 1.0) effect sizes on insulin and C-peptide levels. An integrated population PK/PD model jointly described insulin, C-peptide and glucose change, in response to a meal, as a function of ulotaront plasma concentrations.

Conclusions: The effects of ulotaront on metabolic markers, derived from plasma samples collected in the course of clinical pharmacology studies, suggest that the beneficial effects observed in animal models may translate to humans. Phase 1 clinical studies are currently ongoing to test the direct effects of ulotaront on metabolic parameters in schizophrenia patients (NCT05402111, NCT05463770). The healthcare burden of hyperglycemia, diabetes mellitus, dyslipidemia, weight gain and NAFLD in the treatment of schizophrenia with the current antipsychotic class would be avoidable with the advent of a new pharmacological class demonstrating benefit on these metabolic parameters.

Keywords: TAAR1, Ulotaront, Metabolic Biomarker, Schizophrenia Novel Treatment

Disclosure: Sunovion Pharmaceuticals, Inc.: Employee (Self)

P493. Gender Differences in the Relationship Between Trauma Exposure and Symptoms in First Episode Psychosis

Julie McCarthy*, Jacqueline Dow, Kim Mueser

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

Background: A history of trauma is common in people with psychotic disorders, and it is associated with more severe symptoms. Men and women with psychosis often experience differences in their course of illness and symptom profile. Past studies have found gender differences in trauma among those with first episode psychosis, but data in this area are limited. The current project aims to examine the relationship between trauma exposure and baseline quality of life and symptom profiles by gender, as well as the association of trauma exposure and change in 12- and 24-month quality of life and symptom profiles by gender.

Methods: Included in this secondary data analysis were n = 404 participants from the NIMH Recovery After an Initial Episode of Schizophrenia-Early Treatment Program (RAISE-ETP) study, which examined the effects of the NAVIGATE treatment group compared to the community care (CC) control group. The sample consisted of participants with schizophrenia, schizoaffective disorder, or other non-affective psychosis diagnoses. Trauma exposure was

measured using an abbreviated version of the Traumatic Life Events Questionnaire (TLEQ); outcome measures were the Quality-of-Life Scale (QLS), Calgary Depression Scale for Schizophrenia (CDSS), and the Wallwork five-factor model for the Positive and Negative Syndrome Scale (PANSS). At baseline, individuals with TLEQ data were included for analyses ($n = 400$; $n = 291$ men, $n = 109$ women); at follow-up, subjects were included with baseline and 12-month data ($n = 252$; $n = 185$ men, $n = 67$ women) and baseline and 24-month data ($n = 205$; $n = 151$ men, $n = 51$ women). Spearman correlations were examined between baseline trauma exposure and baseline symptom and quality of life measures (QLS, CDSS, PANSS total and PANSS factor scores). Reported correlations survived Bonferroni correction (.05/16 tests = .003). Linear mixed effects models of baseline trauma exposure on change in QLS, CDSS, and PANSS outcomes were examined at 12 and 24 months. Covariates included treatment group, study site, low or high duration of untreated psychosis, substance use disorder history, and baseline outcome of interest. Parallel analyses were conducted by gender.

Results: There was a positive correlation at baseline between trauma exposure and depression (CDSS) for men ($r = .18$, $p = .0019$) and women ($r = .28$, $p = .0027$). Positive correlations were also found at baseline between trauma exposure and PANSS depression/anxiety factor in men ($r = .25$, $p < .001$), and PANSS excited factor in women ($r = .30$, $p = .002$). A negative correlation was found at baseline between trauma exposure and PANSS negative factor for men only ($r = -.20$, $p = .005$). In the mixed model analyses, trauma exposure was associated with less decrease in the PANSS excited factor ($p = .038$) at 12-months and the PANSS total ($p = .019$) and positive symptom factor ($p = .002$) at 24-months in women; the remaining analyses were not significant.

Conclusions: Results suggest that gender differences in the relationship between trauma exposure and symptoms for individuals with first episode psychosis depend on the timeframe of assessment. At baseline, trauma was related to depression on the CDSS in men and women, and the depression/anxiety factor on the PANSS for men. Depression/anxiety on the PANSS was also related to trauma in women ($r = .19$ vs $r = .25$ in men), but it did not survive Bonferroni correction partly due to smaller sample size. So, the association between trauma and depression was a relatively consistent finding across gender. However, during the first years of treatment, trauma exposure may have a prolonged impact on total, positive, and excited symptom severity especially for women in that they may experience less benefit than men. These results indicate that further clinical interventions may be indicated in the consideration of lifetime trauma exposure by gender for those experiencing their first episode of psychosis.

Supported by: K23DA050808

Keywords: First Episode Psychosis, Gender Differences, Trauma Exposure

Disclosure: Nothing to disclose.

P494. The Effects of Lesion Network Guided Transcranial Electrical Stimulation on Symptoms, Behavior, and Electrophysiology in Patients With Psychosis: An Open Label Clinical Trial

Paulo Lizano*, Nicolas Raymond, Robert Reinhart, Matcheri Keshavan

Beth Israel Deaconess Medical Center, Harvard Medical School, Boston, Massachusetts, United States

Background: Transcranial electrical stimulation (tES) has been shown to improve psychotic symptoms and cognition in psychosis spectrum disorders. However, few investigations have used novel tES approaches, such as high definition tES (HD-tES) to more specifically target brain circuits. Convergent data from neuroimaging, neurophysiological, and cause-effect studies points towards the extrastriate visual cortex (V5/MT) as being causally linked to visual hallucinations and delta frequency deficits being associated with motion processing alterations in psychosis. We aimed to determine if causal lesion network guided HD-tES to the extrastriate cortex is efficacious and safe in improving psychosis symptoms, motion processing, and steady state visual evoked response potentials (ssVEP). Secondary outcomes include determining HD-tES effects on emotion processing evoked potentials, cognition, and depression symptoms.

Methods: To examine the functional role of the extrastriate visual cortex a between-participants, open-label, non-randomized, cross-over design, pilot study at a single site was performed to characterize the efficacy and safety of using cathodal HD-tDCS (transcranial direct current stimulation) or delta frequency HD-tACS (transcranial alternating current stimulation) in psychosis (ClinicalTrials.gov Identifier: NCT04870710). Enrolled participants were allocated to 20 mins of tES twice daily for 5 consecutive days applied bilaterally to the extrastriate visual cortex with one month of wash out between tES conditions. Clinical (psychosis and depression symptoms), cognitive (BACS), ssVEP and emotion processing evoked potential assessments (IAPS) were performed at baseline, day 5 and day 30.

Results: A total of 6 patients with a psychosis spectrum disorder were enrolled. Six individuals received cathodal HD-tDCS with 5 of them completing all follow-up visits. Three individuals received delta frequency HD-tACS and also completed all follow up assessments. HD-tDCS resulted in a significant reduction in psychosis general symptoms and emotion processing evoked potentials at 5 days, which were directly correlated. HD-tACS resulted in longer term reductions in psychosis general symptoms at 30 days, as well as cognitive improvements. Both type of tES were well tolerated and there were no adverse effects reported.

Conclusions: The findings herein provide proof of concept evidence that causal lesion network mapping can be used to enhance brain targeting using tES in psychosis. We showed that bilateral stimulation of the extrastriate visual cortex resulted in short term effects with HD-tDCS, while HD-tACS resulted in longer term benefits on general psychosis symptoms. Further research is needed in in a larger and more acute psychosis population to determine the clinical efficacy and safety of this type of stimulation.

Keywords: Psychosis, Transcranial Current Stimulation, Cognition, EEG, ERP

Disclosure: Nothing to disclose.

P495. A Phase 2, Randomized, Double-Blind, Placebo-Controlled Study of BIIB104 in Cognitive Impairment Associated With Schizophrenia (CIAS): The Tally Study

Alexander Coppell*, Michael Serenko, Michael Rooney, Jonathan Smith, Donna Babiar, Estibaliz Arce, Christopher L. Shaffer, Michael F. Green

Biogen Inc, Maidenhead, United Kingdom

Background: Certain cognitive impairments in schizophrenia are hypothesized to result from dysfunction in NMDAR glutamatergic neurotransmission. BIIB104 (formerly PF-04958242), a potent and highly selective high-impact AMPA receptor (AMPAR) positive allosteric modulator (PAM), has potential to enhance AMPAR activation and hence augment NMDAR-induced synaptic potentiation. Both preclinical (Shaffer et al., 2015; Shaffer 2018) and early clinical (Evans et al., 2016; Ranganathan et al., 2017) studies

suggest the potential efficacy of BIIB104 for cognitive impairment associated with schizophrenia (CIAS).

Methods: TALLY (263CS201, NCT03745820, EudraCT 2018-003825-27) was conducted in the USA, Japan, Spain, Germany, and the UK. Eligible participants were 18-55 years old with a DSM-5 diagnosis of schizophrenia of at least 2 years and selection criteria conformed to the Measurement and Treatment Research to Improve Cognition in Schizophrenia (MATRICS) recommendations (with additional criteria). Participants were randomized to receive oral doses (BID) of BIIB104 (0.15 mg or 0.5 mg) or placebo in a 1:1:1 ratio for 12 weeks. Dose selection was intended to permit the evaluation of both phasic and tonic dosing regimens relative to the preclinically predicted clinically efficacious plasma concentration.

The primary endpoint was the change from baseline in MATRICS Consensus Cognitive Battery (MCCB) Working Memory (WM) Domain T-score to Week 12. The University of California, San Diego Performance-Based Skills Assessment–Brief (UPSA-B), Schizophrenia Cognition Rating Scale (SCoRS) total interviewer score, MCCB Neurocognitive Composite (NCC), and Positive and Negative Syndrome Scale (PANSS) change from baseline to Week 12 were assessed as secondary endpoints. Safety and pharmacokinetics (PK) were also assessed. A standardized effect size of 0.35, which was considered a reasonable clinically meaningful treatment effect, was targeted.

Results: 195 participants were randomized from 52 centers; mean age was 39.8 years (range 21-55 years), 136 (69.7%) were male and 155 (79.5%) completed the 12-week treatment period. Demographic characteristics were well-matched across treatment groups.

PK sampling demonstrated consistent and expected drug plasma exposures for both BIIB104 doses over the entire 12-week evaluation period. Dosing compliance exceeded 95% across all treatment groups. Analyses of the primary endpoint showed no statistically significant improvement for BIIB104 (0.15 mg or 0.5 mg) versus placebo at Week 12 ($p = 0.85$ and $p = 0.81$, respectively). Least squares (LS) mean differences from placebo in MCCB WM were -0.26 (95% confidence interval [CI] -2.88, 2.37) and -0.33 (95% CI -2.94, 2.29) for BIIB104 at 0.15 mg and 0.5 mg, respectively. Analyses of the secondary endpoints also showed no statistically significant improvements for either BIIB104 dose across MCCB NCC (LS mean difference from placebo -1.10 [95% CI -3.14, 0.94], $p = 0.29$ and 0.49 [95% CI -1.55, 2.53], $p = 0.63$); UPSA-B (LS mean difference from placebo 3.43 [95% CI -0.20, 7.06], $p = 0.06$ and 3.42 [95% CI -0.23, 7.06], $p = 0.07$); or SCoRS (LS mean difference from placebo 0.43 [95% CI -1.43, 2.29], $p = 0.65$ and -0.43 [95% CI -2.28, 1.42], $p = 0.65$), for BIIB104 at 0.15 mg and 0.5 mg, respectively. There was no worsening or improvement of PANSS total score. BIIB104 was generally well tolerated.

Conclusions: BIIB104 did not show statistically significant efficacy in the treatment of CIAS at either tested dose. The hypothesis that the high-impact AMPAR PAM BIIB104 would demonstrate clinically meaningful efficacy in the treatment of CIAS was not supported.

Keywords: BIIB104, PF-04958242, Schizophrenia, Cognitive Impairment, AMPA

Disclosure: Biogen Inc.: Employee (Self), Biogen Inc.: Stock / Equity (Self)

P496. Longitudinal Ecological Momentary Assessments of the Behavioral Indicators of Avolition in Schizophrenia Identify Changes that Are Correlated With Clinical Ratings of Negative Symptoms

Philip Harvey*, Colin Sauder, Soumya Chaturvedi, Stephen Targum

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

Background: Reductions in emotional experiences in subjects with schizophrenia (avolition, asociality, and anhedonia) are associated with the real-world social deficits experienced by these individuals. The assessment of negative symptoms is more difficult than assessment of positive symptoms because it requires self-awareness and corroborative information from knowledgeable informants. Recent advances in the observational assessment of negative symptoms have employed participant reports using ecological momentary assessment (EMA) which can capture critical behavioral features of avolition and asociality, including the amount of time spent home and alone, and engagement in passive and unproductive activities. We report interim, but substantive pilot data from the first half of a 12-month longitudinal study of EMA, combined with clinical ratings of negative symptoms during an unblinded open label safety study of an antipsychotic medication in development.

Methods: Stable outpatients (PANSS ≤ 80) with schizophrenia entered a 12-month open-label treatment study of the candidate drug that included monthly clinical ratings of the PANSS and negative symptom ratings with the NSA-16. Longitudinal EMA assessments were delivered in 7-day bursts, 3 surveys per day for one week intervals monthly throughout the study. The EMA surveys were delivered by a smartphone and queried location and social context (home vs. away; alone vs. with someone), positive and negative affect (PA, NA), hallucinations and delusions, and 1 of 3 targeted activity surveys that were customized for home alone, home with someone, and away from home. A total of 23 different activities were sampled across the three surveys with a sampling window of "the last hour". Participants also wore a "fitbit" actigraph daily during EMA sampling weeks. Three individual NSA items were selected for analysis because they most closely defined avolition (reduced activities, reduced sense of purpose, and reduced social drive). Because this is an exploratory assessment of EMA, the analyses were limited to subjects who answered at least 33% of the EMA surveys. Mixed Model Repeated-Measures Analysis of Variance (MMRM) strategies were used for EMA data analysis, including use of dynamic correlates to predict activity outcomes. Correlations (and regression analyses) with aggregated EMA variables and scores on the NSA items were also computed.

Results: A total of 4138 fully completed EMA surveys have been answered to date by 54 subjects with completed clinical assessments during the sampling period. Scores on momentary PA as measured by EMA increased significantly ($X2 = 47.30$, $p < .001$). Step counts increased in concert with momentary PA ($p < .001$). The increases in PA also correlated with significant increases in the time-synchronized proportion of at-home productive activities and decreases in passive and unproductive activities. Further, significant decreases in the proportion of surveys answered at home were associated with increases in momentary PA ($p < .001$). Regression analyses indicated that more EMA surveys answered while at home, alone, and engaging in unproductive activities shared 21% of the variance with higher the NSA "reduced activity" item scores. More away from home activities, fewer EMA surveys answered home and alone, and more daily steps shared 31% of the variance with lower ratings of the NSA "reduced sense of purpose" item. Fewer EMA surveys answered home and alone and more surveys answered while engaging in productive activities shared 29% of the variance in the NSA item ratings of "reduced social drive". In contrast to these consistent correlations with negative symptom items rated with the NSA, scores on the PANSS reduced emotional experience items manifested much smaller correlations with EMA-derived variables. Numbers of surveys answered at home and alone, and

PA all shared less than 4% variance with PANSS reduced emotional experience items. The only correlation with PANSS reduced emotional experience that was substantial was the correlation between lower reduced emotional experience scores and more away from home activities (r = -.40).

Conclusions: These preliminary findings are the first report to document that treatment related changes in a constellation of the behavioral indicators of avolition can be detected with EMA surveys. The results of these surveys were substantially correlated with clinician ratings of NSA items indexing avolition (i.e., reduced emotional experience) that are the primary predictors of the social deficits associated with schizophrenia. Increases in PA after baseline were robustly detected and predicted increased engagement in positive daily activities measured on a momentary basis over a 6-month period, including more surveys answered away from home that corresponded with more productive activities and reductions in at-home unproductive activities. Increased physical activity was also longitudinally associated with increased PA. These EMA findings were validated by their convergence with clinician-rated scores on negative symptoms generated with a "gold standard" instrument (NSA-16) using a clinician-based, monthly rating strategy.

Keywords: Schizophrenia; Technology, Ecological Momentary Assessment, Schizophrenia Negative Symptoms

Disclosure: Allermes, Bioexcel, Boehringer-Ingelheim, Karuna Therapeutics, Merck, Minerva Pharma, Sunovion Pharma, WCG Clinical: Royalties (Self), EMA Wellness: Consultant (Self), i-Function, Inc.: Owner (Self)

P497. Metformin and Lorcaserin Combination Treatment for Antipsychotic-Associated Weight Gain

Lars Jarskog*, Thomas Stroup, Kai Xia, Anastasia Ivanova, Kim Brownley, Joseph McEvoy, Robert Millet

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

Background: Weight gain is a common and serious side effect of antipsychotic treatment and there are few effective options to mitigate this risk. Metformin has been identified as the most effective adjunctive treatment for antipsychotic-associated weight gain, with meta-analyses demonstrating ~3 kg weight loss in studies of 12-24 weeks duration. Lorcaserin, a 5-HT2C agonist, is an effective weight loss agent in obese but otherwise healthy individuals. Some combination drug approaches for weight loss using agents with different mechanisms have demonstrated promise over monotherapy treatment but this approach had not been studied for antipsychotic-associated weight gain.

Methods: This 52-week double-blind, randomized trial compared lorcaserin/metformin (LOR/MET, 10 mg BID/1,000 mg BID, n = 23) combination treatment, lorcaserin (LOR, 10 mg BID, n = 24) monotherapy and placebo (PBO, n = 24) in 71 non-diabetic outpatients with schizophrenia and schizoaffective d/o with a body mass index (BMI) > 27. All subjects also received a behavioral intervention aimed at improving diet and increasing physical activity. The primary outcome measure was change in weight at study endpoint compared to baseline. Secondary outcomes included change in measures of glucose and lipid metabolism.

Results: LOR/MET combination treatment was associated with significantly greater weight loss compared to placebo, with an estimated least-squares mean change from baseline to 52 weeks of -8.4 kg (p = 0.010). The effect of LOR monotherapy on weight was not significantly different compared to placebo at 52 weeks (-0.7 kg, p = 0.817). Of the secondary outcome measures of lipid and glucose metabolism, only fasting glucose in the LOR/MET arm was significantly reduced compared to placebo by -10.1 mg/dL (p = 0.030). Neither hemoglobin A1c nor any of the markers of lipid metabolism differed significantly from placebo for LOR/MET or LOR arms. Treatments were generally well tolerated, with somewhat higher rates of diarrhea, headache, dizziness, restlessness and diaphoresis in the treatment arms compared to placebo. Notably, the study was prematurely terminated on 02/13/2020 due to withdrawal of FDA approval for lorcaserin due to evidence of a small excess in cancer incidence following long-term lorcaserin treatment (median 3.3 years) in a large Phase 4 study aimed at assessing cardiovascular safety and efficacy. There were no new cases of cancer in the current study.

Conclusions: Combination treatment with LOR/MET was effective and generally well-tolerated in reducing body weight in outpatients with schizophrenia or schizoaffective disorder who were on stable antipsychotic regimens. The magnitude of weight loss for LOR/MET combination treatment was nearly 3-fold greater compared to weight loss seen for metformin monotherapy from prior studies in the same population. Among secondary outcome measures, LOR/MET appeared to improve fasting glucose levels, but hemoglobin A1c was unchanged as were markers of lipid metabolism. LOR monotherapy did not show efficacy at 52 weeks in primary or secondary outcomes. Two important limitations of the study include the current inability to use lorcaserin in clinical practice as well as reduced statistical power due to premature termination of the study. While lorcaserin is no longer available to prescribe for weight loss in the US, these data suggest viability of the 5-HT2C agonist mechanism as a generally well-tolerated and effective adjunct when used in combination with metformin to achieve clinically meaningful weight loss for antipsychotic-associated weight gain. The development of novel 5-HT2C agonists for weight loss represents a potentially important approach for addressing antipsychotic-associated weight gain. More generally, the findings also highlight the potential benefits of combining agents with distinct mechanisms for addressing antipsychotic-associated weight gain.

Keywords: Metformin, Lorcaserin, Antipsychotic Induced Weight Gain

Disclosure: Boehringer Ingelheim: Contracted Research (Self), Corcept Pharmaceuticals: Contracted Research (Self), UpToDate: Royalties (Self), SignantHealth: Consultant (Self), Otsuka: Contracted Research (Self)

P498. Cerebellar TMS Dosing Reveals Cerebellar-Cortical and Cerebellar-Basal Ganglia Connectivity

Mark Halko*, Roscoe Brady, Laura Holsen, Jing Xie, Adam Beerman, Won Young Cheong, Ivy Lee, Uzma Nawaz, Emily Payne, Emma Joncas, Madelaine Nye, Michael Esterman, Simon Laganiere

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

Background: Cerebellar stimulation has a renewed interest for the treatment of psychiatric and neurological conditions. Recent discoveries of motor function within the cerebellum have made understanding the mechanics of cerebellar stimulation imperative. Critically, we have previously demonstrated that cerebellar stimulation impacts network connectivity at rest in acute studies (Halko et al 2014) and across multiple stimulation sessions (Brady et al 2019), showing promise for reduction of symptoms of schizophrenia (Brady et al 2019). Here, we examined if dosage of TMS can be titrated by network connectivity.

Methods: In a repeated measures study, 26 healthy participants received 3 different intensities of intermittent theta-burst simulation determined by active motor threshold (100% aMT, 87.5%

aMT, 75% aMT). Stimulation was targeted at the cerebellar vermis representation of the dorsal attention network, with a standard figure of 8 coil, handle facing upward. Functional connectivity during a continuous performance task (gradCPT) was collected before and immediately after stimulation.

Results: Increasing cerebellar connectivity between default network regions of the cerebellum and default network regions of cortex following stimulation at 100% AMT regions of cortex was observed, but did not reach significance, when compared to 87% and 75% conditions ($p = .21$). However, cortical network connectivity change between default network and dorsal attention network was strongly linked to attentional performance improvement ($r = -0.225$, $p = 0.002$). Preliminary analysis suggests that cerebellar-basal ganglia connectivity may be intensity dependent ($r = 0.19$, $p = 0.07$).

Conclusions: Overall, our study indicates that titrating the dose of TMS based on motor threshold is an ineffective strategy for cerebellar modulation. We also demonstrate that connectivity change is a required component of behavioral change, suggesting that parameters that can deterministically impact connectivity at the targeted site of intervention may be more relevant considerations for determining effective TMS intensity dosages.

Keywords: Theta Burst Transcranial Magnetic Stimulation, Cerebellum, Basal Ganglia, Non-Invasive Brain Stimulation

Disclosure: Nothing to disclose.

P499. Cognitive Flexibility is Differently Linked to Callosal Fibre-Bundles and Functional Connectivity Between Homotopic Grey Matter Regions in Antipsychotic-Naïve Patients with First Episode Schizophrenia

Tina Kristensen*, Karen Ambrosen, Jayachandra Raghava, Egill Rostrup, Warda Syeda, Thijs Dhollander, Cecilie Lemvigh, Kirsten Borup Bojesen, Simon Anhøj, Birgitte Fagerlund, Mette Nielsen, Christos Pantelis, Birte Glenthoj, Bjørn Ebdrup

Center for Neuropsychiatric Schizophrenia Research (CNSR), Glostrup, Denmark

Background: Cognitive impairments are a core feature of schizophrenia. Abnormalities in the neuronal connectivity of the brain have been proposed as an essential biomarker of schizophrenia. Higher cognitive processes, such as executive functions, rely on an interplay between several distributed regionally based processes, and have been investigated in conjunction with a range of neuroimaging modalities. Typically, each biomarker has been analysed separately. However, due to the interconnected nature of brain processing, changes in one modality may modulate changes in other modalities. Little is known about how the structural connectivity (white matter (WM) fiber bundles connecting grey matter (GM) regions) and functional connectivity (concurrent functional activity between GM regions) associate in parallel to higher order cognitive functioning. Nonetheless, it has become increasingly clear that combining multi-modal brain imaging data is able to provide more detailed information about the neurobiological underpinnings of higher order cognitive processes.

Fixel-based analysis (FBA) is a novel statistical framework to analyse fibre-specific measures of WM, such as fibre density (FD) and fibre-bundle cross-section (FC). We used FBA and resting state functional MRI to investigate associations between WM micro- and macrostructure, functional connectivity and executive functions in 64 antipsychotic-naïve patients with first-episode schizophrenia and 95 matched healthy controls.

Methods: All participants underwent MRI (diffusion weighted imaging and resting state functional MRI). Executive functioning was assessed using tests from the Cambridge Neuropsychological Test Automated Battery (cognitive flexibility (IED) and planning (SOC); and the Brief Assessment of Cognition in Schizophrenia (verbal working memory (digit sequencing) and verbal fluency). Group comparisons and associations between fixel-based measures and executive functions were analysed using multivariate linear regression. Post hoc, homotopic connectivity was estimated as the correlation between the functional activity of corresponding grey matter regions of interest (ROIs) in each hemisphere connected via the isthmus, namely the left and right isthmus cingulate, precuneus, postcentral, and superior parietal regions, respectively. Associations between the homotopic connectivity between each ROI pair and cognitive flexibility, as well as to fixel-based measures were tested using partial correlation analyses. Analyses were covaried for age, sex, cohort, intracranial volume, 6 motion regressors, and the relative motion during the fMRI acquisition as appropriate, and results were corrected for multiple comparisons using FWE with a threshold of $p < 0.05$. There was an equal distribution of male/female sex in patients and matched controls (52% males, 48% females)

Results: Patients displayed cognitive impairments on all executive functions ($p < 0.001$) and reduced FD in the body of corpus callosum and cingulum($p < 0.05$) compared to controls. When modelling FC, we found a significant interaction between group and cognitive flexibility in the isthmus of corpus callosum ($p < 0.05$), i.e., larger callosal fibre-bundles of the isthmus was linked to increased cognitive flexibility in patients, but not in controls. We did not identify any significant interaction effect with planning, verbal working memory, or verbal fluency. Furthermore, when post hoc modelling homotopic connectivity, we identified a significant interaction effect between group and cognitive flexibility in the precuneus and postcentral ROIs. In patients, increased homotopic connectivity in the precuneus was associated with more cognitive flexibility ($p = 0.028$), while in controls, increased homotopic connectivity in the postcentral region was associated with more cognitive flexibility ($p = 0.012$). We did not identify any significant interactions between fixel-based measures and homotopic connectivity, but a trend level negative correlation between FC in isthmus and homotopic connectivity between postcentral ROIs in controls ($p = 0.08$, RHO -0.19), but not in patients ($p = 0.86$, $RHO = -0.02$).

Conclusions: Cognitive flexibility is linked to macroscopical characteristics of the fibre bundle cross-section of the isthmus of corpus callosum and is differentially associated with homotopic connectivity in the precuneus and postcentral GM regions in antipsychotic naïve patients with first-episode schizophrenia compared to controls. These group specific links may reflect that the structural and functional neurobiological underpinnings of cognitive flexibility in antipsychotic-naïve patients with first episode schizophrenia differs from controls.

Keywords: Antipsychotic-Naïve First-Episode Schizophrenia, Executive Function, Diffusion Weighted Imaging, Resting State Functional Connectivity

Disclosure: Nothing to disclose.

P500. Neurocognitive and Sensorimotor Biomarkers of Psychosis-Risk in 22q.11.2 Deletion Syndrome and Their Relationship to Clinical Symptoms

David Parker*, Sid Imes, Brett Henshey, Nicholas Massa, Bruce Cuthbert, Grace Lee, Elaine Walker, Erica Duncan, Joseph Cubells, Opal Ousley

Emory University, Athens, Georgia, United States

Background: The 22q.11.2 deletion syndrome (22q11DS) is one of the most robust genetic predictors of the development of psychosis and other psychiatric illnesses. We aimed to untangle

the complex relationship between 22q11DS and mental illness by utilizing a specific battery of cognitive tests and psychophysiological biomarkers known to be associated with psychosis-risk. In this study we examined performance on neurocognitive tests, motor speed, target detection, and auditory EEG responses in 22q11DS individuals and controls to understand the relationship between cognition, sensorimotor responses, and clinical symptoms.

Methods: We recruited 15 22q11DS individuals (Mean age=30, M/F = 9/6) and 19 healthy controls (HCs; Mean age=34, M/F = 5/14) from the local community. Each individual completed the MATRICS Consensus Cognitive Battery (MCCB), the Wechsler Abbreviated Scale of Intelligence, Second edition (WASI-II) Verbal IQ subtests, and the computerized Wisconsin Card Sorting Task (WCST). Sensorimotor reactivity was measured via a finger-tapping task (i.e., both dominant and non-dominant hands) and a visual oddball target detection task. For the auditory EEG task, each participant completed the "Double-Deviant" target detection paradigm, which presents a pseudorandom sequence of frequent standard tones and infrequent deviant tones (i.e., 90% of trials, 633 Hz, 50 ms duration; 10% of trials, 1000 Hz, 100 ms; respectively). Mismatch negativity (MMN) metrics were generated from this assessment. Current symptoms were assessed with the Structured Interview for Psychosis-Risk Syndromes (SIPS), including subscales that assess prodromal symptoms of schizophrenia (i.e., Positive, Negative, Disorganized, and General).

Data analysis: Group comparisons were examined for all measures. Welch's t-tests were completed for all age/sex adjusted standardized neurocognitive variables. One-Way ANOVAs were completed to examine sensorimotor and EEG results, with sex entered as a separate factor and age entered as a covariate for non-normed data (sensorimotor and EEG results). All p-values were false discovery rate (FDR)-adjusted. We tested for correlations between significant variables and clinical measures from the SIPS Subscales.

Results: Significant group differences were found in 8 of the 9 neurocognitive measurements (FDR-adjusted p's< 0.02, average Cohen's d = 1.62, average observed power= 0.91) indicating widespread cognitive deficits in 22q11DS subjects across multiple domains. Significant differences were found for the dominant and non-dominant hands on the motor finger tapping task (Cohen's d = [0.558, 0.716], average observed power: 0.68). Group differences were not found for accuracy or latency of detecting a visual target (FDR-adjusted p's>0.16). The Double-Deviant MMN ERP response was significantly smaller in absolute magnitude in the 22q11DS group (FDR-adjusted p = 0.048, Cohen's d = -0.864, observed power= 0.58). The MMN ERPs for the frequency and duration deviants were not significantly different (FDR-adjusted p's> 0.33). No group by sex interactions were observed in any of the measures.

Two neurocognitive variables and the finger-tapping for the non-dominant hand were significantly negatively correlated with the SIPS Positive symptoms (i.e., MCCB Working Memory, MCCB Verbal Learning, finger tapping non-dominant hand; Spearman's rho = -0.473, -0.403, -0.396, respectively; FDR-adjusted p's< 0.049). These results indicated that poorer neurocognitive and motor speed performance were associated with greater SIPS symptoms. Verbal IQ and each of the MCCB cognitive domains were negatively associated with Negative and Disorganized symptom scores on the SIPS (i.e., Spearman's rho range = -0.436 to -0.71, FDR-adjusted p's< 0.049), indicating that greater prodromal symptoms were associated with poorer cognition. Lastly, MCCB Reasoning and Problem-Solving score was negatively associated with the SIPS General Symptom score (Spearman's rho = -0.486, FDR-adjusted p = 0.015), indicating that more severe General symptoms were associated with poorer cognitive performance.

Conclusions: Identification and validation of psychosis-risk biomarkers in 22q11DS could provide important translational targets for future clinical trials for individuals with 22q11DS and other individuals at-risk for psychosis syndromes. Through the use of a dense battery of cognitive and psychophysiological biomarkers, these preliminary results indicate robust deficits across cognitive, motor, and auditory neural processing domains in 22q11DS that associate with clinical symptoms of the schizophrenia prodrome. Future work will build upon and attempt to verify the findings seen in this preliminary study. We plan to compare these data to individuals who are at clinical high-risk for psychosis to untangle genetic versus idiopathic risk. Furthermore, our current work in progress is examining deficits in pluripotent stem cell-derived neurons from 22q11DS and control subjects to understand abnormalities at the cellular level that could contribute to 22q11DS pathology.

Keywords: 22q11.2 Deletion Syndrome, Psychosis-Risk, Biomarkers, Cognition, EEG

Disclosure: Nothing to disclose.

P501. Astrocyte Subtype Gene Enrichment in Psychiatric Disorders and Psychotropic Medication Datasets

Sinead O'Donovan*, Xiaolu Zhang, Alyssa Wolfinger, Xiaojun Wu, Anna Lundh, Vladimir Parpura, Robert McCullumsmith, Rammohan Shukla

University of Toledo, Toledo, Ohio, United States

Background: Astrocytes have many important functions in the brain, but their roles in psychiatric disorders and their responses to psychotropic medications are still being elucidated. Reactive astrogliosis describes a spectrum of heterogeneous changes in astrocyte gene expression, morphology, and overall function, which can have a protective or pathological effect, depending on the subtype of astrocyte involved and the type of injury sustained. Different subtypes of astrocyte are classified based on their cellular morphology, location and primary functions, and can have potentially different responses to insult. Our understanding of the different types of astrocytes in humans and their roles in psychiatric disorders is still limited.

Methods: Gene and pathway enrichment of astrocyte subtypes, psychiatric disease and psychotropic medication gene-sets was assessed using hypergeometric overlap analysis with a background of 21,196 genes (GeneOverlap R v1.26.0). A density index (DI) was applied to quantitatively summarize how common or unique a biological theme is across different astrocyte subtypes and disease and drug gene-sets. Gene-sets were derived from publicly available transcriptomic datasets and gene-set size was restricted to between 10 to 500 significant ($p < 0.05$) differentially expressed genes. Confirmation studies included qPCR assays of astrocyte marker gene expression in chronic haloperidol- and vehicle- treated rat brain (n = 20), and exploratory kinome analysis (Pamgene12) of gliosome fraction isolated from postmortem brain tissue.

Results: Bioinformatic analysis identified gene enrichment [–log10($p < 0.05$)] of different astrocyte subtypes in psychiatric disorders. The highest level of enrichment (DI = 0.6) was found in schizophrenia. Astrocyte subtypes were differentially enriched in specific biological processes, including protein phosphorylation (DI = 0.25). Enrichment of protein phosphorylation in astrocytes was confirmed by increased (FC 1.22) kinome signal intensity in gliosome fraction relative to total homogenate. Common gene enrichment of different psychotropic medications and astrocyte subtypes was limited. qPCR analysis also found little effect of psychotropic medication (haloperidol-decanoate, Student's t-test, $p > 0.05$) on common astrocyte marker gene expression.

Conclusions: Gene enrichment analyses suggest enrichment of astrocyte subtype-specific genes in psychiatric disorders like

schizophrenia, indicating unique roles for different astrocyte subtypes in these disorders. Psychotropic medication and astrocyte gene enrichment are low suggesting that astrocytes are not significant targets of these medications. Overall, this study provides a unique view of astrocyte subtypes and the effect of medications on astrocytes in disease.

Keywords: Astrocyte, Schizophrenia (SCZ), Major Depression Disorder, Psychotropic Medications, Phosphorylation

Disclosure: Nothing to disclose.

P502. Assessing the Roles of Prefrontal Excitatory and Parvalbumin-Expressing Neurons During Working Memory

Tyler Dexter*, Megan Attard, Daniel Palmer, Lisa Saksida, Tim Bussey

University of Western Ontario, London, Canada

Background: Working memory (WM) is the process by which information is temporarily held available to aid goal-directed behavior. The prefrontal cortex (PFC) is a major modulator of WM function, and PFC disruption is known to impair WM processes across species. Synchronous neuron firing at the gamma frequency (gamma oscillations) in the PFC has been shown to increase alongside WM task difficulty, which has been implicated in stabilizing information in WM. Previous studies have demonstrated that inhibitory GABAergic neurons expressing the protein parvalbumin (PV) are critical for facilitating gamma oscillations in the cortex. Additionally, specific disruption of this population has been shown to impair PFC functions such as attention and cognitive flexibility. However, studies in mice have produced conflicting findings as to whether prefrontal PV neurons are necessary for WM task performance. Here, we combined in vivo optogenetics with a touchscreen-based paradigm of WM to 1) assess how prefrontal excitatory neurons contribute to maintaining WM over a delay period; and 2) assess how prefrontal PV neurons contribute to WM processes.

Methods: We used in vivo optogenetics to inactivate either prefrontal excitatory or inhibitory PV neurons during the trial unique non-match to location (TUNL) task. The TUNL paradigm contains three phases, where mice are presented a stimulus on the screen, they must retain the location of this stimulus over a delay, and then correctly chose a novel stimulus when it is presented alongside the original stimulus. All mice were trained on the TUNL task prior to surgery. To inactivate excitatory neurons, we injected male C57BL/6 mice ($n = 9$) bilaterally with a CAMKii-dependent archaerhodopsin (Arch) virus targeted to the medial PFC (mPFC; AP + 1.8, ML ± 0.3, DV -2.2). Two fiber optic probes were implanted above the injection sites. To inactivate PV neurons, male and female mice ($n = 7$ each) were bred to selectively express Arch under the PV promoter. These mice were then implanted with bilateral fiber optics at the same coordinates described above. Following surgery, mice were re-baselined on the TUNL task prior to optogenetic manipulations. To assess the role of mPFC excitatory neurons during WM maintenance, mice received optogenetic stimulation at temporally specific points of the delay (early phase of the delay, middle of the delay, or end of the delay). For these experiments, the delay period was held fixed at 2 s. For PV neuron inactivation experiments, WM maintenance was assessed by increasing the delay duration across separate sessions (0 s, 1 s, and 2 s). Additionally, to test whether PV neuron activity protected against proactive interference, mice were assessed on an increased interference probe. Here, the intertrial interval (normally 15 s) was removed, resulting in trials occurring back-to-back and increasing the potential for memories of previous trials to interfere with the current trial configuration.

Results: First, we observed that optogenetic inhibition of prefrontal excitatory neurons impairs task performance during specific temporal phases of the delay. Inhibition during the early and middle, but not late (choice phase), significantly impaired choice accuracy ($p = 0.002$, $F(2, 12) = 29.9342$). Next, two-way ANOVA revealed that inhibiting prefrontal PV neurons throughout the delay produced a significant delay-independent reduction in choice accuracy ($p = 0.025$, $F(2, 28) = 6.344$). Further, we observed a significant impairment when PV neurons were inactivated during the high interference condition ($p < 0.001$, $F(1, 28) = 19.625$). There were no significant effects of optogenetic stimulation under any conditions in a control group of PV:Arch- mice who underwent identical protocols ($n = 10$). Further, there were no significant effects of optogenetic stimulation on response or reward collection latencies under any conditions.

Conclusions: Our results describe the involvement of relevant PFC circuitry during a touchscreen-based task of WM designed to emulate paradigms used in human testing. We show that performance on TUNL not only depends on PFC excitatory neuron functioning, but that information held in WM is stable against PFC disruption by the choice phase. While multiple recent studies have evaluated the role of prefrontal PV neurons during WM paradigms in mice, the results have been inconsistent as to whether this population is necessary for optimal task performance. With TUNL, we are able to present sample and choice stimuli across multiple locations on the touchscreen. Therefore, this paradigm provides a high number of stimuli configurations, which differs from common two-choice working memory tasks. As such, TUNL may introduce a greater degree of interfering information on a given trial, as there may be lingering memories of past trial configurations that could disrupt the current information held in WM. PV neurons are critical for the generation and maintenance of gamma oscillations, which have been implicated in holding information in WM and protecting against competing task-irrelevant information. Therefore, our data suggests that prefrontal PV neurons facilitate the maintenance of WM information during TUNL, potentially by protecting against distracting information that interferences with the target held in WM.

Keywords: Visuospatial Working Memory, Medial Prefrontal Cortex, Parvalbumin Neurons, Touchscreen Cognitive Testing

Disclosure: Nothing to disclose.

P503. Impaired Face Emotion Recognition in Schizophrenia: Task Performance, Eye-Tracking and Neurophysiological Analyses and Comparison to Autism Spectrum Disorder

Antigona Martinez*, Elisa Dias, Pamela Butler, Russel Tobe, Daniel Javitt

Nathan S Kline Institute for Psychiatric Research, Orangeburg, New York, United States

Background: A key component of human social behavior is orientation to faces in general and eyes in particular during social interaction. In both schizophrenia (Sz) and autism spectrum disorder (ASD), deficits in social cognition have been attributed to reduced orientation to eyes while processing faces. Nevertheless, the mechanisms underlying reduced eye fixation remain incompletely understood. In the present study we performed a fine-grain analysis of fixation patterns during face viewing in Sz and ASD participants, along with electrophysiological recordings of event-related potentials (ERPs). The study builds from prior studies in significant ways. First, it includes both Sz and ASD subjects, permitting direct comparison of mechanisms underlying social cognitive deficits across the two disorders. Second, it uses incongruent as well as congruent face stimuli to better clarify the role of eye versus mouth fixation in face-emotion recognition

(FER) deficits across the disorders. Third, it assesses fixation patterns relative to responses on a trial-by-trial basis in order to assess cause and effect relationships between alterations in gaze and alterations in behavior. Finally, it builds from recent studies showing differential patterns of early visual processing alterations in Sz and ASD compared to neurotypical (NT) individuals to assess potential neurophysiological contributions to altered behavior. To our knowledge, this is the first study to directly compare fixation patterns related to FER across Sz and ASD groups, to utilize trail-by-trial fixation information, and to combine these with neurophysiological analyses tied both to stimulus and fixation onset.

Methods: Participants were 23 patients diagnosed with Sz (mean age 37.1 years), 22 ASD individuals (29.2 years) and 24 demographically-matched NT subjects (36.0 years). Participants were presented with 324 human face stimuli and reported the perceived emotion in each (happiness, sadness, anger, fear or neutral). The face stimuli were either congruent (top and bottom halves of faces conveyed the same emotion) or incongruent (top and bottom halves conveyed different emotions). Eye movements (gaze position) were recorded on each trial and the ongoing electroencephalogram (EEG) was recorded from 64 scalp channels.

Results: As expected, the percent correct performance for congruent stimuli differed significantly across groups ($p = .01$). Post-hoc differences between Sz ($p = .004$) and ASD ($p = .027$) versus NT subjects were also significant. The severity of FER deficits was similar in Sz and ASD participants ($p = .20$), however, despite the much different level of overall socioeconomic function.

Relative to NT subjects, both Sz ($p < .001$) and ASD ($p < .001$) participants showed reduced fixation on eyes and increased fixation on mouths and were more likely than NT individuals to use mouth-, rather than eye-, information to disambiguate chimeric faces (SZ: $p < .001$; ASD: $p = .006$). However, whereas ASD individuals correctly identified faces in trials where they did fixate on eyes, Sz subjects remained impaired despite their fixation location ($p = .002$).

In the time-domain, ERP responses elicited by all face stimuli consisted of a P1 followed by N170 potential over the ventral occipital scalp. Overall, the peak amplitude of the P1 and N70 was significantly different across groups ($p = .011$) and in Sz compared to NT ($p = .013$). Lastly, as in previous reports, the onset of the N170 was delayed in ASD relative to NT subjects ($p = .035$). When the spectral composition of EEG responses was assessed using time-frequency analyses, a significant across-group difference in evoked power in the theta (4-7 Hz) frequency band was observed ($p = .02$) with a significant post-hoc difference between Sz and NT individuals ($p = .016$). The onset of face stimuli also modulated ongoing oscillatory activity in the alpha (8-12 Hz) band with reduced modulation in Sz ($p = .024$) (relative to NT) but significantly enhanced modulation in ASD ($p = .026$) which additionally predicted their delayed N170 latencies ($p = .048$).

Conclusions: These findings show the potential value of the use of chimeric face stimuli, and, despite similar levels of overall dysfunction, our findings also point to significant mechanistic differences underlying FER deficits in Sz and ASD participants. In Sz patients, the deficit patterns are consistent with impaired magnocellular function and impaired bottom-up input to visual sensory systems. Deficits in ASD are consistent with hyperactivity of the retinocollicular system as indexed electrophysiologically by alpha-band activity. Abnormalities in stimulus-induced alpha modulation may also underlie the well-described N170 latency increases in ASD and may thus serve as an additional sensitive biomarker of early face-processing dysfunction in ASD.

Keywords: Schizophrenia (SCZ), Autistic Spectrum Disorders, Face Emotion Processing, EEG Electrophysiology, Eye-Tracking

Disclosure: Nothing to disclose.

P504. Brain and Physiological Stress Responses in Early Psychosis

Brandee Feola*, Elizabeth Flook, Dongju Seo, Victoria Fox, Jesse Oler, Neil Woodward, Stephan Heckers, Jennifer Blackford

Vanderbilt University Medical Center, Nashville, Tennessee, United States

Background: Stress is proposed to contribute to the onset and expression of psychosis. Yet, how people with psychosis differ in responses to stressors remains unclear, especially in early-stage psychosis. The current study examined whether brain or physiological stress responses differ in people with early-stage psychosis.

Methods: 20 people with early psychosis (schizophrenia spectrum disorders/bipolar disorder with psychotic features) and 20 healthy control participants completed a fMRI stress task that involved viewing stress images, neutral relaxing images, and fixation baselines. Additional assessments included physiological stress responses (cortisol, heart rate), self-report of stress, and psychosis symptoms (PANSS). Region of Interest (ROI) analyses were conducted for a prior stress network (amygdala, hippocampus, striatum, hypothalamus, prefrontal cortex). Linear mixed models compared differences in ROI activation using group (psychosis/control), phase (baseline, provocation), and emotion (stress, neutral). Correlations were conducted between brain activation, stress responses, and clinical characteristics.

Results: During the stress task, significant group differences in brain activation were found for the hippocampus (group x phase x emotion interaction $p < 0.001$) and prefrontal cortex (group x phase interaction $p = 0.04$). Within the psychosis group, activation in the hippocampus, amygdala, and prefrontal cortex correlated with physiological stress responses and clinical characteristics (all $p < 0.05$).

Conclusions: Our findings suggest that people with early psychosis have altered brain responses to stress. Within the psychosis group, brain responses to stress correlated with clinical characteristics. Future studies utilizing multi-method assessment of stress may provide the most insight into the role of stress in early psychosis.

Keywords: Acute Stress, Early Psychosis, Brain Imaging, fMRI

Disclosure: Nothing to disclose.

P505. Functional Imaging and Genetic Predictors of Future Employment in Patients With Schizophrenia Performing a Working Memory Task

Rachael Blackman*, Daniel Eisenberg, Dwight Dickinson, Michael Gregory, Karen Berman

Clinical and Translational Neuroscience Branch, National Institute of Mental Health, Intramural Research Program, Bethesda, Maryland, United States

Background: With a dearth of treatment options, cognitive deficits in patients with schizophrenia are well known to be linked to real-life functional outcomes in patient populations. There is great heterogeneity in this regard; some individuals become active members of the workforce despite their illness, while others do not have this resilience. Uncovering predictors of positive outcomes may inform the development of better treatment targets for patients with schizophrenia. Deficits in working memory have been well characterized in patients, for example using the N-Back task which activates frontal networks during task performance. Here we examine whether neural

activation during this task may predict whether patients are employed or not at a future timepoint. In addition, as schizophrenia is a highly heritable illness, we test whether overall genetic risk moderates these effects of cognition on future employment.

Methods: Patients with schizophrenia spectrum illness (n = 125, 31.2% female, mean age 32.5 + /- 10.5 years) participated in a research study at the National Institute of Mental Health Intramural Research Program's Clinical Center and were recontacted at a later timepoint (8.7 + /- 4.0 years after initial visit) when outcomes information such as employment status (employed vs. unemployed) was collected. At the initial visits, patients participated in neuropsychological testing, functional neuroimaging (3 T fMRI during N-Back performance), and provided blood samples for genotyping. Cognitive measures including g (a general measure of cognition), IQ, and six cognitive subdomain scores (verbal memory, N-Back working memory, visual memory, processing speed, card sorting/executive function, and span working memory) calculated at the initial visit were tested as predictors of employment status at follow-up using logistic regressions. Covariates in the models included age at the initial visit, sex, length of time to follow-up, and initial visit employment status. In a subset of participants (n = 102, 28.4% female, mean age 32.9 + /- 10.9 years), polygenic scores for schizophrenia risk (PGSscz) were derived using summary statistics from genome wide association studies. Logistic regressions including PGSscz and the first three ancestry-related genetic principal components as covariates were added to the models and repeated. Finally, for the subset of participants who performed the N-Back working memory task during fMRI scanning at their initial visit (n = 54, 31.5% female, mean age 33.2 + /- 11.6 years), neural activation during task performance (2-Back > 0-Back) was tested using AFNI software to identify regions that differed between the individuals who were employed vs. unemployed at follow-up. To further focus on a sub-group who may benefit from targeted intervention, we repeated the fMRI analyses restricting the analyses to only those participants who were unemployed at the initial visit (n = 37) to characterize regions that predict employment status change (i.e., unemployed to employed) at follow-up.

Results: 44.8% of the total participants (n = 56) were employed at follow-up, with 23.2% of participants at the initial visit having an employment change such that they were unemployed at the initial visit and became employed by the follow-up timepoint. Of the cognitive domains examined at the initial visit, g (n = 115, B = 0.61, SE = 0.30, Wald=4.08, p = 0.043), IQ (n = 116, B-0.06, SE = 0.02, Wald=7.07, p = 0.008), and verbal memory (n = 115, B = 0.43, SE = 0.21, Wald=4.23, p = 0.040) predicted employment status at follow-up such that higher scores were observed in those who were employed compared to those who were unemployed. We further found that PGSscz moderated the effect of N-Back behavioral scores at the initial visit on the ability to predict employment status at the later timepoint (n = 83, interaction N-Back score by PGSscz p = 0.022). Participants who were unemployed at follow-up and had higher PGSscz had worse performance at the initial visit, whereas the reverse was true in the employed population. Greater neural activation in right dorsolateral prefrontal cortex during the N-Back task (peak p = 0.002, uncorrected) and right cingulate cortex (peak p = 0.003, uncorrected) was observed for employed individuals at follow-up. Results from the analysis examining only participants unemployed at the initial visit were consistent.

Conclusions: The results of our study demonstrate that in addition to general cognitive predictors of future employment there are other subdomains that may be relevant, specifically verbal and working memory. The N-Back behavioral effects were evident when considering PGSscz, highlighting the need to consider the heterogeneity of the schizophrenia patient population. The fMRI findings suggest avenues for further investigation into the neural underpinnings of real-world outcome measures in this population.

Keywords: Cognitive Impairment Associated With Schizophrenia, Functional Outcomes, Polygenic Risk Score, Functional Neuroimaging

Disclosure: Nothing to disclose.

P506. The Impact of Early Risk Factors on Cognition in Children, Adolescents, and Adults With First-Episode Psychosis

Cecilie Lemvigh*, Birgitte Fagerlund, Merete Osler, Jens Richardt Møllegaard Jepsen, Mette Nielsen, Jacob Rydkjaer, Kirsten Borup Bojesen, Christos Pantelis, Birte Glenthoj, Anne Katrine Pagsberg, Bjørn Ebdrup

Center for Neuropsychiatric Schizophrenia Research (CNSR), Mental Health Center, Glostrup, Copenhagen University Hospital, Valby, Denmark

Background: Schizophrenia is associated with widespread cognitive deficits that often precede the onset of psychosis by several years. Twin studies have demonstrated that cognitive functions are heritable and show genetic overlap with schizophrenia risk. Moreover, several early risk factors for schizophrenia have been identified, and some studies report and association between low birth weight and impaired cognition. Nevertheless, the potential additive or interactive impact of multiple early risk factors on cognition and the relation to age of illness onset remains unclear.

Methods: Clinical cohorts of patients with first-episode psychosis (FEP) and matched healthy controls (HC) with comparable cognitive measures (recruited from 1998 – 2017) were combined and linked with information from the medical birth registry. The final sample consisted of 608 participants, including 166 children and adolescents with FEP and 148 matched HCs (age 9-17), and 146 adults with FEP and 148 HCs (age 18-45). From the medical birth registry, we included gestational age, Apgar score, birth weight and length, parental age, maternal smoking, and winter/spring birth. Specific cognitive functions were assessed at illness onset using the Brief Assessment of Cognition in Schizophrenia, and intelligence (IQ) was estimated using selected subtests from the Wechsler child or adult intelligence scales. Multiple linear regression was used to explore if the early risk factors significantly predicted cognitive performance.

Results: FEP patients performed worse than HCs on all cognitive measures (all p's < .001). In the whole sample, children and adolescents performed worse than adults on verbal memory, working memory, planning, fluency and motor speed (all p's < .001). Among the early risk factors, FEP patients only displayed significantly lower birth length (p = .007) and birth weight (p = .032). Linear regression with IQ as the dependent variable revealed gestational age (p = .006), birth weight (p = .047) and group (FEP vs HC) as significant independent variables, while birth length, Apgar score, paternal age, winter birth and age (child vs adult) did not contribute significantly (adjusted R2 for the model = 0.164). Only birth weight (p = .043), group (p < .001) and age (p = .015) were significant for processing speed (adjusted R2 = 0.162). We observed no interaction effects between the significant early risk factors and group (FEP vs HC) or age (child vs adult). Including sex in the models did not change the findings. The remaining cognitive measures were not significantly influenced by the included early risk factors. Only a small subsample (N = 85) including both FEP patients and HCs had information on maternal smoking status, but here individuals with a positive history of maternal smoking during pregnancy showed significant

impairments in processing speed ($p = .003$), working memory ($p < .001$), and planning ($p = .018$).

Conclusions: The FEP groups showed widespread cognitive deficits compared to HCs regardless of age of onset, consistent with the current evidence implicating cognitive deficits as a core feature of schizophrenia. However, the majority of the cognitive functions examined were not significantly influenced by the included early risk factors for schizophrenia. Only IQ and processing speed were significantly associated with gestational age and birth weight, although the observed effects were small. Low birth weight and premature birth has previously been associated with impaired cognition in both healthy individuals and schizophrenia subjects, yet the available literature is sparse. Exploratory analyses of maternal smoking status revealed a possible, but imprecisely measured association with lower cognition. More studies are needed to gain a better understanding of the complex processes leading to cognitive deficits in schizophrenia.

Keywords: Cognition, First Episode Psychosis, Neurodevelopment

Disclosure: Nothing to disclose.

P507. EEG Correlates of Aggression in Patients With Psychotic Disorders

Michael Murphy*, Ricardo Carrion, Jose Rubio, Anil Malhotra

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

Background: Active psychosis symptoms have been associated with an elevated acute risk of violence. However, most patients with active psychosis symptoms do not commit acts of violence. It is unclear why some patients commit violent acts and current prediction tools are insufficient. Identification of easily-measurable neurological correlates for aggression could help reduce the risk of harm to patients, caregivers, and clinicians. Electroencephalography (EEG) is a widely used and easily applied g technique that can provide millisecond-level resolution of neural activity. Literature suggests that the alpha rhythm is linked to cognitive ability and can be impaired in schizophrenia. We hypothesized that larger alpha abnormalities would be associated with more impulsivity and aggression. Since the alpha rhythm is the primary generator of EEG microstates, we hypothesized that microstate parameters would also be abnormal in patients with higher levels of aggression.

Methods: Eyes-closed resting EEG data was collected from 31 patients with psychotic disorders (18 males) and 18 age matched controls (7 males) using an EEG cap with 64 Ag-AgCl electrodes. Bad channels were spline-interpolated and data was re-referenced to average reference. For individual alpha peak frequency analysis (IAPF), we estimated the power spectral density using Welch's method with non-overlapping Hamming windows of 2 seconds in length and identified a peak in the power spectral density between 7 and 13 Hz. Microstate analysis was performed using CARTOOL. A k-means clustering algorithm was used to identify microstate topographies and a meta-criterion was used to select k. Participants completed the four questionnaires about aggression: the Barratt Impulsivity Scale (BIS), the Reactive Proactive Aggression Questionnaire (RPAQ), the Buss-Perry Scale (BPS), and the Urgency, Premeditation (lack of), Perseverance (lack of), Sensation Seeking, Positive Urgency, Impulsive Behavior Scale (UPPS-P). In addition, participants completed a questionnaire about their history of aggressive behavior, the Lifetime History of Aggression (LHA). P values from statistical tests were adjusted with the Dubey Armitage-Parmar method.

Results: Patients scored significantly higher on all aggression scales compared to controls (all $p < .037$). There were no statistically significant relationships between delta, theta, alpha, beta, or gamma power and any of the aggression scales. After correction for multiple comparisons, in the combined group, IAPF was inversely correlated with BPS and RPAQ total scores (both Spearman's $R = -.33$) and Hostility ($R = -.39$) and Physical Aggression ($R = -.33$) subscores of the BPS. These effects were stronger in women than in men. In patients only, IAPF was inversely correlated with RPAQ Proactive Aggression subscore ($R = -.39$). There were no statistically significant relationships between IAPF and LHA. We then repeated the analysis using only a single channel (FPz) and found that in patients there was a statistically significant relationship between IAPF and BPS. For the microstate analysis, the segmentation algorithm produced four canonical microstates that have been repeatedly demonstrated in the literature. There were no case-control differences in microstate duration, occurrence, or transition probabilities. In patients only, uncorrected exploratory analyses suggested that microstate A duration was associated with LHA, Antisocial Behavior, and Verbal Aggression ($R = .39, .37, .47$ respectively). Transitions between microstates A and B were inversely correlated with UPPS-P as well as subscales for Non-planning, Proactive Aggression, Perseveration, and Hostility (all $R > .37$). The overall deviation from a completely random transition matrix was inversely associated with Physical Aggression ($R = -.37$).

Conclusions: We used resting state EEG frequency and microstate analyses to identify correlates of aggression in controls and patients with schizophrenia. We found that IAPF was inversely correlated with self-assessed aggression but not lifetime history of aggression. We note that these results could be robustly demonstrated with a single EE electrode. We also found that transitions between canonical microstates A and B were correlated with aggression.

Keywords: Irritability/Aggression, Psychotic Disorders, Electroencephalography

Disclosure: Nothing to disclose.

P508. BDNF Levels as a Predictor of Cognitive Response to Antipsychotic Treatment. Preliminary Results

Rodrigo Nieto*, Hernan Silva, Manuel Kukuljan

Universidad de Chile, Santiago, Chile

Background: Cognitive symptoms are a central manifestation of schizophrenia and are significantly related to patients´ social functioning and quality of life. Several studies have linked Brain-Derived Neurotrophic Factor (BDNF) not only to the pathogenesis of schizophrenia, but also to neuronal plasticity, learning, and memory. Changes in BDNF levels have been related to improvements in cognition in schizophrenia patients during cognitive remediation. Some patients improve their cognitive function during pharmacological antipsychotic treatment, but some patients do not. A personalized medicine approach to help identify which patients will have a better cognitive outcome is necessary. We hypothesized that baseline BDNF levels could be related to cognitive response to antipsychotic treatment in schizophrenia patients.

Methods: Patients with schizophrenia ($n = 24$) were evaluated with Montreal Cognitive Assessment (MoCA). Those with cognitive deficit ($n = 18$) were reassessed after 6 months of treatment with atypical antipsychotics, and 61% of them ($n = 11$) improved enough to reach a normal MoCA score. Plasma and serum BDNF levels were measured with ELISA at baseline evaluation. We compared baseline plasma BDNF levels in two group of patients: those that improved cognition enough to reach a normal MoCA score vs those that did not. We also searched for correlations between changes in MoCA score and baseline BDNF levels.

Results: Baseline plasma BDNF levels showed a trend to be higher in the subgroup of patients that has cognitive deficit and that improve cognition enough to reach a normal MoCA score at follow-up evaluation, although this difference is not significant (U Mann Whitney; $p = 0.29$). A trend towards a positive correlation was found between baseline serum BDNF levels and improvement in MoCA score (Spearman Rho 0.32; $p = 0.19$).

Conclusions: The study of BDNF as a biomarker of cognitive improvement in patients with schizophrenia requires further research, with potential clinical implications for personalized medicine in the treatment of cognitive symptoms in schizophrenia patients.

Keywords: BDNF, Cognition, Schizophrenia

Disclosure: Nothing to disclose.

P509. Network Hub Strength as a Predictor of Working Memory Performance in Schizophrenia

Hamdi Eryilmaz*, Melissa Pax, Mark Vangel, Ibai Diez, Daphne Holt, Joan Camprodon, Jorge Sepulcre, Joshua Roffman

Massachusetts General Hospital, Charlestown, Massachusetts, United States

Background: Working memory impairment is a debilitating and treatment-resistant aspect of schizophrenia, which is also a strong predictor of functional outcome. Breakdown in brain network hubs, putatively related to altered neurodevelopment may underlie the cognitive deficits associated with this illness. However, our understanding of network hub characteristics in schizophrenia and their relationship to working memory impairment remain elusive.

Methods: Here, we used weighted degree, a robust graph theory metric representing the number of weighted connections to a node based on resting state functional connectivity. In doing so, we quantified node strength across 333 cortical hubs (based on Gordon et al. parcellation) in 29 patients with schizophrenia and 29 age- and gender-matched healthy controls (including both male and female subjects). We assessed the relationship between participants' behavioral performance (retrieval accuracy and reaction time) in a verbal working memory task (Sternberg Item Recognition Paradigm) and node strength of the regions across the cortex using a correlation analysis. We utilized bootstrapping and False Discovery Rate to correct for multiple comparisons and determine significance ($p < 0.025$, for two behavioral measures). In an additional analysis, we employed a connectome-based predictive modeling framework and leave-one-out cross-validation to test whether a summary degree score can be used to predict participants' working memory performance.

Results: In both patients and controls, elevated weighted degree in the default mode network (DMN) regions was generally associated with poorer performance (retrieval accuracy and reaction time, $p < 0.025$). Higher weighted degree in the ventral attention network (VAN) nodes along the right superior temporal cortex was associated with better performance (retrieval accuracy) in both groups ($p < 0.025$). Weighted degree in several prefrontal and parietal areas was also associated with behavioral performance only in patients ($p < 0.025$). In regions that are critical for sustained attention, these correlations were primarily driven by between-network connectivity in patients ($p < 0.05$). Finally, our cross-validated prediction analysis revealed that a linear model using a summary weighted degree score can be used to predict an individual's working memory performance in patients with schizophrenia ($r = 0.36$ for retrieval accuracy and 0.68 for reaction time).

Conclusions: Our results indicate that schizophrenia is associated with dysfunctional hubs across the cortical systems supporting internal and external cognition and highlight the importance of topological network analysis in the search of biomarkers for cognitive deficits in schizophrenia.

Keywords: Working Memory, Schizophrenia (SCZ), Graph Theory, Cognitive Outcome Prediction

Disclosure: Elsevier Inc.: Employee (Spouse)

P510. Mice Exhibit Distinct Behavioral Signatures While Executing a Multi-Alternative Spatial Working Memory Task in an Automated Radial Arm Maze

Joshua Taliaferro*, Julia Greenwald, Elizabeth Pekarskaya, Alexandra Tarasenko, Clay Lacefield, Christoph Kellendonk

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

Background: Working memory (WM) is the cognitive capacity for temporarily holding information in mind for processing or use and undergirds most of our actions and activities. Despite a rich history of WM investigation, the characterization of the neural basis of WM is still emerging. In rodents, unlike in nonhuman primates (NHPs), robust neural encoding of retrospective actions or stimuli —theoretically a quintessential WM characteristic—is not regularly observed, calling into question the congruity of neural WM mechanisms across species.

Spatial working memory in particular is usually tested in rodents using tasks with two spatial choices. One possibility is that such paradigms do not provide the spatial complexity and alternative choices needed to evoke explicit neural representations of retrospective actions, locations, or stimuli.

Methods: Adult mice ($n = 8$) were trained in a novel spatial working memory task in an automated, 8-arm radial arm maze with a large, open center. Inspired by multi-alternative NHP WM tasks, the task was designed to increase choice optionality, while minimizing the possibility of behavioral subversion of the working memory maintenance requirement, likely thus necessitating clearer neural representations of WM content for successful execution.

Results: Despite task difficulty, mice learn the task to proficiency, consistently operating well beyond chance. During task acquisition, mice develop distinct behavioral signatures, which help reveal the underlying cognitive frameworks mice are using to solve the task.

Conclusions: Here we present a novel spatial WM paradigm for freely moving mice in an automated radial arm maze. Even amidst variability in behavioral signatures, mice perform the task well, suggesting that this multi-alternative behavioral paradigm is a promising testing ground for a more complete elucidation of the neural basis of working memory.

Keywords: Working Memory, Behavioral Tasks, Behavioral Variability

Disclosure: Nothing to disclose.

P511. Increased Rostral Medial Frontal GABA in Early Psychosis is Obscured by Levels of Negative Affect

Molly Simmonite*, Beier Yao, Robert Welsh, Stephan Taylor

University of Michigan, Ann Arbor, Michigan, United States

Background: Evidence suggests dysfunction of gamma-aminobutyric acidergic (GABAergic) interneurons in psychosis, and prior research has linked GABAergic function with a tendency toward negative affective states and poor tolerance for stress. Magnetic resonance spectroscopy (MRS) studies measuring GABA

have yielded inconsistent findings. Here we investigate GABA concentrations in young adults with attenuated psychosis syndrome (APS) and first episode psychosis (FEP), as well as testing the hypothesis that negative affect is a clinical phenotype that is associated with reduced GABA, a relationship which may confound GABA measurement in psychosis.

Methods: MRS were obtained from 14 patients with FEP (9 men, 5 women), 7 patients with APS (5 men, 2 women) and 15 healthy controls (HC, 10 men, 5 women), using a MEGA-PRESS sequence on a 3 T Philips Ingenia scanner. Voxels were placed in rostral MFC and midline occipital cortex. Gannet 3.1 was used to determine GABA + and Glx (glutamate and glutamine combined) concentrations.

Results: We found a trend towards increased rostral MFC GABA + concentrations in FEP ($F_{2,28} = 3.04$, $p = .06$), but no group differences in the occipital cortex GABA + concentrations ($F_{2,32} = 0.49$, $p = .62$) or Glx concentrations were found (rostral MFC Glx: $F_{2,28} = 0.08$, $p = .93$; midline occipital Glx: $F_{2,32} = 0.34$, $p = .72$). When covarying for scores on the Psychological Stress Index, rostral MFC GABA + levels in FEP were significantly greater than APS and HC ($F_{2,28} = 4.26$, $p = .02$). Planned comparisons also revealed a trend towards increased rostral MFC GABA + in APS relative to HC ($t27 = 2.00$, $p = .0548$). No group differences in Glx (rostral MFC Glx: $F_{2,31} = 0.26$, $p = .77$; midline occipital Glx: $F_{2,32} = 1.03$, $p = .23$) or midline occipital GABA + were found ($F_{2,31} = 0.48$, $p = .62$).

Conclusions: These results, considered alongside previously published findings, suggest multiple factors influencing GABA + levels in psychosis. We conclude a process exists which drives up GABA + levels in early psychosis, alongside a separate process in which reduced GABA + is associated with increased negative affect. These multiple processes have resulted in contradictory findings in the literature, and their untangling is critical to the understanding of GABA + in psychosis.

Keywords: Magnetic Resonance Spectroscopy, Psychosis, GABA, Negative Affect

Disclosure: Nothing to disclose.

P512. Medial Prefrontal Cortex Dysfunction Mediates Working Memory Deficits in Patients With Schizophrenia

John Williams*, Zu Jie Zheng, Philip Tubiolo, Jacob Luceno, Roberto Gil, Ragy Girgis, Mark Slifstein, Anissa Abi-Dargham, Jared Van Snellenberg

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

Background: Schizophrenia (SCZ) is marked by deficits in working memory (WM), which are strongly predictive of poor functional outcomes. WM performance has been found to depend critically on the function of circuits in prefrontal cortex (PFC) and their modulation by dopamine (DA). Considerable evidence has implicated pathology of the PFC in the etiology of SCZ, including findings of dramatically reduced cortical DA release in SCZ. Reduced WM task-evoked deactivation in regions of the default mode network (DMN), including medial PFC (mPFC), has been shown to correlate with reduced task performance in both SCZ and HC. However, the mechanisms by which cortical hypodopaminergia may produce WM deficits in SCZ, including relationships with mPFC hypodeactivation, are unclear.

Methods: Patients with SCZ ($N = 41$) and HC ($N = 40$) performed a visual object n-back WM task across 4 functional magnetic resonance imaging (fMRI) runs, each comprising two 40-second 1-back and 2-back blocks. Performance was quantified as the proportion of correct 2-back trials, with the significance of group differences tested using a Mann-Whitney-Wilcoxon rank-sum test. In subject-level analysis, deactivation during 2-back blocks was quantified using generalized linear models in Statistical Parametric Mapping version 12 (SPM12). An mPFC region of interest (ROI) was produced from the Gordon (2016) parcellation by selecting DMN parcels in PFC, dilating each by 8 mm, uniting them, and eroding the result by 5 mm. Group-level analyses were conducted in Permutation Analysis of Linear Models (PALM), using threshold-free cluster enhancement (TFCE) and family-wise error rate correction. Within mPFC, we first isolated voxels significantly deactivating during 2-back blocks. We then localized 205 voxels (5.535 cm3) additionally showing associations between deactivation and performance, controlling for diagnosis. Mean mPFC deactivation was obtained for each participant by averaging across all 205 voxels; the significance of group differences was assessed by Welch's t-test. mPFC deactivation was evaluated as a mediator of the impact of diagnosis on task performance using the CANlab M3 Mediation Toolbox (significance evaluated by bias-accelerated and corrected bootstrap procedure with 10 million resamplings).

Prefrontal synaptic dopamine release was additionally quantified in a subset of 9 unmedicated patients with SCZ and 14 HC via two positron emission tomography (PET) scans with the DA D2/3 receptor antagonist [11 C]FLB457: a baseline scan, and a second scan 3 hours after oral administration of 0.5 mg/kg of amphetamine. For both PET scans, binding potential relative to the non-displaceable compartment (BPND) was quantified in ROIs drawn on each participant's T1-weighted anatomical magnetic resonance image. DA release capacity was quantified in medial frontal cortex (MFC) as the percentage change in BPND after amphetamine challenge (ΔBPND). mPFC deactivation was input as the dependent variable into a robust regression model including intercept, MFC ΔBPND, diagnosis, ΔBPND×Diagnosis interactions, and age.

Results: Relative to HC ($N = 41$), SCZ ($N = 40$) exhibited worse performance (median 2-back proportion correct, SCZ: 0.781, HC: 0.914, $p = 0.0015$). SCZ showed significantly reduced task-evoked mPFC subregion deactivation relative to HC ($p = 0.0351$). Mediation analysis revealed a significant mediated (indirect) effect ($p = 0.0431$) of diagnosis on WM task performance, and an unmediated (direct) effect with only trend level significance ($p = 0.0545$). The regression model for mPFC deactivation as a function of MFC ΔBPND (in 9 SCZ and 14 HC) included: Intercept ($B = -0.430$, $p = 0.0269$), Diagnosis ($B = 0.0703$, $p = 0.503$), ΔBPND ($B = -0.913$, $p = 0.0257$), ΔBPND×Diagnosis ($B = 0.888$, $p = 0.311$) and age ($B = 0.00308$, $p = 0.569$); greater release capacity was associated with reduced mPFC deactivation during the WM task. When evaluated in each group separately, MFC DA release capacity (ΔBPND) was associated with mPFC deactivation in HC ($B = -1.335$, $p = 0.0268$), but not SCZ ($B = -0.280$, $p = 0.543$).

Conclusions: Failure of task-evoked mPFC deactivation is a mediator of and potential substrate for WM impairment in SCZ. HC participants show an association between mPFC deactivation magnitude and DA release capacity, which is absent in SCZ. This suggests that DA deficit in PFC in SCZ is a potential mechanism underlying failure to deactivate mPFC, thus limiting WM performance. A better understanding of how DA deficit affects microcircuitry within this region would allow better targeted therapies for cognitive impairments in SCZ.

Keywords: Schizophrenia (SCZ), Working Memory, Functional Magnetic Resonance Imaging (fMRI), Task fMRI, Positron Emission Tomography (PET)

Disclosure: Nothing to disclose.

P513. Cerebellar Effects on Abnormal Psychomotor Function are Mediated by Processing Speed in Psychosis Spectrum

Alexandra Moussa-Tooks*, Anna Huang, Baxter Rogers, Julia Sheffield, Neil Woodward, Stephan Heckers

Vanderbilt University Medical Center, Nashville, Tennessee, United States

Background: Psychomotor disturbance has been identified as a key feature of psychotic disorders, with motor signs observed in upwards of 66% of unmedicated, first-episode patients. Aberrations in the cerebellum have been directly linked to sensorimotor processing deficits including processing speed, which may underly psychomotor disturbance in psychosis, though these brain-behavior-symptom relationships are unclear.

Methods: In a sample of 339 psychosis patients (242 schizophrenia-spectrum, 97 bipolar with psychotic features) and 217 controls, we evaluated the relationship between cerebellar grey matter volume in the Yeo sensorimotor network and processing speed (assessed via the Screen for Cognitive Impairment in Psychiatry [SCIP]). To further test the role of these variables in predicting psychomotor disturbance (mannerisms and posturing, retardation, excitement of the Positive and Negative Syndrome Scale [PANSS]), a mediation analysis was performed for an a priori model in the psychosis group: predictor=cerebellar volume; mediator=processing speed; outcome=psychomotor disturbance. Cerebellar volumes were calculated using optimized processing through the Spatially Unbiased Infratentorial (SUIT) toolbox. Statistical analyses were performed in R; all models included total intracranial volume, age, sex, and chlorpromazine equivalents as covariates.

Results: As expected, sensorimotor cerebellar volume was positively associated with processing speed in the whole sample ($t = 3.428$, $p < 0.001$, $d = 0.30$). Effects were present in both the control sample ($t = 2.345$, $p = 0.020$, $d = 0.32$) and the psychosis sample ($t = 2.188$, $p = 0.029$, $d = 0.24$). Next, we tested whether processing speed was a significant mediator between cerebellar volume and psychomotor aberrations. Within the psychosis group, no significant direct effect (cerebellar volume --> psychomotor disturbance) was observed in this mediation model (ß=-0.035, $p = 0.59$, $d = 0.06$). Rather, the indirect path (average combined effect: ß=-0.043, $p = 0.014$) showed a significant partial mediation by processing speed, with a small effect of cerebellum on processing speed (ß=0.172, $p = 0.029$, $d = 0.24$) and medium effect of processing speed on psychomotor disturbance (ß=-0.254, $p < 0.001$, $d = 0.60$).

Conclusions: The current findings suggest a critical role of cerebellar circuitry in a well-established sensorimotor aberration in psychosis (processing speed) and the presentation of related psychomotor phenotypes within psychosis. Due to the nature of the PANSS, our mediation model could not be evaluated in the control sample. Future work will employ more dimensional measures of psychomotor disturbance to capture normative and aberrant brain-behavior-symptom relationships. Future research may also determine the magnitude of these relationships within subtypes of psychosis (e.g., disorganized behavior or catatonia) as well as the specificity to motor, versus non-motor (e.g., cognitive, social), cerebellar functions.

Keywords: Psychomotor Symptoms, Cerebellum, Brain-Behaviour Relationships, Structural MRI, Processing Speed

Disclosure: Nothing to disclose.

P514. Prioritization of Potential Causative Genes for Schizophrenia in Placenta

Gianluca Ursini, Pasquale Di Carlo, Sreya Mukherjee*, Qiang Chen, Shizhong Han, Jiyoung Kim, Maya Deyssenroth, Carmen Marsit, Jia Chen, Ke Hao, Giovanna Punzi, Daniel Weinberger

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

Background: The placenta plays a critical role since very early phases of development, paving the path for brain development. Disruptions in placenta functioning may later result in neurodevelopmental psychiatric disorders, such as schizophrenia. Our prior findings show that genomic risk for schizophrenia converges with early-life complications (i.e., prenatal, intrapartum, and early postnatal complications, such as serious obstetric complications) and affects risk for the disorder, in a sex-biased fashion. Computational approaches such as Transcriptome Wide Association Studies (TWAS) and Summary data based Mendelian Randomization (SMR) have the ability to integrate summary statistics from GWAS data with expression mapping studies, to allow predicting gene expression in specific tissues for individuals in case-control cohorts. This makes possible to estimate, in larger cohorts, the association of predicted gene expression in a specific tissue with traits or diseases. Here, we use these methods to identify specific genes and potential mechanisms that, in placenta, may condition trajectories of risk that lead to schizophrenia.

Methods: We performed TWAS in healthy term placentae ($N = 147$) samples to derive candidate placental causal genes that were confirmed with SMR. To search for placenta and schizophrenia-specific associations, we performed an analogous analysis in fetal brain ($N = 166$) samples, and additional placenta TWAS for other disorders/traits. We also analyzed if there were differences in sex-biased placental processes relevant for brain development, as prior data suggest. Finally, we performed placental TWAS for other disorders and traits, to further support that the placental transcriptome is relatively specific to schizophrenia.

Results: The analyses in the whole sample and stratified by sex highlight 186 genes whose predicted expression in placenta is associated with schizophrenia (Bonferroni corrected p-value<0.05); 138 of these placenta and schizophrenia specific risk genes are SMR-validated, and many risk genes are sex-biased. The molecular mechanisms highlighted by gene ontology prediction converge on nutrient-sensing capabilities of placenta and trophoblast invasiveness. These genes do not show analogous effects on risk for other developmental disorders and traits and do not show a potentially causal role in similar analyses in fetal brain. This result suggests an etiological role in the risk for schizophrenia (versus other disorders of development) relatively specific of placenta (versus brain). Additionally, transcript-level analysis detects gene isoforms not linked with schizophrenia at the gene level, thus implicating splicing events in the placental transcriptome that do not seem biased by sex-related differences.

Conclusions: Placenta and brain might converge in contributing to early, though reversible, trajectories of risk for the disorder. However, most research on brain development and schizophrenia has been exclusively focused on brain, a strategy that may miss relevant discovery opportunities. In this study we show that the investigation of placental mechanisms of risk leads to the detection of potential causal genes for schizophrenia and points to novel candidate mechanisms that may inform new venues of for prevention.

Keywords: TWAS, Psychiatric Disorders, Schizophrenia (SCZ), Placenta, Mendelian Randomization

Disclosure: Nothing to disclose.

P515. Evaluating the Psychometric Reliability of Nonlinear Dynamics in Schizophrenia

Christopher Gonzalez*, Claudia Lainscsek, Juan Molina, Yash Joshi, Terrence Sejnowski, Neal Swerdlow, Gregory Light

Department of Veterans Affairs, San Diego, California, United States

Background: Schizophrenia (SZ) is a chronic psychiatric disorder that is highly heterogenous in clinical manifestations, and sensitivity to therapeutics such as medications or cognitive interventions. Identifying biomarkers that characterize treatment-sensitive subpopulations can match patients with treatments that improve outcomes. EEG measures such as event-related potentials (ERPs) serve as such biomarkers in SZ and demonstrate high psychometric reliability. In addition to classical ERPs, novel EEG measures of nonlinear dynamics (NLD) could similarly serve as biomarkers, though the reliability of these measures have not been assessed. This study applies a nonlinear signal processing technique, Delay Differential Analysis (DDA), to capture dynamical features inaccessible from traditional neurophysiological, linear techniques. Because brain processes are intrinsically nonlinear, DDA is well-suited to study both group differences between healthy controls and SZ patients, as well as individual differences within SZ. Here, we estimate the test-retest reliability of the NLD measure for the first time alongside traditional ERPs computed from the same trials.

Methods: This study is part of an ongoing clinical trial, with N = 24 to date (SZ = 20, healthy control=4, age=46.9, M/F 14/6). Each person is administered a passive auditory oddball task with EEG for two sessions, separated by 1 week, both prior to intervention. Traditional ERP analysis follows standard pre-processing pipelines, whereas DDA pre-processing is minimal. DDA calculates NLD features in a sliding window manner by fitting a three-term delay differential equation to each time window. The estimated coefficients from this model serve as NLD features. Model parameters were defined in a previous publication and were selected for classifying SZ from healthy control EEG dynamics. At each EEG session, amplitudes for Mismatch Negativity (MMN), P3a, and a peak in the NLD feature were extracted. Reliability metrics were calculated using a frontal electrode (Fz) from 150 randomly selected standards and deviants, with the same trials for both approaches. Reliability was assessed using Pearson correlations.

Results: MMN ($r = 0.75, t = 5.3(22), 95\%CI = 0.5\text{-}0.9, p < 0.001$) and P3a ($r = 0.74$, $t = 5.1(22)$, 95% CI = 0.47-0.88, $p < 0.001$) showed high reliability across a 1 week interval for all individuals. The NLD measure showed moderate reliability ($r = 0.52$, $t = 2.87(22)$, 95% CI = 0.15-0.76, $p < 0.01$).

Conclusions: MMN, P3a, and NLD exhibited significant test-retest reliability, though NLD reliability was lower than that of traditional ERP measures. Notably, this study modeled subsets of random trials and likely underestimates true reliability. Future studies are needed to determine whether comparable data pre-processing improves reliability for NLD and whether this measure is associated with demographic, clinical, cognitive and functional outcomes in SZ.

Keywords: Test-Retest Reliability, Auditory Mismatch Negativity, Nonlinear Analysis

Disclosure: Nothing to disclose.

P516. Normative Modelling of Brain Morphometry in Individuals at Clinical High-Risk for Psychosis From the ENIGMA CHR Working Group

Shalaila Haas*, Ruiyang Ge, René Kahn, Maria Jalbrzikowski, Dennis Hernaus, Cheryl Corcoran, Sophia Frangou

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

Background: Psychosis is characterized by substantial heterogeneity at the level of brain organization, cognitive dysfunction and clinical severity. However, group-level inferences derived from case-control differences is not representative of individual-level deviations. In this study, we used normative modeling of brain morphometry to investigate whether the normative deviations derived from harmonized MRI data from a large independent sample of healthy individuals could help inform associations with positive symptoms and general intellectual ability.

Methods: T1-weighted MRI brain scans were available for 1237 healthy controls (HC; 684 [55.3%] males; mean [SD] age = 20.75 [4.74])) and 1340 CHR (709 [52.9%] males; mean [SD] age = 22.32 [4.95]) with positive symptom data from 29 sites. Structural neuroimaging data was processed within each respective site using Freesurfer analysis software using standardized ENIGMA protocols to derive measures of cortical thickness (CT), surface area (SA), and subcortical volume (SV). The dataset underwent site effect correction using ComBat-GAM harmonization. Normative modeling of neuroanatomical data was generated using Fractional Polynomial Regression separately for males and females from an independent sample of 38,050 healthy individuals (53.3% female) aged 3-90 years pooled across 81 datasets and was used to compute individual-level statistically quantified deviations from the normative range by computing a z-score. Subsequently, we averaged the regional z-scores separately for each phenotype in each participant to generate an "average deviation score" for cortical thickness (ADS-CT), cortical surface area (ADS-SA), subcortical volume (ADS-SV), and across all measures to generate a global average deviation score (ADS-G). Mixed linear models including age were used to determine associations between ADS with positive symptoms and general intellectual ability in CHR, correcting for multiple comparisons using FDR correction. In posthoc follow-up analyses, we examined the possibility that associations between psychopathology and IQ are not linear but are restricted to most deviant groups in CHR individuals with ADS below the 5th percentile ("infranormal") and above the 95th ("supranormal") centiles for each of these metrics and compared them to those in between these percentiles. The three groups were compared in terms of positive symptoms and IQ using analysis of covariance with age as covariates.

Results: CHR individuals showed widespread decrements in regional mean deviation of cortical thickness, particularly in superior temporal and frontal regions. Regional mean deviations in surface area also indicated decrements with the exception of visual, sensory, and motor regions. Regional mean deviation in subcortical volumes showed pronounced decrements, especially in the thalamus, putamen, and hippocampus. Within the CHR group, there was a negative association between positive symptoms and ADS-SA ($\beta = -0.08$, P-FDR = 0.02) and positive associations between IQ with ADS-SA ($\beta = 0.08$, P-FDR = 0.03) and ADS-G ($\beta = 0.10$, P-FDR = 0.03). Comparison of CHR individuals with supranormal, average, and infranormal ADS-CT, ADS-SA, ADS-SV, ADS-G did not identify significant group differences at P-FDR < 0.05 in terms of positive symptoms and IQ. At a nominal level, CHR individuals with infranormal ADS-G had higher positive symptom scores ($F = 4.07$, $P = 0.01$) and a trend towards lower IQ ($F = 2.71$, $P = 0.07$) compared to those with average or supranormal ADS-G.

Conclusions: The present study identified marked neuroanatomical heterogeneity in CHR states based on the largest dataset of individual-level neuroimaging measures in CHR individuals using robust estimates of the normative range of regional cortical thickness, cortical surface area and subcortical volume. Moreover, we identified a subset of CHR individuals that showed reduced global brain morphometry, with corresponding higher positive symptom severity and lower IQ. These findings suggest normative modeling of brain morphometry may help identify individuals at extremes with both preserved and atypical brain morphometry which could help inform clinical and cognitive heterogeneity in other psychiatric disorders.

Keywords: Normative Modeling, Clinical High Risk for Psychosis, FreeSurfer, Clinical Heterogeneity, Stratification

Disclosure: Nothing to disclose.

P517. Computer-Vision Analysis of Craniofacial Dysmorphology in 22q11.2 Deletion Syndrome and Psychosis Spectrum Disorders

David Roalf*, Donna McDonald-Mcginn, Joelle Jee, Mckenna Krall, T. Blaine Crowley, Paul Moberg, Christian Kohler, Alice Bailey, Monica Calkins, Ian Campbell, Kelly Clark, Andrew Crow, Nicole Fleischer, R. Sean Gallagher, Virgil Gozenbach, Ruben Gur, Emily McClellan, Daniel McGinn, Arianna Mordy, Kosha Ruparel, Russell Shinohara, Bruce Turetsky, Lauren White, Elaine Zackai, Raquel Gur

University of Pennsylvania, Philadelphia, Pennsylvania, United States

Background: Minor physical anomalies (MPAs) are phenotypic abnormalities of aberrant development. MPAs are common in 22q11.2 deletion syndrome (22q11DS) and psychosis spectrum disorders (PS). MPAs in these disorders include abnormalities of the eyes, ears, mouth, and head. Since MPAs likely represent a disruption of early embryologic development, in-depth study of MPAs offers a promising entry point for better understanding the neurodevelopmental neuropathology associated with 22q11DS and PS.

Methods: 2D photographs of individuals using standard digital photography. Participants included 22q11DS (n = 150), PS (n = 55), and typically developing (TD; n = 93) subjects. Photographs were analyzed using two computer-vision techniques: 1) Facial Dysmorphology Novel Analysis (Face2Gene (F2G)) technology to identify the presence of genetically mediated facial disorders, and 2) Emotrics—a recently developed semi-automated machine learning technique that localizes facial landmarks and computes regional facial measurements.

Results: F2G detected 22q11DS in 99% of 22q11DS individuals. F2G scores were robust and significantly higher in 22q11DS as compared to PS or TD. PCA-derived factor loadings of all F2G scores indicated unique and overlapping facial patterns that were related to both 22q11DS and PS. Regional facial measurements of the eyes (marginal reflex distance) and nose (philtrum length) were shorter in 22q11DS as compared to TD, while PS showed an intermediate pattern.

Conclusions: These preliminary results indicate overlap in facial dysmorphogenesis between 22q11DS and PS. The extent to which these developmental facial markers are evident before the impairment or distress of sub-psychotic symptoms may allow us to identify at-risk youths more reliably and at an earlier stage of development.

Keywords: Psychosis-Risk, 22q11.2 Deletion Syndrome, Facial Dysmorphology

Disclosure: Nothing to disclose.

P518. Quantifying Speech and Language Disturbance in Schizophrenia With Neural Language Models

Yan Cong*, Amir Nikzad, Sameer Pradhan, Sunghye Cho, Katrin Hänsel, Aarush Mehta, Sarah Berretta, Leily Behbehani, Mark Liberman, Sunny Tang

Feinstein Institute for Medical Research, Glen Oaks, New York, United States

Background: Disorganized speech is a hallmark of schizophrenia spectrum disorders (SSD) and related psychotic disorders and represents complex language disturbances which cannot be easily captured on a semantic or syntactic level. Since their introduction in 2017, pre-trained neural language models (nLMs) continue achieving state-of-the-art results on a variety of natural language processing tasks. We hypothesized that nLMs are promising tools for automatically quantifying disorganized speech in SSD because nLMs are effective at encoding contextual content and more complex referential information. We tested multiple validated semantic similarity strategies to examine the extent to which nLMs can improve accuracy of detecting and characterizing different types of speech and language disturbances, especially incoherent and inefficient disorganized speech among people with SSD.

Methods: Healthy volunteers (HV) and participants with SSD were recruited from inpatient and outpatient facilities (SSD = 74, HV = 37). They were rated on the Scale for the Assessment of Thought Language and Communication (TLC) and two items from the Scale for the Assessment of Negative Symptoms (SANS; decreased vocal inflection and increased latency). Open-ended speech (162 ± 121 words) was recorded and underwent transcription and processing for cosine similarity of word- and sentence-level embeddings. Three levels of pre-processing were conducted incrementally. Level 1 included verbatim transcripts excluding non-speech verbalizations like laughter. Level 2 excluded disfluencies such as repetitions and filled pauses. Level 3 additionally excluded NLTK stop words. We adopted 3 primary strategies to measure semantic similarity, sometimes referred to as "semantic coherence" in other studies: (1) the word-to-word variability at K inter-word distances, with K ranging from 2 to 10 (K2:10); (2) average semantic similarity of each word in 5- or 10-words window (W5/10); (3) First Order similarity of consecutive phrase vectors and Second Order similarity between phrase separated by another intervening phrase (FO/SO). Vector embeddings were generated from a static model GloVe and 3 nLMs (BERT, T5, GPT3) to compute similarity. Median, inter-quartile range (IQR), 5th percentile (Q5), and 95th percentile (Q95) values were then derived. Welch's t-test were used to examine group diagnosis (SSD vs. HV). Significance was two-tailed with alpha=0.05.

Results: Using the K2:10 semantic similarity strategies, across all levels of pre-processing, GloVe showed statistically significant lower similarity in SSD than in HV for 7 measures with medium effect size (Cohen's d 0.5-0.8) in median, Q5, and Q95 similarity. BERT showed lower similarity in SSD for 9 measures with medium effect size, and for 46 measures with large effect size (Cohen's d >= 0.8) in median, Q95, and IQR values. T5 showed lower similarity in SSD for 43 measures with medium effect size, and for 37 measures with large effect size in median, Q95, and IQR values. In contrast, GPT3 showed higher similarity in SSD for 39 measures but lower IQR in SSD for 2 measures (Cohen's d >= 0.5). For each of the models, more extensive data pre-processing generally resulted in fewer significant results. Using the W5/10 semantic similarity strategies, GloVe showed statistically significant higher similarity in SSD for 1 measure (Cohen's d 0.41) with Level 1 pre-processing, and no significant results with more pre-processing. BERT showed lower similarity in SSD for 18 measures (Cohen's d >= 0.5), including 13 measures with large effect size. The results were consistent across pre-processing levels. T5 showed lower similarity in SSD for 15 measures (Cohen's d >= 0.5), including 6 measures with large effect size. In contrast, GPT3 showed higher similarity in SSD for 8 measures of median, Q5 and Q95 values but lower IQR in SSD for 3 measures (Cohen's d >= 0.5). For both T5 and GPT3, Level 3 pre-processing resulted in fewer significant effects. Using the FO/SO strategies, GloVe showed statistically significant lower similarity in SSD for SO Q95 (Cohen's d -0.77) and FO Q95 (Cohen's d -0.81) with Level 1 pre-processing, but higher similarity in SSD for FO Q5 (Cohen's d 0.43) with Level 2, and null results for Level 3. BERT showed higher similarity in SSD for 4 measures (Cohen's d 0.2-0.5), and T5 for 2 measures with medium effects. The results improved with more extensive pre-processing for both BERT and T5. GPT3 showed higher similarity in SSD for 6

measures (Cohen's d 0.5-0.8), including 2 measures with large effect size. The results were mostly consistent across levels of preprocessing.

Conclusions: We evaluated three different types of nLMs to quantify speech and language disturbances in SSD, using static embeddings from GloVe as baseline. nLMs were, in general, more sensitive than GloVe in identifying language disturbances in SSD, reflecting that nLMs are better tools for studying SSD. Preprocessing by removing disfluencies did not greatly alter the results but removing stop words masked the SSD group difference in some cases, particularly for GPT3. GloVe, BERT and T5 all generally show lower similarity in SSD, across analysis frameworks, while GPT3 mostly shows higher similarity in similar tests. We hypothesized that the differences can be explained by how nLMs are pre-trained and what data they are trained on. Future directions include evaluating the robustness of language measures generated from nLMs across speech tasks and samples, and finetuning nLMs with domain specific data.

Keywords: Schizophrenia Spectrum Disorders, Natural Language Processing (NLP), Digital Phenotyping, Computational Psychiatry, Biomarkers

Disclosure: Nothing to disclose.

P519. Cognitive Flexibility and Psychotic Symptoms: A Computational Analysis of Learning in a Dynamic Environment

James Waltz*, Adam Culbreth, Pantelis Leptourgos, Sonia Bansal, Philip Corlett, Molly Erickson, James Gold

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

Background: While deficits in reinforcement learning (RL) and decision-making (DM) in psychosis are well-established, it is not clear precisely which aspects of RL and DM relate to which psychotic symptoms. We have previously shown that the ability to adaptively modulate learning rates dynamically, around contingency shifts, relates to negative symptoms in schizophrenia. However, we have also found that rates of switching in response to positive and negative outcomes also predict positive symptom severity. We hypothesized that, in a large sample of schizophrenia patients (SZs) and non-clinical voice-hearers (VHs), the ability to adaptively modulate learning rates in a dynamic learning environment would distinguish SZs and VHs from healthy controls (HCs), and that measures of the ability to adaptively modulate learning rates would relate systematically to the severity of positive symptoms.

Methods: We administered 68 schizophrenia patients, 33 non-clinical voice-hearers, and 47 healthy controls an RL task where they chose from three decks of cards and were probabilistically reinforced for their choices. After the participant reached a criterion, the contingencies changed, with a new deck becoming the best. Participants achieved as many stages as possible in 240 trials. We quantified overall switch rates, as well as switch rates after rewards and punishments. Furthermore, we used a computational model to estimate learning rates (α), on a trial-wise basis, as a function of the slope of prediction errors. We estimated the impact of uncertainty (operationalized through the slope of prediction errors) on learning rate change, on a subject-wise basis. We used ANOVAs with post-hoc tests to examine differences between participant groups, and we used correlation analyses to investigate relationships between learning measures and symptom severity. We also examined differences between subgroups of SZ patients, based on their duration of illness (less than 3 years vs. greater than or equal to 3 years).

Results: We found that switch rates for SZs early in their illness course ($n = 19$; $p < 0.001$) and non-clinical VHs ($p = 0.028$) were significantly greater than those of controls, and that uncertainty had a greater impact on learning rate change in both SZs ($p = 0.029$) and VHs ($p = 0.003$) than in HCs. However, more paranoid SZs ($p = 0.002$) and VHs ($p = 0.008$) showed lower mean learning rates (α) under greater uncertainty, and SZs and VHs with more severe overall psychosis ($p = 0.024$) showed less learning rate modulation around a contingency shift.

Conclusions: These results provide further evidence that people with clinical psychosis are less able to adaptively use uncertainty to modulate the impact of feedback on learning and decision-making. Furthermore, these results indicate that this form of cognitive flexibility may be disrupted in people with unusual percepts and beliefs who do not seek help. Consistent with prior findings, these results suggest a particularly close relationship between learning rate and psychotic symptoms, and paranoia, in particular. Further researcher is needed to examine whether abnormalities in learning rate modulation in schizophrenia patients and non-clinical voice-hearers have the same or different neural substrates.

The research was supported by NIH Grant R01 MH112887 (PIs: J. Gold, P. Corlett).

Keywords: Schizophrenia (SCZ), Delusions, Decision Making, Reinforcement Learning Modelling, Uncertainty

Disclosure: Nothing to disclose.

P520. A Functional Connectome-Based Neural Signature for Individualized Prediction of Antipsychotic Response in First-Episode Psychosis

Hengyi Cao*, Todd Lencz, Juan Gallego, Anita Barber, Michael Birnbaum, Delbert Robinson, Anil Malhotra

Feinstein Institute for Medical Research, Queens, New York, United States

Background: Identification of robust biomarkers that predict individualized response to antipsychotic treatment at the early stage of psychotic disorders remains a challenge in precision psychiatry. This study aimed to investigate if any functional connectome-based neural traits would serve as such a biomarker.

Methods: In a discovery sample, 49 first-episode patients with psychosis received multi-paradigm fMRI scans at baseline and were clinically followed up for 12 weeks under antipsychotic monotherapies. Treatment response was evaluated at the individual level based on the psychosis scores of the Brief Psychiatric Rating Scale (BPRS). Cross-Paradigm Connectivity and Connectome-based Predictive Modeling were employed to train a predictive model that uses baseline connectomic measures to predict individualized change rates of psychotic scores. The generalizability of the model performance was further examined in an independent validation sample of 24 first-episode patients with a similar design.

Results: The results revealed a paradigm-independent connectomic trait that significantly predicted individualized treatment outcome in both the discovery sample ($r[\text{predicted vs observed}] = 0.44$, $P = 0.007$) and the validation sample ($r[\text{predicted vs observed}] = 0.50$, $P = 0.005$). This neural trait involved connections predominantly related to the cerebellar-cortical circuitry and spanned multiple sensory (e.g., sensorimotor, auditory, visual) and cognitive (e.g. default-mode, frontoparietal, cingular-opercular) systems.

Conclusions: This study discovers and validates a connectome-based functional signature as a promising early predictor for individualized response to antipsychotic treatment in first-episode

psychosis, thereby highlighting the potential clinical value of this biomarker in precision psychiatry.

Keywords: Early Psychosis, Treatment Outcome Prediction, Functional MRI (fMRI), Brain Connectome, Machine Learning

Disclosure: Nothing to disclose.

P521. Contrasting Case-Control and Normative Reference Approaches to Capture Clinically Relevant Structural Brain Abnormalities in Antipsychotic Medication-Naïve First-Episode Psychosis Patients

Natalie Remiszewski, James Edward Bryant, Saige Rutherford, Andre Marquand, Eric Nelson, Ibrahim Askar, Adrienne Lahti, Nina Kraguljac*

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

Background: The central premise of precision medicine is that an individual's unique physiologic characteristics play a significant role in disease vulnerability and response to specific therapies. To make progress towards this goal, it is imperative to move beyond group-based studies that disregard individual variations as noise, and instead, interpret them in the context of normal-range of biological systems.

Normative modeling ("brain growth charting") is a statistical technique that allows characterization of disease signatures at the individual level, where positive (higher volumes compared to the normative range) and negative (lower volumes compared to the normative range) deviations can be calculated. Recent applications in psychiatry have shown that heterogeneity in structural brain abnormalities is substantial and that group averages do not accurately reflect patterns of atypicality at the individual level. However, it remains to be determined whether these structural deviations map onto clinical outcomes in psychosis spectrum disorders.

We enrolled antipsychotic medication-naïve first-episode psychosis patients (FEP) in a 16-week trial with risperidone and quantified raw subcortical and ventricle volumes as well as individual deviations from a normative reference model at baseline. We chose subcortical regions because they include principal sites of dopaminergic drug action and ventricles because these are one of the hallmark structural features in schizophrenia. We hypothesized that subcortical and ventricle volume deviations would be highly heterogeneous, and that deviations derived from the normative model in subcortical regions would be superior to raw volumes in predicting treatment response.

Methods: We enrolled 98 antipsychotic medication-naïve first episode psychosis patients (age 23.4 + /- 5.7, 61% male, 63% identified as racial/ ethnic minority) and treated them for sixteen weeks with risperidone. We used data from 92 controls (age 23.9 + /- 5.4, 55% male, 43% identified as racial/ ethnic minority) as comparison group for case control contrasts and calibration of normative curves.

Imaging was performed on a 3 T Siemens Magnetom Prisma using an MPRAGE [TR/TE/TI: 2400/2.22/1000 ms; 0.8mm3 voxels]. Data were processed in FreeSurfer 7.1.18.

We quantified raw subcortical volumes in FreeSurfer, as well as individual deviations based on a reference model (n = 58,836; age 2-100) using the PCN and braincharts toolkits.

Results: We found a significant group difference in raw volume measures when variables were considered jointly on all measures (Pillai's Trace= 0.18; $F = 3.23$; $p < 0.01$). Post hoc analyses demonstrated that FEP had lower thalamus ($F = 9.36$; $p < 0.01$), hippocampus ($F = 17.23$; $p < 0.01$), amygdala ($F = 6.55$; $p = 0.01$), ventral diencephalon ($F = 4.84$; $p = 0.03$), and brain stem volumes ($F = 8.39$; $p < 0.01$) compared to controls.

Across all regions, 36% of FEP displayed extreme deviations, 22% had them in ventricles, and 22% had them in subcortical regions. Extreme deviations were shared among patients more commonly in ventricle regions (6-9% of FEP) compared to subcortical regions (0-5%). They were twice as likely to be in the negative (lower volume compared to the normative reference) than positive (higher volume) direction in subcortical regions, whereas 62% of extreme ventricle deviations were in the positive direction.

While subcortical raw volumes at baseline did not predict treatment response, deviations in the caudate, putamen, and ventral diencephalon did. For these regions, correlations significantly differed between raw volume and deviation measures in the caudate ($z = -2.17$; $p = 0.03$) and putamen ($z = -2.15$; $p = 0.03$). Ventricle raw volumes or deviations did not predict response.

Conclusions: Our finding that structural deviations are heterogeneous in antipsychotic-naïve FEP is consistent emerging literature reporting this in several major psychiatric disorders. We extend the literature by providing empirical evidence that greater negative deviations in key dopaminergic brain regions better predict antipsychotic treatment response in FEP compared to raw volumes.

Our data suggests that this normative modeling allows to capture inter-individual heterogeneity of regional brain volumes in FEP and to characterize structural pathology in a clinically relevant fashion. This holds great promise for progress in precision medicine in psychiatry where group-level studies have failed to derive reliable maps of structural pathology.

Keywords: Medication-Naive First Episode Psychosis, Predictor of Treatment Response, Normative Modelling, Subcortical Volume

Disclosure: Neurocrine Biosciences, Inc.: Advisory Board (Self), American Board of Psychiatry and Neurology: Other Financial or Material Support (Self)

P522. Prolonged Kynurenic Acid Elevation During the Prenatal Period Elicits Electrophysiological and Behavioral Changes in Adult Mice

Robert Schwarcz*, Sarah Beggiato, P. Leon Brown, Snezana Milosavljevic, Marian Thomas, Maria Piroli, Korrapati Sathyasaikumar, Francesca Notarangelo, Ana Pocivavsek

Maryland Psychiatric Research Center, Baltimore, Maryland, United States

Background: Neuroactive metabolites of the kynurenine pathway (KP) of tryptophan degradation are believed to be involved in the pathophysiology of several psychiatric diseases, including schizophrenia (SZ) and bipolar disorder (BPD). Kynurenic acid (KYNA) has received special attention in this regard since its levels are elevated in the brain and CSF of persons with SZ and may be causally related to the cognitive dysfunctions seen in the disease. Previous studies in rats had indicated that abnormally high KYNA levels in the fetal brain may play a pathophysiologically significant role in this context ("EKyn model"; Notarangelo and Pocivavsek, 2017; Beggiato et al., 2018).

Methods: In anticipation of future studies with genetically altered animals, we now fed pregnant C57Bl/6 J mice either with the immediate KYNA precursor kynurenine (10 mg or 30 mg/day; EKyn) or with control mash (ECon) from embryonic day (ED) 11 to ED 18. Separate cohorts of adult, male and female offspring were then assessed in electrophysiological, behavioral, and biochemical studies on postnatal day (PD) 56-70. Electrophysiology was conducted by ex vivo recording of evoked local field potentials in coronal brain slices (N = 10 per group). Barnes maze, a spatial learning task, was used to evaluate hippocampal-dependent acquisition training (2 trials per day for 3 consecutive days) and

prefrontal cortex-dependent reversal learning (2 trials on the 4th day of testing; N = 20 – 28 per group). Biochemical analyses were designed to measure the KP metabolites 3-hydroxykynurenine (3-HK) and KYNA in tissue homogenates from hippocampus and cortex (N = 7 – 8 per group) and to determine extracellular KYNA levels by in vivo microdialysis in the prefrontal cortex (N = 4 – 5 per group).

Results: Ex vivo recordings revealed that prenatal treatment with 30 mg kynurenine/day promoted a longer contralateral response latency in EKyn compared to ECon mice (*P < 0.05), likely reflecting a decline in white matter function. Adult male EKyn mice also showed significantly longer latency (*P < 0.05) and lower speed to enter the escape box during acquisition training when spatial learning was tested in the Barnes maze. During subsequent reversal learning, the latency to escape into the goal box and the number of re-visits to the previous escape location were higher in male EKyn offspring compared to controls (*P < 0.05). Of note, female EKyn mice were not impaired in the Barnes maze. Measured in the cortex or hippocampus in adulthood, the tissue levels of 3-HK and KYNA did not differ between EKyn and ECon groups. Separately, in preparation of experiments designed to investigate the role of KYNA in the adverse long-term effects seen in EKyn mice, we verified by in vivo microdialysis that inhibition of KYNA neosynthesis with the potent kynurenine aminotransferase inhibitor PF-04859989 (30 mg/kg, s.c.) induces a rapid decrease in extracellular KYNA in the prefrontal cortex of adult EKyn animals.

Conclusions: The observation that physiological and behavioral abnormalities are seen in adult EKyn mice without concurrent elevations in brain KYNA levels indicates that prolonged treatment with kynurenine during the embryonic period causes permanent, functionally significant changes in brain development in these experimental animals. Elaboration of the mechanisms underlying these changes, especially of the role of cerebral KP metabolism in this context, can be expected to provide useful information regarding the etiology of neuropsychiatric illnesses.

Keywords: Kynurenic Acid, Perinatal Development, Schizophrenia

Disclosure: Nothing to disclose.

P523. Incomplete Hippocampal Inversion Impacts Hippocampal Subfield Morphology in Schizophrenia

Maxwell Roeske*, Maureen McHugo, Simon Vandekar, Kristan Armstrong, Stephan Heckers

Vanderbilt University School of Medicine, Nashville, Tennessee, United States

Background: Hippocampal morphology is altered in schizophrenia, but it is unclear whether deficits in specific regions and subfields are linked to altered neurodevelopment. Converging shape and volumetric analyses suggest that the anterior region and cornu ammonis (CA) 1 subfield are affected in the early stages of illness. A recent study revealed that incomplete hippocampal inversion (IHI), an anatomical variant of the human hippocampus resulting from an arrest during neurodevelopment, is more prevalent and severe in patients with schizophrenia. We hypothesized that IHI contributes to hippocampal subfield differences in schizophrenia.

Methods: We studied 199 schizophrenia patients and 161 healthy individuals with 3-Tesla MRI and measured the prevalence and severity of IHI in each hippocampus bilaterally using established, quantitative criteria. We generated hippocampal surface reconstructions using a combination of manual and Freesurfer automated segementation and the SPHARM-PDM toolkit. We regressed the average local shape vertex displacement onto IHI score to test the effect of IHI on hippocampal shape variation and conducted a sensitivity analysis to test for group shape differences with and without IHI included as a main effect. We completed the shape analysis using SurfStat and applied random field theory to account for multiple comparisons. We measured volumes of subfields (CA1, CA2/3, dentate gyrus, subiculum) across anterior and posterior hippocampal regions using ASHS automated segmentation in an overlapping group of participants: 86 individuals in the early stage of a psychotic disorder and 67 demographically similar healthy individuals. We completed statistical analysis of hippocampal subfield volume using linear mixed models in R. We included age, sex, and intracranial volume as covariates in all models.

Results: IHI is associated with rounding of hippocampal shape. Linear models not including IHI as a main effect replicated well-known hippocampal shape differences in schizophrenia patients localized to the CA1 region of the antero-lateral hippocampus. Including IHI as a main effect in the model eliminated the bilateral significant shape differences in the CA1 subfield. The relationship between IHI score and hippocampal volume varied by region and subfield ($F_{2,2253} = 11.76$, $P < 0.001$). Increased IHI score was associated with lower volume in the anterior CA1 (slope = -21.46, 95% CI = [-27.32, -15.61]) and anterior dentate gyrus (slope = -14.24, 95% CI = [-20.09, -8.39]) subfields.

Conclusions: IHI impacts hippocampal shape and volume with regional and subfield specificity. Our results suggest that abnormal development of the hippocampus contributes to morphologic differences, particularly in the anterior CA1 subfield, observed in patients with schizophrenia.

Keywords: Hippocampal Subfields, Schizophrenia (SCZ), Human Neuroimaging, Neurodevelopment

Disclosure: Nothing to disclose.

P524. Altered Maturation of Functional Connectivity in Thalamocortical Networks in 22q11.2 Deletion Syndrome

Charles Schleifer*, Kathleen O'Hora, Amy Lin, Leila Kushan-Wells, Carrie Bearden

University of California, Los Angeles, Los Angeles, California, United States

Background: 22q11.2 Deletion Syndrome (22qDel) is a recurrent copy number variant (CNV) in which a hemizygous deletion of ~46 protein-coding genes from the long arm of chromosome 22 results in greatly increased risk for schizophrenia and other developmental neuropsychiatric conditions. Studying individuals with 22qDel can provide a powerful framework for linking genes to brain phenotypes and behaviors relevant to these conditions. The cortex and thalamus are anatomically and functionally organized into a set of distributed circuits that support diverse brain functions. Dysconnectivity within these thalamocortical circuits has been consistently implicated in idiopathic neuropsychiatric disorders, and in 22qDel. Specifically, resting-state functional MRI has reproducibly revealed thalamic dysconnectivity to a network of frontoparietal regions and a network of somatomotor regions as a biomarker of both schizophrenia and 22qDel. Psychosis spectrum disorders generally emerge in mid-adolescence to early adulthood, yet no study to date has investigated longitudinal development of functional connectivity in thalamocortical networks in 22qDel during this sensitive age period.

Methods: In a longitudinal sample of resting-state fMRI from 22qDel (n = 100, 62% female) and typically developing (TD) youth (n = 100, 48% female) ages 6-22 y, we used an atlas-based approach to compute thalamocortical functional connectivity across a set of functionally defined networks. Thalamocortical

connectivity was measured as the correlation in fMRI time-series between thalamic and cortical regions classified as belonging to the same functional network based on an a priori parcellation. For both groups, non-parametric age-related curves for thalamocortical connectivity were generated using Generalized Additive Mixed Models.

Results: We report a novel finding of altered thalamocortical developmental trajectories, particularly in the frontoparietal and somatomotor networks. In both networks, TD controls exhibit a relatively flat developmental trajectory with no significant effect of age on connectivity (Somatomotor $F = 0.11$, $p = 0.75$; Frontoparietal $F = 0.97$, $p = 0.33$). However, 22qDel show significant changes across the age range in both networks (Somatomotor $F = 8.13$, $p = 0.005$; Frontoparietal $F = 7.11$, $p = 0.0007$). GAMM curves were compared for regions of non-overlap based on 95% confidence intervals. In the youngest 22qDel patients, thalamic-frontoparietal connectivity is lower than TD but switches to overconnectivity in mid-adolescence. The opposite pattern is observed for somatomotor connectivity.

Conclusions: This pattern implicates dysfunction and altered maturation of circuits involved in sensory and executive function which are disrupted in 22qDel and related conditions like Autism Spectrum Disorder and schizophrenia, and broadly aligns with findings of age-related functional connectivity changes in the 22q11 mouse model.

Keywords: 22q11.2 Deletion Syndrome, Thalamo-Cortical Connectivity, Longitudinal MRI, Copy Number Variants

Disclosure: Nothing to disclose.

P525. Cannabis Use is Associated With More Symptoms and Better Functioning but Not Psychotic Conversion in Participants at Clinical High Risk (CHR) for Psychosis in the NAPLS 3 Study

Kristin Cadenhead*, Jean Addington, Carrie Bearden, Tyrone Cannon, Matcheri Keshavan, Daniel Mathalon, Diana Perkins, Elaine Walker, Scott Woods

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

Background: Multiple studies have demonstrated an association between cannabis use and future risk of psychosis. In patients with established psychosis, cannabis use is also associated with worsening psychotic symptoms and better functioning in some studies. The aim of the present study was to examine cannabis use patterns across CHR and Health Comparison (HC) participants in the NAPLS 3 consortium and determine the association to symptoms, functioning and psychotic conversion.

Methods: The sample included 699 participants at CHR for psychosis and 96 HCs. All participants were queried about current and past cannabis use, symptoms and functioning. A total of 71 CHR participants are known to have converted to psychosis during the course of the study.

Results: The CHR participants had a higher rate of cannabis use compared to HCs (50.9% vs 32.3%, $p < 0.001$) and the majority used at least once per week. Within the CHR sample who had used cannabis, an earlier age of first use was significantly correlated ($r = -0.15$, $p < 0.001$) with greater use in the last 6 months. Heavier use in the last 6 months was also associated with greater positive ($r = 0.20$, $p < 0.001$) and disorganized ($r = 0.11$, $p < 0.02$) symptoms at baseline assessment. Heavier cannabis users had greater positive symptoms ($F = 4.6$, $p < 0.01$) but better global ($F = 3.9$, $p < 0.05$) and social ($F = 8.8$, $p < 0.001$) functioning than those with none or minimal cannabis use. There was no significant difference in the rate or pattern of cannabis use between the CHR sample who converted to psychosis and those who did not.

Conclusions: The CHR sample in the NAPLS 3 study has a higher rate and heavier pattern of cannabis use compared to HC participants. Heavier use was associated with more positive symptoms but not conversion to psychosis in this sample. Heavier cannabis users tended to have better functioning consistent with prior studies in schizophrenia populations.

Keywords: Clinical High Risk for Psychosis, Cannabis Use, Social Functioning

Disclosure: Nothing to disclose.

P526. Risk of Sudden Cardiac Death Among Patients With Schizophrenia, and Other Psychotic Disorders: An Exploratory Analysis Using the Million Veteran Program Dataset

Rishab Gupta*, Nina Schooler, Panos Roussos, Mihaela Aslan, Philip Harvey, Tim Bigdeli

SUNY Downstate Medical Center, Jamaica Plain, Massachusetts, United States

Background: Patients with schizophrenia typically die ~15-20 years earlier compared to those without any psychiatric illness. The two most common causes of death in patients with schizophrenia are cardiovascular disease (most commonly, myocardial infarction) and suicide. In particular, patients with schizophrenia are up to 4.5 times more likely to die from sudden cardiac death (SCD). SCD in schizophrenia may occur in the context of either a structural heart disease (coronary heart disease, congestive heart failure) or due to arrhythmia (genetic or multifactorial). Psychotropic medications also predispose to SCD by prolonging QTc interval and leading to Torsades de pointes. To date, most studies have focused on schizophrenia only to estimate the risk of SCD even though people with other psychotic disorders also share risk factors similar to schizophrenia.

Methods: We conducted a cross-sectional observational study to evaluate the prevalence of SCD in patients with schizophrenia, and a broader psychosis phenotype enrolled in the VA Million Veteran Program study. We operationalized some definitions for this study: schizophrenia was defined as patients assigned phecode 295- (created using ICD-9 and ICD-10 equivalent codes) over at least two visits; a broader psychosis phenotype was defined based on two or more phecodes for psychotic disorders, 295-298 (created using ICD-9/10 codes); and SCD was defined based on phecode 427.5 (created using ICD-9/10 codes) for cardiac dysrhythmia not otherwise specified (NOS) (as a proxy).

Results: Among 654,023 participants with available data, 17,900 met criteria for schizophrenia (~2.7%). There were 46,885 patients with history of cardiac dysrhythmia NOS in the total eligible sample, out of which 1,839 also had co-morbid schizophrenia (~7.25%). We calculated the odds ratio of 1.45 ($p < 0.0001$) for those with history of cardiac dysrhythmia NOS and co-morbid schizophrenia versus those with cardiac dysrhythmia NOS but without co-morbid schizophrenia. We next applied general linear modeling, with age, age-squared, gender, and self-identified race as covariates to examine the relationship between schizophrenia and cardiac dysrhythmia NOS. We found that cardiac dysrhythmia NOS was positively predicted by schizophrenia ($\beta = 0.4881$; $p < 2\times10\text{-}16$), African American ancestry ($\beta = 0.122$; $p < 2\times10\text{-}16$), and age ($\beta = 0.1122$; $p < 2\times10\text{-}16$), while Asian ancestry ($\beta = -0.7569$; $p < 2\times10\text{-}16$) and male gender ($\beta = -0.1145$; $p = 2.48\times10\text{-}9$) were protective factors. Surprisingly, age-squared was also a negative predictor of cardiac dysrhythmia NOS ($\beta = -0.6369 \times10\text{-}3$; $p < 2\times10\text{-}16$). Among more than 30,000 participants meeting criteria for the broader psychosis phenotype, 5,371 also met criteria for cardiac dysrhythmia NOS, corresponding to a 1.73-fold increased risk ($p < 0.0001$). Demographic covariates showed a near-identical pattern of association as observed for schizophrenia.

Conclusions: Among individuals receiving health care at the VA, we observed significantly elevated odds of sudden cardiac death among individuals with diagnosed schizophrenia or a related psychotic disorder. We observed risk-increasing effects of age and African American ancestry and protective effects of Asian ancestry, male gender, and age-squared. Implications of this work include recognition of potentially modifiable mortality risk, need for a close examination of the risk factors for SCD, and possible ethnic disparities in cardiac outcomes of patients with a psychotic disorder. Ongoing analyses are leveraging available mortality records to identify confirmed cases of SCD, and evaluation of how polygenic risk for schizophrenia and heart disease interplay to moderate cardiac outcomes in our patients. This study will build on our recent work showing that polygenic risk score for schizophrenia increase the risk for a range of medical disorders.

Keywords: Sudden Cardiac Death, Schizophrenia, Psychotic Disorder

Disclosure: Nothing to disclose.

P527. The Relationship Between Breast Cancer and Long-Term Exposure to Prolactin Sparing Anti-Psychotic Medications Among Schizophrenia Patients: A Population-Based Study

Amir Krivoy, Ariel Asper, Noa Menkes-Caspi, Michael Davidson, John M Davis, Mark Weiser*

Sheba Medical Center, Ramat Gan, Israel

Background: Most anti-psychotics cause an increase in prolactin, a hormone hypothesized to increase risk for breast cancer.

Methods: We utilized population-based data from the Clalit Health Services (CHS) database, the largest provider of health insurance in Israel (N = 5.5 million persons), followed-up between 2000 and 2020. In 15,686 female patients with schizophrenia, we examined the relationship between anti-psychotic medication and later breast cancer. Specifically, we wanted to see whether female schizophrenia patients who were exposed to at least 4 years of prolactin sparing anti-psychotic medication (clozapine, quetiapine, aripiprazole) were at a lower risk of developing breast cancer compared to female schizophrenia patients who were exposed to at least 4 years of non-prolactin sparing anti-psychotic medication and were not exposed to prolactin sparing anti-psychotic medications.

Results: Female schizophrenia patients who were exposed to at least 4 years of prolactin sparing anti-psychotic medication were at a lower risk of developing breast cancer compared to female schizophrenia patients who were exposed to at least 4 years of anti-psychotic medication (HR = .44, $p < .001$). This trend remained significant even after adjusting for age, socioeconomic status, smoking, diabetes and obesity (HR = .5, $p < .001$).

Conclusions: In female schizophrenia patients, prolactin sparing antipsychotic medications decrease the risk for breast cancer compared to non-sparing anti-psychotic medications. This should be taken into consideration when prescribing anti-psychotic medications to female schizophrenia patients.

Keywords: Antipsychotics, Breast Cancer, Schizophrenia (SCZ), Prolactin

Disclosure: Jansen, Lundeck: Advisory Board (Self), Teva, Dexcel, Msd, Orion, Clearmind: Consultant (Self), Pfizer: Honoraria (Self), Minerva: Contracted Research (Self)

P528. Violent Behavior Among Young Adults Receiving Treatment for Early Psychosis

Stephanie Rolin*, Leah Pope, Elizabeth Ford, Michael Compton, Paul Appelbaum, Lisa Dixon

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

Background: Overall, the absolute risk of violence for individuals with serious mental illnesses (SMI) is low. However, when compared to the general population, people with SMI have an increased absolute risk of engaging in violent behavior. This seems to be particularly true for young adults experiencing early stages of psychosis, with research suggesting that violence risk peaks during the period of early psychosis. This study analyzed the prevalence of and risk factors for violent behavior in a sample of young adults receiving treatment for non-affective psychosis in an early intervention services (EIS) setting.

Methods: A total of 1726 young adults (ages 16-30) within two years of onset of non-affective psychosis were enrolled in 21 EIS sites in New York State between October 2013 and January 2020. Clinical teams completed standardized forms on all enrolled participants every 3 months, including measures of demographics, clinical outcomes, and violent behavior. Sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) of violent behavior at time of entry into treatment in predicting future violent behavior was calculated.

Results: The total sample was mostly men (n = 1263, 73%) and included 599 Black (35%), 454 Latinx (26%), 417 White (24%) and 153 Asian (9%) individuals. Approximately three-quarters (n = 1238, 73.4%) of the sample had no violent behavior at any time point. Of individuals with violent behavior at time of entry into treatment at an EIS clinic (n = 452, 26.7%), approximately half had violent behavior that persisted during treatment (n = 228, 50.4%) and half had violent behavior that resolved during treatment (n = 224, 49.6%). The sensitivity, specificity, PPV and NPV of violent behavior at time of entry for predicting future violent behavior during treatment was 32.4%, 82.5%, 18.3%, and 91.0%.

Conclusions: Overall, violent behavior is relatively uncommon among a sample of young adults receiving treatment in an EIS setting for nonaffective psychosis. Among individuals who have violent behavior at time of entry into treatment, about half do not have violent behavior following entry into treatment. Overall, past violent behavior had low sensitivity in predicting future violent behavior.

Keywords: Early Psychosis, Violence, Early Intervention Services

Disclosure: Nothing to disclose.

P529. Lifetime Consequences and Correlates of Anticholinergic Medication Burden in the VA Healthcare System

Tim Bigdeli*, Yash Joshi, Yuli Li, Nallakkandi Rajeevan, Fred Sayward, Ronald Przygodzki, Grant Huang, Saiju Pyarajan, Sumitra Muralidhar, J Michael Gaziano, David Braff, Gregory Light, Mihaela Aslan, Panos Roussos, Philip Harvey

SUNY Downstate Medical Center, Brooklyn, New York, United States

Background: Cognitive impairments are a hallmark of schizophrenia (SCZ) and can be worsened by medications with anticholinergic properties used to mitigate side-effects of commonly prescribed antipsychotics. Following recent reports from the COGS and BSNIP consortia that demonstrate the robust association of anticholinergic medication burden (ACB) across multiple cognitive domains, we replicated these findings in VA Cooperative Studies Program (#572), a cohort of 9,378 patients with confirmed diagnoses of SCZ or bipolar I disorder (BIP). We extend these findings here, hypothesizing that cumulative ACB from multiple medication classes may underlie the increased prevalence of dementia among individuals diagnosed with SCZ.

Methods: Leveraging the VA's extensive electronic health record (EHR), we constructed individual-level ACB scores from prescription records for over 700,000 participants in the Million Veteran Program (MVP). We included first- and second-generation antipsychotics, mood stabilizers, antidepressants, anticholinergics, antiparkinsonian medications, anticonvulsants, and benzodiazepines. We examined the distribution of ACB among diagnosed cases of SCZ and BIP, and in the VA patient population at large, and modeled the associations of ordinally-coded ACB (0-2 => "low"; 3-4 => "intermediate"; 5 or greater => "high") on ~1,600 categories of ICD-9/10 billing codes ("phecodes"). Our preliminary analyses consider all inpatient and outpatient prescriptions within 90 days of study enrollment.

Results: Among 16,287 diagnosed SCZ patients, and after controlling for recent inpatient hospitalization, high ACB corresponded to a 1.4-times higher risk (95% CI: 1.2,1.6; p < 10^-5) of being diagnosed with dementia (phecode 290.1). Among SCZ patients, high ACB also significantly increased risk of being diagnosed with mild cognitive impairment (phecode 292; OR = 1.44, 95% CI: 1.3,1.6; p < 10^-17), suicidal behavior or ideation (phecode 297; OR = 1.41, 95% CI: 1.3,1.6; p < 10^-14), and tobacco use disorder (phecode 318; OR = 1.36, 95% CI: 1.2,1.5; p < 10^-10). We observed comparable effects of high ACB on dementia risk in 32,022 BIP patients (OR = 1.46, 95% CI: 1.3,1.6; p < 10^-13). This effect was still detectable in 9,935 patients who had not been treated with antipsychotics or mood-stabilizers and had no record of inpatient treatment (OR = 1.53, 95% CI: 1.27,1.83; p < 10^-5).

Conclusions: We have previously reported that ACB robustly influences cognitive performance in SCZ and BIP patients, even after adjusting for the influences of age, sex, gender, and IQ polygenic scores. In the current analysis, we explore the broader consequences of chronic ACB exposure in a broader cohort of participants. Planned follow-up analyses include joint modeling of ACB and polygenic scores for SCZ, Alzheimer's Disease, and executive functioning, and comparative analyses of partial-ACB attributable to particular classes of medications.

Keywords: Schizophrenia (SCZ), Anticholinergic Medication Burden, Electronic Health Record (EHR), Veterans, Polygenetic Risk Score

Disclosure: Nothing to disclose.

P530. Examining Mitochondrial DNA Variants in the Risk for Schizophrenia

Fernanda Souza, Ana Paula Mendes-Silva, Wei Deng, Lei Sun, James Kennedy, Marquis Walter, Vanessa Gonçalves*

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

Background: Schizophrenia is a complex disorder with clinical and biological heterogeneity. Despite of the recent success in identifying contributing genetic variants from the nuclear genome, the effective diagnosis, treatment and prevention strategies are largely absent. Mitochondria are crucial for the brain physiology, and mitochondrial DNA (mtDNA) variants may contribute to the etiology of mental illnesses. Previous studies of primary mitochondrial diseases have observed impairment in brain function and high prevalence of psychosis among cases. Here, we aim to identify novel mitochondrial variants associated with schizophrenia using a subset of the Psychiatric Genomics Consortium (PGC) schizophrenia samples, which had mtDNA available for analysis.

Methods: In this study, we assess the contribution of mtDNA and jointly the influence of nuclear genome on the risk of SCZ using a subset of carefully curated samples (n = 37,500) with high-quality mtDNA data available in the Psychiatric Genomics Consortium. The mtDNA associations were performed individually for common single nucleotide variants (minor allele frequency > 1%) and aggregated at the gene-level for rare variants (minor allele frequency < 1%). Mitochondrial haplogroups were assigned using HaploGrep2 and were tested at macro-group level (H (H-HV-V), JT (J-T), UK and Others (W-X-I). To harmonize mtDNA variants across the genotyping platforms and to increase the number of variants for analysis, we performed imputation using a custom reference panel.

Results: After standard quality control and imputation, we recovered 370 mtDNAs that were partially shared across the 37,500 samples (17,283 cases and 20,217 controls), including 144 common variants (MAF > 0.01) and 226 rare variants (MAF between 0.001 and 0.01). Among the macro-haplogroups, only JT was marginally associated with a decreased risk of SCZ (Odds ratio = 0.94, p = 0.035) while adjusting for sex. For common variants (N = 144), the SNP MT-15924 was significant after correcting for macro-haplogroup H (p = 0.00026). The gene-set analysis revealed three potentially interesting gene complexes: complex I (gene ND1-6), complex IV (cytochrome c oxidase 1-3), and complex III (cytochrome b) that deserved further investigation into individual gene component.

Conclusions: To the best of our knowledge, the current analysis is the largest in major psychosis. These positive findings are expected to shed light on the roles of mitochondrial variants in schizophrenia etiology and complement discoveries from the nuclear genome.

Keywords: Mitochondrial DNA, Genomics, DNA, Whole-Genome, Sequencing, Schizophrenia

Disclosure: Nothing to disclose.

P531. The Three Dimensional Landscape of Open Chromatin Regions in Schizophrenia

Kiran Girdhar*, Jaroslav Bendl, Pengfei Dong, Steven Kleopoulos, Samir Rahman, Vahram Haroutunian, Gabriel Hoffman, John Fullard, Panos Roussos

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

Background: To translate scientific discoveries from lab to clinic for treatment requires deeper understanding of molecular mechanisms of disease. One of the ways to gain more insights in mechanism is to investigate the structure and function of human genome in affected cell type in diseases. For example, to identify how risk variants are organized in three-dimensional genome and what specific cellular functions are affected in disease. In this study, we investigate how majority of schizophrenia (SCZ) variants underneath non-coding regions are organized in three-dimensional genome in a critical cell type neurons in schizophrenia (SCZ). To answer this, we profile population scale (N = 1,394) chromatin accessibility libraries from neurons and non-neurons in two neocortical brain regions.

Methods: Using frozen postmortem tissue from 469 cases with SCZ and controls, we performed ATAC-seq to profile chromatin accessibility in neuronal and glial populations of cells, isolated by FACS from two different brain regions, i.e. dorsolateral prefrontal cortex and anterior cingulate cortex. Then, we characterized epigenetics changes associated with SCZ phenotypes. The availability of RNA-seq and whole-genome data for this cohort allowed us to measure the overlap between differentially regulated transcriptome and epigenome signatures and related pathways. Finally, we determined enhancer-promoter interactions, by utilizing additional omics in the brain tissue (Hi-C and H3K27ac ChIP-seq) and jointly analyzing with ATAC-seq based on the "activity-by-contact" (ABC) approach.

Results: First, multi-omic data integration associated global patterns of changes in chromatin accessibility with gene expression and identified enhancer-promoter interactions that are impaired in disease phenotypes. Second, we explored cis-regulatory domains (CRDs), a structural attribute of 3D genome architecture and demonstrated how schizophrenia specific dysregulation in CRDs contributes to nuclear topography. We found genome wide mapped CRDs delimited the cell-region specific OCRs (~35%) into active chromatin regions enriched for SCZ risk loci. Interestingly, SCZ heritability was found significantly enriched at CRD boundaries and unique to neuronal cell types indicating borders OCRs prominently represented by promoters as sites of dysregulation in SCZ patients. Third, trans interactions of diseased CRDs revealed a separate cluster of domains governing fetal development in higher order A (Active) compartments.

Conclusions: This finding links the disease risk loci to the chromatin landscape that could be reconfiguring during the early stages of fetal development. Overall, this dataset provides a unique insight into molecular mechanisms underlying brain region and cell type-specific vulnerability in both schizophrenia and bipolar disorder.

Keywords: Neuropsychiatric Disorders [Schizophrenia, Parkinson's Disease, Major Depressive Disorder], Computational Genomics, Open Chromatin Regions

Disclosure: Nothing to disclose.

P532. Diurnal Alterations in Gene Expression across the Human Dorsal and Ventral Striatum in Psychosis and Unaffected Comparison Subjects

Kyle Ketchesin*, Wei Zong, Mariah Hildebrand, Madeline Scott, Marianne Seney, Kelly Cahill, Vaishnavi Shankar, Jill Glausier, David Lewis, George Tseng, Colleen McClung

University of Pittsburgh, Pittsburgh, Pennsylvania, United States

Background: Psychosis is a defining feature of schizophrenia and highly prevalent in bipolar disorder. Individuals suffering with these illnesses also have major disruptions in sleep and circadian rhythms, and disturbances to sleep and circadian rhythms can precipitate or exacerbate psychotic symptoms. A prior study from our group used a time-of-death analysis of RNA-seq data and found that subjects with schizophrenia show altered gene expression rhythms in the human postmortem dorsolateral prefrontal cortex (dlPFC), a region associated with the cognitive symptoms of schizophrenia. Positive symptoms (i.e., psychosis) are associated with the striatum, though no study to date has directly measured molecular rhythms and determined how they are altered in the striatum of subjects with psychosis.

Methods: In the current study, we performed both differential expression and rhythmicity analyses to determine diurnal alterations in gene expression across the nucleus accumbens (NAc), caudate, and putamen in subjects with psychosis relative to unaffected comparison subjects. NAc, caudate, and putamen samples were collected from male and female subjects with psychosis [$n = 36$; schizophrenia/schizoaffective disorder ($n = 28$) and bipolar disorder with psychosis ($n = 8$)] and unaffected comparison subjects ($n = 36$). Total RNA-seq was performed on the striatal tissue samples. Differential expression analyses were first performed between psychosis and comparison subjects in each region regardless of time of death (TOD). Given our previous findings in the dlPFC, we then split the cohorts into subjects who died either during the day or during the night and performed differential expression analyses. Transcripts were considered differentially expressed if $p < 0.01$ and log2 fold change $\leq$ -0.26 or $\geq$0.26. Circadian patterns of gene expression were detected using nonlinear regression based on individual subject TOD. Sinusoidal curves were fitted to the expressed data using the nonlinear least-squares method and coefficient of determination (R2) was used as a measure of goodness-of-fit. Transcripts with either a gain or loss of rhythmicity between psychosis and matched comparison subjects were determined using the difference in R2 between the cohorts.

Results: Across the three striatal subregions, we found differential expression of immune-related transcripts and a substantial loss of rhythmicity in core circadian clock genes in subjects with psychosis. In the NAc, mitochondrial-related transcripts showed decreased expression in psychosis subjects, but only in those who died at night. Moreover, we found a loss of rhythmicity in small nucleolar RNAs and a gain of rhythmicity in glutamatergic signaling in the NAc of psychosis subjects. Between region comparisons revealed that rhythmicity in the caudate and putamen is highly similar in subjects with psychosis, suggesting synchronization in daily rhythms across the dorsal striatum in these subjects.

Conclusions: Overall, we have identified multiple gene expression differences in the striatum in subjects with psychosis, as well as changes in rhythmic gene expression that could underlie disease pathology or response to treatment. Ongoing studies focused on between-region differential expression analyses in both unaffected comparison and psychosis subjects will provide insight into how the identified gene expression differences are coordinated across ventral and dorsal striatal subregions.

Keywords: Human Postmortem Brain Tissue, Circadian Rhythms, Psychosis, Schizophrenia (SCZ), RNA-seq

Disclosure: Nothing to disclose.

P533. Pathways From Genes to Functioning: Integrating Genetic Risk, Neurophysiology, Anticholinergic Medication Burden, Cognition, and Symptom Contributions to Disability in Schizophrenia

Yash Joshi, Michael Thomas, Tim Bigdeli, Juan Molina, David Braff, Neal Swerdlow, Gregory Light*

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

Background: Emerging disease-relevant measures generated from large schizophrenia (SZ) psychiatric consortium studies invite opportunities to investigate multivariate contributions to psychosocial disability. Clarification of pathways which lead to disability have substantial implications for drug development, research funding allocation, clinical care delivery, and health policy. The objective of this study was to assess the contributions of genetic risk, neurophysiologic abnormalities, medication burden, cognition, and symptoms to disability in SZ.

Methods: SZ outpatients ($n = 1120$) were assessed via their participation in the Consortium on the Genetics of Schizophrenia-2 (COGS-2) study. Polygenic scores for SZ risk (SZ PRS) and for intelligence (IQ PS) were derived via the PsychArray and Multi-Ethnic Global Array. Neurophysiologic indices of early auditory information processing (EAIP) included mismatch negativity, P3a, and reorienting negativity. A modified Anticholinergic Cognitive Burden (ACB) Scale served to generate ratings of anticholinergic medication burden. Positive (POS) and Negative (NEG) symptoms were assessed via the Scales for the Assessment of Positive and Negative Symptoms. Cognition (COG) was measured via the Penn Neurocognitive Battery. Functional outcomes (FNX) were measured by the Role Functioning Scale. Informed by prior studies, an iterative multivariate approach testing 28 separate models using structural equation modeling (SEM) was used to generate an integrative model of SZ disability.

Results: An optimized model illustrated that FNX was affected by COG (via EAIP), and separately, NEG. ACB impacted FNX (d = -0.44) via independent effects on EAIP, COG and NEG. By contrast, POS was only associated with NEG, and both SZ PRS and IQ PS did not affect FNX.

Conclusions: Genetic factors and positive symptoms have little impact on disability in SZ outpatients stabilized on psychotropics. Anticholinergic medication burden reduction and remediation of information processing are predicted to have beneficial effects on cognition, symptoms and psychosocial outcomes in SZ. Iterative SEM-based modeling may be a useful tool in clarifying multivariate effects in large SZ studies.

Keywords: Polygenetic Risk Score, Cognition, Neurophysiology, Schizophrenia (SCZ), Anticholinergic Medication Burden

Disclosure: Astellas, NeuroSig, Sosei-Heptares: Consultant (Self)

P534. Additive and Multiplicative Influence of CNVs and Common SNPs on Cognition

Emma Knowles, Josephine Mollon, Laura Schultz, Guillaume Huguet, Samuel Mathias, Amanda Rodrigue, Aaron Alexander-Bloch, Zohra Saci, Laura Almasy, Sebastien Jacquemont, David Glahn*

Boston Children's Hospital, Harvard Medical School, Boston, Massachusetts, United States

Background: Numerous copy number variants (CNVs) have been identified as risk markers for neurodevelopmental and psychiatric disorders. Such disorders are characterized by decrements in cognitive performance. Yet, the influence of these CNVs, and their joint influence in interaction with known common risk variants for psychiatric illness, on neurotypical cognitive performance remains unclear. We investigate the influence of common CNVs and polygenic risk for both psychiatric illness and intelligence related phenotypes (e.g., brain morphology) on general cognitive ability (g) and reaction time in the UK Biobank (UKBB).

Methods: Reaction time data were available in 491,185 individuals (mean age = 57.59, sd = 8.11, range = 39.17 - 76.54 years, 54.39% female). Reaction time data was winsorized and inverse normalized. g was calculated in a subset of the sample (N = 257,976) in line with a previously published investigation of the integrity and structure of the cognitive data from the UKBB (Lyall et al, 2016). To minimize missingness all cognitive data was collapsed across time points, where missing data at a preceding time point was directly imputed with available data from the following one. CNVS were called using PennCNV and QuantiSNP via an established pipeline from genotypic data derived by the UKBB. CNVs were annotated using Gencode for overlapping gene components. Polygenic scores were calculated using an established pipeline for White British individuals in the sample. PRS for disorders related to cognitive ability were calculated using summary statistics derived from samples with minimal overlap with the UKBB. Linear regression models, run in python (statsmodels) and R (lm), were used to test for main effects of CNV, PRS, and CNV*PRS interactions on cognitive ability. Deletions or duplications with fewer than five copies were dropped prior to regression analyses.

Results: Effects for overall CNV effect were small by significant for both cognitive phenotypes. CNV carriers showed lower reaction time (B = -0.10, p = 1.78×10-13) and g scores (B = -0.10, p < 2.00×10-16). Significant effects were observed for the presence of any deletion and any duplication also. Nominally significant CNV*PRS interaction effects included schizophrenia (B = -0.1, p = 0.03), brain surface area (B = -0.05, p = 0.04), autism spectrum disorder (B = 0.03, p = 0.02). Top-ranked individual CNVs with small to large effect on reaction time included deletions in 3q29 (B = 1.17, p = 4.10×10-04), 7q11.23 (B = -0.52, p = 3.62×10-03), 15q13.1 (B = -0.53, 3.52×10-05), 16p13.11 (B = -0.40, p = 7.40×10-07), 15q11.2 (B = 0.19, p = 1.10×10-16), duplications in 22q11.2 (B = 0.60, p = 7.28×10-10), and deletion (B = 0.52, p = 4.29×10-09) and duplication (B = 0.54, p = 3.69×10-12) in 16p11.2. A similar profile of individual CNV associations was observed for g, though effect sizes tended to be larger. Additional effects were observed for duplication in 1q21.1 (B = 0.40, p = 8.62×10-08), 16p12.1 (B = 0.52, p = 2.87×10-06), 17q12 (B = 0.61, p = 2.81×10-04), and 1q21.1 (B = 0.39, p = 7.08×10-04) as well as duplication (B = 0.52, p = 4,49×10-04), and deletion (B = 0.23, p = 2.35×10-05) within 16p13.11.

Conclusions: CNVs that are established risk factors for neurodevelopmental and psychiatric disorders have moderate effect on both reaction time and general (g) cognitive ability. When considering the overall effect of the presence of any CNV effects are small and interact with polygenic risk for various psychiatric diseases and brain morphology traits such that effects are potentiated

Keywords: CNV, Polygenetic Risk Score, Cognition, IQ

Disclosure: Nothing to disclose.

P535. Polygenic Risk Scores for Psychiatric Disorders in a Diverse Postmortem Brain Tissue Cohort

Laramie Duncan*, Hanyang Shen, Anton Schulmann, Tayden Li, Bhaskar Kolachana, Ajeet Mandal, Ningping Feng, Pavan, K. Auluck, Stefano Marenco

Stanford University, Stanford, California, United States

Background: A new era of human postmortem tissue research has emerged thanks to the development of 'omics technologies that measure genes, proteins, and spatial parameters in unprecedented detail. Also newly possible is the ability to construct individual-level metrics of genetic risk (polygenic risk scores, PRS, based on genome-wide association studies, GWAS). Here we report on clinical, educational, and brain gene expression correlates of polygenic scores in ancestrally diverse samples from the Human Brain Collection Core (HBCC).

Methods: Genotypes from 1,418 donors, derived from cerebellum tissue, were subjected to quality control filters, imputed, and used to construct PRSs. Gene expression from dorsolateral prefrontal cortex (DLPFC), for a subset of 312 samples, was obtained from the CommonMind Consortium.

Results: Polygenic scores for schizophrenia predicted schizophrenia status in donors with African ancestry (p = 1.6×10-5, 10.4% of phenotypic variance explained) and in donors of European ancestry (p = 4.7×10-8, 17.2%). This pattern of higher variance explained among European ancestry samples was also observed for other psychiatric disorders (depression and bipolar disorder) and for height, body mass index, and years of education. Schizophrenia polygenic scores also predicted an aggregate measure of schizophrenia differential gene expression (African ancestry: p = .04, European ancestry: p = .01).

Conclusions: Polygenic scores performed as expected in ancestrally diverse samples, given historical biases toward use of European ancestry samples and variable predictive power of polygenic scores across phenotypes. The transcriptomic results reported here suggest that inherited schizophrenia genetic risk influences gene expression, even in adulthood. For future research, these and additional polygenic scores are available for analyses, and for selecting samples, using postmortem tissue from the Human Brain Collection Core.

Keywords: GWAS, Postmortem Brain Tissue, Schizophrenia (SCZ), Schizophrenia, Bipolar Disorder, Depression, Dorsolateral Prefrontal Cortex (DLPFC)

Disclosure: Nothing to disclose.

P536. Differences in Alternative Splicing and Transcript Isoform Usage in Layer 3 Pyramidal Neurons Across the Visuospatial Working Memory Network

Kevin Dowling*, John Enwright, Dominique Arion, David Lewis

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

Background: Human visuospatial working memory (VSWM) depends on a network of functionally distinct cortical regions, including primary visual (V1) and association cortices in the dorsolateral prefrontal cortex (dlPFC) and posterior parietal cortex (PPC). Across these regions, layer 3 pyramidal neurons (L3PNs) differ in excitability, morphology, and activity during VSWM tasks, partly due to regional differences in gene expression, which are particularly pronounced between the dlPFC or PPC and V1. Alternative splicing of pre-mRNA (AS), a process that alters the composition of exons and retained introns in mature RNA transcripts, is evident in nearly all genes. AS occurs more extensively in the CNS than in other tissues, where it contributes to the regulation of neuron physiology and morphology. However, it is unclear whether AS differs between L3PNs across regions of the VSWM network. Moreover, other processes concomitant with AS further augment transcript isoform diversity (e.g., usage of alternative transcription start sites and alternative polyadenylation sites); the cumulative effects of these processes are indexed by the abundance of distinct transcript isoforms in a given tissue. However, as with AS, it is unclear whether there are regional differences in the proportion of distinct transcript isoforms (a measure known as differential transcript usage; DTU) in L3PNs of the VSWM network.

Methods: Samples of 100 L3PNs were collected from the dlPFC, PPC, and V1 of neurotypical adults ($n = 39$, 11 female) by laser capture microdissection, sequenced to an average depth of 50 million 100-nt paired end reads, and aligned to the genome using STAR. Genome-guided de-novo transcript isoform assembly was performed using StringTie. LeafCutter and IsoformSwitchAnalyzeR were used to analyze regional differences in AS and DTU. All analyses were FDR-corrected for multiple comparisons ($\alpha = 0.05$) and included age, sex, tissue pH, RNA integrity number, PMI, and sequencing batch as covariates. PANTHER was used for pathway enrichment analyses. As many AS events are evolutionarily conserved between humans and macaques, samples of 120 long-range projection L3PNs collected from the dlPFC and PPC of post-pubertal rhesus macaques ($n = 5$, 2 female) were sequenced and used to validate AS differences in human cortex.

Results: A greater number of genes were alternatively spliced between association cortices (dlPFC or PPC) and V1 (877 and 794 genes, respectively) than between the dlPFC and PPC (144 genes); of these genes, 35%-74% were not differentially expressed across regions. Likewise, >79% of genes differentially expressed across regions did not demonstrate significant DTU or AS. Genes exhibiting AS across all three regions were enriched for pathways related to excitatory neurotransmission, synaptic plasticity, and calcium flux. Of AS events that differed between the human dlPFC and PPC, and which occurred at splice sites evolutionarily conserved between the human and the macaque (105 events), 59% of the AS differences between the dlPFC and PPC were replicated in the macaque (62/105 events). As with AS, a greater number of transcripts demonstrated significant DTU between dlPFC (4,109 transcripts) or PPC (3,643 transcripts) and V1 than between dlPFC and PPC (21 transcripts). To predict the functional consequences of DTU, transcript isoform open reading frames were annotated for protein domains, intrinsically disordered regions, coding potential, and sensitivity to nonsense mediated decay (NMD) using PFAM, IUPRED2A, and CPAT3. Transcript isoforms more abundant in V1 were significantly enriched for non-coding potential, intron retention, and sensitivity to NMD – a homeostatic mechanism that degrades mRNA with a premature termination codon prior to translation. Of genes with both expression and NMD sensitivity differences between dlPFC (180 genes) or PPC (223 genes) and V1, NMD sensitivity differences at least partially mediated regional differences in the expression of 49% and 69% of genes, respectively.

Conclusions: Many exons and transcripts demonstrated significant AS or DTU between dlPFC or PPC and V1, a pattern consistent with prior gene expression findings. However, a majority of all genes with significant AS or DTU were not differentially expressed across regions. Likewise, the majority differentially expressed genes did not demonstrate differences in AS or DTU. These results are consistent with prior findings that AS and DTU capture distinct aspects of the transcriptome. Although fewer genes demonstrated significant AS between L3PNs in the dlPFC and PPC than between either region and V1, many AS events that differed across all 3 regions have previously been shown to be functionally significant. For example, splicing of GRIN1 exon 5, which may be altered in schizophrenia and autism spectrum disorder, has been experimentally shown to influence the number of pyramidal neuron spines. GRIN1 exon 5 splicing (but not GRIN1 expression) significantly differed among all three cortical regions, a pattern consistent with spine density on L3PNs being highest in dlPFC, intermediate in PPC and lowest in V1. Lastly, differences in transcript sensitivity to degradation by NMD between dlPFC or PPC and V1 suggest a mechanism by which splicing may regulate regional gene expression differences in L3PNs of the adult VSWM network. Together, the present results suggest AS and DTU may represent important processes contributing to regional differences in L3PN physiology and morphology across the VSWM network.

Keywords: Visuospatial Working Memory, Alternative Splicing, Pyramidal Neuron, Genomics, Transcript Isoforms

Disclosure: Nothing to disclose.

P537. An Investigation of the Prevalence of Copy Number Variants Associated With Neurodevelopmental Disorders in a Multi-Ancestry Brain Biobank

Rebecca Birnbaum*, Joo Heon Shin, Joshua Stoltz, Joel E. Kleinman, Thomas M. Hyde, Leonardo Collado-Torres, Daniel Weinberger

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

Background: Several large, copy number variants (CNVs), deletions or duplications >1 kb, each spanning single or multiple genes, are known to be pathogenic for neurodevelopmental disorders (NDD), rare in frequency, but of relatively large effect ("NDD-CNVs"). NDD-CNVs are notable for pleiotropy, variable expressivity and penetrance, underlying multiple disorders, including autism spectrum disorders, schizophrenia, intellectual disability and seizure disorder, as well as global developmental delay. Underscoring their clinical relevance, testing for NDD-CNVs is being increasingly considered in neuropsychiatry clinics broadly, while diagnostic testing for NDD-CNVs for some childhood-onset disorders is already standard-of-care.

Numerous studies over past decades have elucidated clinical, cognitive and neuroimaging outcomes of NDD-CNV carriers in clinical and population cohorts, and animal and cellular studies have attempted their functional genomic characterization. To date however, investigation of NDD-CNVs in human brain has been

comparatively limited, owing to rare frequency and difficulty accessing human neural tissue, precluding direct neurobiological genomic and transcriptomic characterization of NDD-CNVs. Furthermore, despite the clinical import of NDD-CNVs (discovered in peripheral blood-based analyses), the precise mechanism of central nervous system pathogenicity remains unknown. A rigorous, functional genomic characterization of NDD-CNVs, specifically in human brain, is therefore critical, to directly investigate NDD-CNVs in neurobiological context and to elucidate potential NDD-CNV pathogenic mechanisms. From a translational perspective, such direct query may enable "precise," genetic-based therapeutic interventions. The current analysis leveraged a cross-disorder, multi-ancestry post-mortem brain bank from the Lieber Institute for Brain Development (LIBD) to identify the prevalence of NDD-CNVs.

Methods: Brain tissue collection methods, procurement and dissection by LIBD were previously described. DNA was extracted from post-mortem cerebellum and dura matter from 2,348 brain donors across multiple disorders and across ancestry and genotyped on at least one genotyping array (Human 1 M Duo, Human Hap650Y, Infinum Omni2.5-8, Infinum Omni5-4). PennCNV was applied to genotype data from each brain sample to call CNVs, genome-wide. Subsequent to QC/filtering (exclusions for waviness factor >0.05 or <0.05, B-allele frequency drift>0.01, LogR Ratio standard deviation>0.3, and CNV burden>3 sd of median), CNVs were evaluated for overlap with 121 published NDD-CNVs, a threshold of at least 40% overlap for multigenic NDD-CNVs and overlap of at least one exon for single gene NDD-CNVs.

Results: Initial QC/filtering resulted in the exclusion of 547 samples, yielding 1,801 samples for downstream analyses, multi-ancestry (75% European, 21% African, 2% Hispanic) and cross-disorder, including: 414 neurotypical controls, 522 major depressive disorder, 292 bipolar disorder, 234 schizophrenia, 130 post-traumatic stress disorder, 58 preclinical Alzheimer's, 38 Alzheimer's, 29 autism spectrum disorder, 15 substance use disorder, 14 eating disorders, 12 obsessive compulsive disorder, 10 alcohol use disorder, 6 bipolar disorder NOS, 5 Depression NOS, 4 Attention Deficit Hyperactivity Disorder, 3 Tic Disorder, 1 generalized anxiety disorder and 1 Williams Syndrome. Overall, NDD-CNVs across 16 loci were identified in a total of ~1.8% of samples, and excluding the common 15q11.2 duplication, in 1.3% of brain samples. NDD-CNV prevalence was comparable among major diagnostic groups (1.5% of major depressive disorder, 1.3% schizophrenia, 1.0% of bipolar disorder) but was not enriched compared to neurotypical controls, with 1.4% of neurotypical controls harboring at least one NDD-CNV. Among 29 ASD samples, there was one NDD-CNV carrier, a NRXN1 deletion, and the current analysis confirmed the 7q11.23 deletion in Williams syndrome. Interestingly, there appeared to be ancestry bias in prevalence, as nearly all NDD-CNV carriers, 31 of the 33 identified, were of European ancestry, and only two NDD-CNV carriers of African ancestry. There was no NDD-CNV sex bias, with females found to constitute 1/3 of NDD-CNV carriers proportionate with their representation in the brain biobank.

Conclusions: The current analysis identified NDD-CNV prevalence in a cross-disorder, post-mortem brain bank, indicating a lack of enrichment among major diagnostic groups, confirming past reports of variable penetrance and expressivity. Given the relatively small sample, this may not deviate from expected probability. Future analyses will query NDD-CNVs in the LIBD post-mortem brain cohort using alternative and additional genotype-based calling algorithms for increased sensitivity and specificity, as well as expand sample size of NDD-CNV analyses to additional genotyped post-mortem brain cohorts. The polygenic risk scores of samples will be considered for multiple disorders, as well as background CNV burden in influencing penetrance. Future analyses will investigate transcriptomic and epigenetic effects of a subset of specific NDD-CNVs and also query potential cell-specificity and mosaic characteristics.

Keywords: Copy Number Variant, Human Post-Mortem Brain, Neurodevelopmental Disorders

Disclosure: Nothing to disclose.

P538. Association Between Schizophrenia Polygenic Risk Score, Treatment Resistance, and Symptomatic Remission in Schizophrenia

Max Lam*, Keane Lim, Todd Lencz, Jimmy Lee

Institute of Mental Health, Singapore, Singapore

Background: Treatment-resistant schizophrenia (TRS) occurs in approximately one-third of individuals who fail to show adequate response to antipsychotic medications, with variability in symptoms severity. This study aims to examine the association between the genetic liability of schizophrenia, TRS status, and symptomatic remission status.

Methods: Individuals diagnosed with schizophrenia (N = 876) were categorised as TRS (n = 150) and non-TRS (n = 726), using well-defined TRS criteria. Schizophrenia polygenic risk score (PRS) was calculated, and logistic regression analyses were conducted to discriminate TRS and non-TRS. The TRS status was further stratified based on symptomatic remission criterion, as indexed by the Positive and Negative Syndrome Scale (PANSS), and regression analyses were conducted to examine the association of PRS with the following symptomatic remission subtypes – antipsychotic remitter; antipsychotic non-remitter; clozapine remitter; clozapine non-remitter.

Results: Schizophrenia PRS discriminated TRS and non-TRS ($R2 = 3.04\%$, $p = 2.27 \times 10\text{-}5$, $0\ R = 1.55$), with higher PRS observed in the TRS group, compared with non-TRS. Stratification of TRS status based on symptomatic remission criterion revealed significant difference in PRS across subtypes ($R2 = 1.62\%$, $p = 2.68 \times 10\text{-}4$, $0\ R = 1.27$). Within TRS, higher schizophrenia PRS was observed in clozapine remitter subtype; while in non-TRS, higher schizophrenia PRS was observed in antipsychotic non-remitter subtype. Compared to the lowest decile, the odds ratio of being classified as TRS in the top decile is 3.29 (95% CI: 1.21-8.94).

Conclusions: The common risk variants for schizophrenia are associated with TRS status, and symptomatic remission. Findings suggest that the genetic burden of schizophrenia may in part explain treatment resistance status in schizophrenia, and indexes treatment outcomes.

Keywords: Schizophrenia (SCZ), Treatment-Resistance, Polygenic Risk Scores, Antipsychotic Treatment, Remission

Disclosure: Nothing to disclose.

P539. Excess Homozygosity in Neuropsychiatric Patients From the Ashkenazi Jewish Populations Implicates TUBGCP4 as a Recessive Locus for Schizophrenia

Todd Lencz*, Jin Yu, Max Lam, Anil Malhotra, Itsik Pe'er

Zucker Hillside Hospital, Glen Oaks, New York, United States

Background: To date, rare variant research in schizophrenia (SZ) has focused on dominant exonic variants; there has been limited research examining the recessive mode of inheritance. The Ashkenazi Jewish (AJ) population is marked by an excess of homozygosity, due to a history of endogamy and a small founder population and may provide enhanced power to detect recessive loci.

Methods: We examined homozygous loci in exome data derived from high-depth (>30x) whole genome sequencing performed on 786 schizophrenia cases and 463 controls drawn

from the AJ population. To enhance power, we merged our dataset with 1934 AJ neuropsychiatric cases from gnomAD and 3106 AJ controls from gnomAD (non-neuro). We compared cases and controls for recessive effects across three types of alleles: synonymous, loss-of-function (LoF), and damaging missense plus loss-of-function (MisLoF). Analagous to our prior work in rare variant discovery (Lencz et al. 2021, Neuron), we hypothesized that significant enrichment would occur if variants observed homozygous even once in gnomAD (non-neuro) and TOPMED databases (total N > 158 K) were filtered out. We compared cases and controls for recessive effects across three allele frequency bins (0.1%<MAF < 1%; MAF < 0.1%; and MAF < 0.1% plus never observed as homozygous in healthy gnomAD and TOPMed samples), and across three types of alleles: synonymous, loss-of-function, and damaging (MPC > 2) missense plus loss-of-function.

Results: Cases were enriched for ultra-rare recessive alleles in the LoF (OR~2) and LoF+Mis (OR~4) categories. Recessive synonymous variants were not enriched in cases relative to controls. While most of the homozygous LoF+Mis variants were observed in poorly annotated genes, a homozygous LoF splice-donor variant (chr15:43696752:T/C) in TUBGCP4 was observed in one patient with schizophrenia. Recessive mutations in TUBGCP4 have been associated with intellectual disability with microcephaly (Scheidecker et al., 2015, Am J Hum Gen).

Conclusions: Results suggest that a small, but detectable, fraction of risk for schizophrenia derives from recessive effects of ultra-rare variants. Unique founder populations, such as AJ, are enriched for such variants and may be worthy of further study. Recessive mutations in TUBGCP4 have been previously associated with intellectual disability with microcephaly, consistent with research showing overlap in dominant rare variants between SZ and neurodevelopmental disorders. This gene encodes a protein that is part of the gamma-tubulin ring complex, which is required for microtubule nucleation, an essential step in the process of neuronal morphogenesis.

Keywords: Human Genetics, Schizophrenia (SCZ), Neurodevelopment

Disclosure: Nothing to disclose.

P540. Investigating Trait Variability of Gene Co-Expression Network Architecture in Brain by Controlling for Genomic Risk of Schizophrenia

Eugenia Radulescu*, Qiang Chen, Giulio Pergola, Pasquale Di Carlo, Joo Heon Shin, Thomas Hyde, Joel Kleinman, Daniel Weinberger

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

Background: Schizophrenia (SCZ) genetic risk is linked with variation in gene expression in brain, but how this affects gene co-expression networks is unclear. To better understand this issue, we implemented a novel approach to network analysis by adjusting gene expression input to account for the effects of SCZ genomic risk score (GRS).

Methods: Postmortem brain samples, RNA-Seq preparation and calculation of variables to enter the selection for gene expression adjustment: data was acquired from assays of postmortem human brain tissue from the LIBD Human Brain Repository, collected under a standard protocol of brain acquisition, processing, and curation. After pre-processing, a final gene expression (N = 18,980 genes) dataset of 78 DLPFC samples (age of death: 43 ± 15.8; M/F: 64/14) from neurotypical adults of European ancestry was retained for further analysis. The gene expression was further adjusted by linear regression models to remove variance explained by RNA quality, cell type proportion, age, sex and genetic ancestry. WGCNA was then used to create DLPFC brain co-expression networks after preserving or removing GRS effects and to calculate consensus networks. The consensus (i.e., background) networks- representative of gene co-expression architecture free of genomic scores effects- were used as baseline for differential network analysis. Specifically, the differential network analysis consisted of calculating the overlap between modules of GRS preserved networks and background modules. Differential modules were considered preserved modules with weak or no overlap in background; they were subsequently tested for biological significance, i.e., correlations with GRS, enrichment in gene ontology (GO) biological processes and enrichment in PGC3 SCZ GWAS priority loci genes. From the same dataset, we created also analogous co-expression networks based on the genomic score of height, as a control normative trait.

Results: We identify key aspects of SCZ GRS effects on brain co-expression networks: 1) Co-expression networks derived from expression input adjusted to account for the genomic scores of SCZ and height traits share a similar architecture (i.e., module composition) with subtle differences between preserved and removed GRS effects.

2) For each trait- SCZ risk and height, modules of networks that preserve GRS effects have a pattern of overlap with the corresponding background modules characterized by strong correspondence for half of the modules and weaker correspondence for the other half.

3) Several SCZ risk and height GRS preserved modules with weak correspondence in the background modules had module eigengenes (MEs) correlated with GRS SCZ or GRS height. Preserved modules with MEs negatively correlated with GRS SCZ were enriched for brain specific functional ontologies and priority PGC3 SCZ GWAS loci genes. Preserved modules with MEs correlated with GRS height were mainly enriched for general cellular processes of transcription, translation and metabolism.

Conclusions: Overall, our results indicate that SCZ GRS is associated with brain gene co-expression networks and creates a molecular background for gene-gene interactions that affect diverse biological pathways. Furthermore, consistent with the pleiotropy phenomenon, SCZ risk genetic influence on gene co-expression architecture shares a part of signal with other complex traits (i.e., height), but has also specific contributions convergent toward brain functionality.

Keywords: Schizophrenia (SCZ), Dorsolateral Prefrontal Cortex, Co-Expression Network Analysis

Disclosure: Nothing to disclose.